Nonpeptide Angiotensin II Receptor Antagonists. I. Synthesis and Biological Activity of Pyridine Derivatives

Naoto UEYAMA, Takashi YANAGISAWA, Tomoyuki KAWAI, Motoharu SONEGAWA, Hiromi BABA, Seiichiro MOCHIZUKI, Kazuhiro KOSAKAI, and Tsuyoshi TOMIYAMA*

Kotobuki Research Laboratories, Kotobuki Pharmaceutical Company, Ltd., 6351 Sakaki-Machi, Nagano 389-06, Japan. Received March 2, 1994; accepted May 18, 1994

Substituted pyridines were synthesized as potential angiotensin II (AII) receptor antagonists. Substitution at the position 2 in the pyridine resulted in potent activity, and the optimal alkyl length was four carbons. The potency further increased with the introduction of a hydroxymethyl group at the position 4. One of the compounds, 2-butyl-6-chloro-4-hydroxymethyl-5-methyl-3-[[2′-(1*H*-tetrazol-5-yl)biphenyl-4-yl]methyl]pyridine 9h (KT3-579) is a competitive AII antagonist with a pA_2 value of 9.31, and is about 10 times more potent than Du Pont 753. It was found to be an AT_1 specific antagonist with an IC_{50} of 3.09 nM.

Keywords nonpeptide AII antagonist; pyridine derivative; competitive AII antagonist; KT 3-579

The renin-angiotensin system (RAS) is one of the important homeostatic mechanisms that regulate hemodynamics and water and electrolyte balance.[1] One approach to controlling the RAS system is to use angiotensin converting enzyme (ACE) inhibitors which inhibit the conversion of angiotensin I (AI) to angiotensin II (AII). However, AII can also be formed *in vivo* by the action of enzymes other than ACE.[2] A more effective approach is to block the action of AII at the AII receptor level. The disclosure of Du Pont 753,[3] the first example of a specific AII receptor antagonist, provided a lead for the development of more potent inhibitors and stimulated a search for agents with improved potency and longer duration of action. Recently more potent nonpeptide AII receptor antagonists such as L-158809[4] and SR-47436[5] have been described.

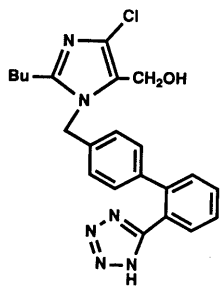
Structure comparison of these compounds reveals two fundamental units : a heterocyclic head and a biphenyl tail having an acidic moiety such as a tetrazole. As a heterocyclic head, 5-membered imidazole (Du Pont 753, SR-47436) and fused imidazole (L-158809) have been reported, but the 6-membered pyridine head has not been reported yet, so we decided to prepare 6-membered pyridine head analogues. Furthermore, we assumed that the 3-nitrogen in imidazole and pyridine nitrogen were possible hydrogen bond acceptors.

In this paper, we report our initial efforts to synthesize and to examine the biological activity of the pyridine derivatives.

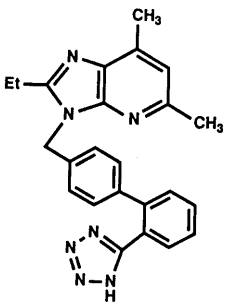
Chemistry

The synthesis of the target compounds was started from the substituted pyridine-3-carbaldehyde **1** and Chart 1 outlines the general method for the synthesis of these compounds. Coupling of **1** with Grignard reagents of 4-bromo-2′-oxazolylbiphenyl **2** or 4-bromo-2′-trityltetrazolylbiphenyl **3** gave the carbinol **4**.[6,7] Chlorination of **4** with thionyl chloride gave **5**, which was reduced with ZnAcOH to give **6** and **7**. Subsequent hydrolysis in 4.5 N HCl or deprotection with 75% AcOH gave **8** and **9**. The biphenyls **2** and **3**[8] were prepared according to the Meyers' method as shown in Chart 2, using an oxazoline to mask the carboxylic group[9] and the nucleophilic displacement of an O-methoxy group of **11** with Grignard reagent. The oxazoline **11**, prepared from **10**, was coupled with *p*-bromophenylmagnesium bromide ($MgBrC_6H_5Br$) **12** to give the biphenyl **2**, which was hydrolyzed to the carboxylic acid **13**, then converted to the amide and dehydrated to the nitrile **14**. Alternatively, **2** was also directly converted to **14** with phosphorus oxychloride in pyridine.[10] Treatment of **14** with trimethyltinazide afforded the trimethyltintetrazole **15**,[11] which was converted to the trityl tetrazole **3** by the standard procedure.

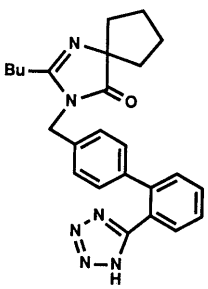
Preparation of mono-, di- and trisubstituted pyridine-3-carbaldehydes **1a**—**4a** is outlined in Chart 3; four principal routes A—D were utilized to prepare these compounds. 2-Mono-, 2,4-di- and 2,4,6-trisubstituted pyridine-3-carbaldehydes **1a** were obtained by condensa-



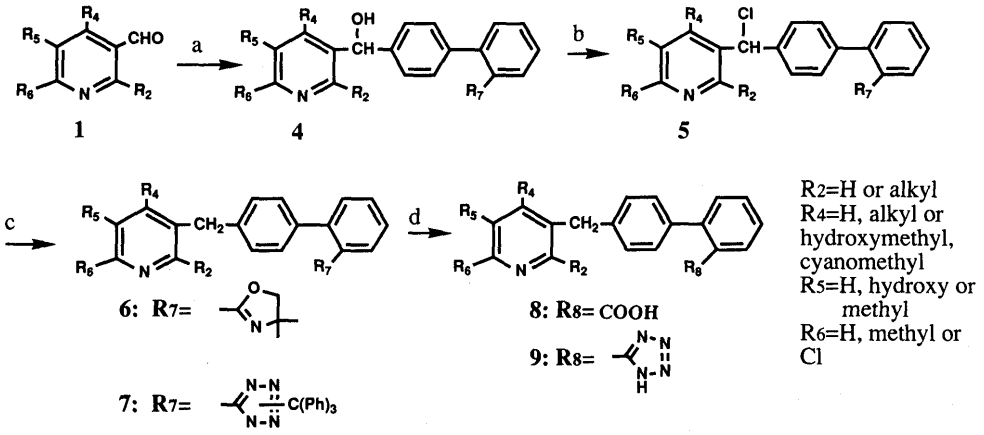
Du Pont 753



L-158809

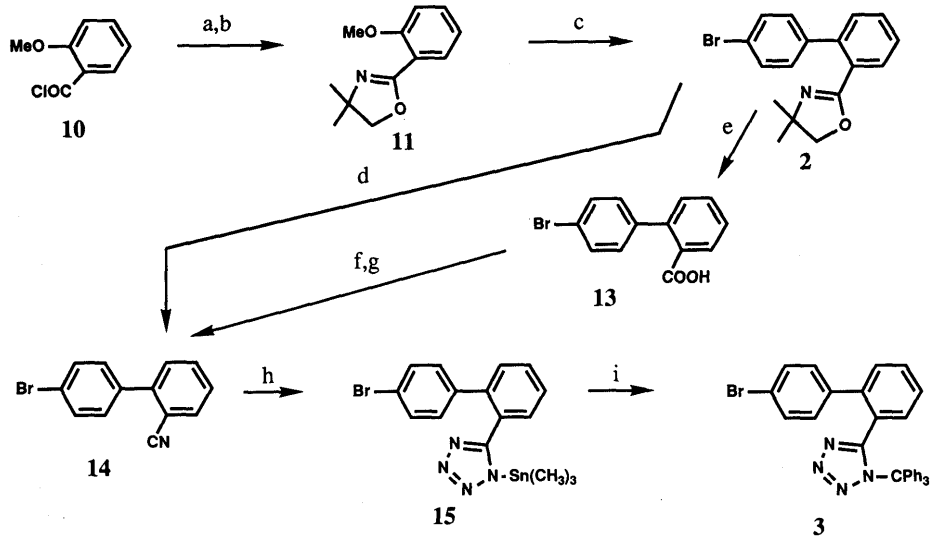


SR-47436



a, Mg/ BrC6H4-C6H4 (**2**) or BrC6H4-C6H4CN4- trityl (**3**)/THF;
b, $SOCl_2$, 60°C ; c, Zn/AcOH, r.t.; d, 4.5 N HCl, reflux (R7=) or 75% HOAc, r.t. (R7=)

Chart 1

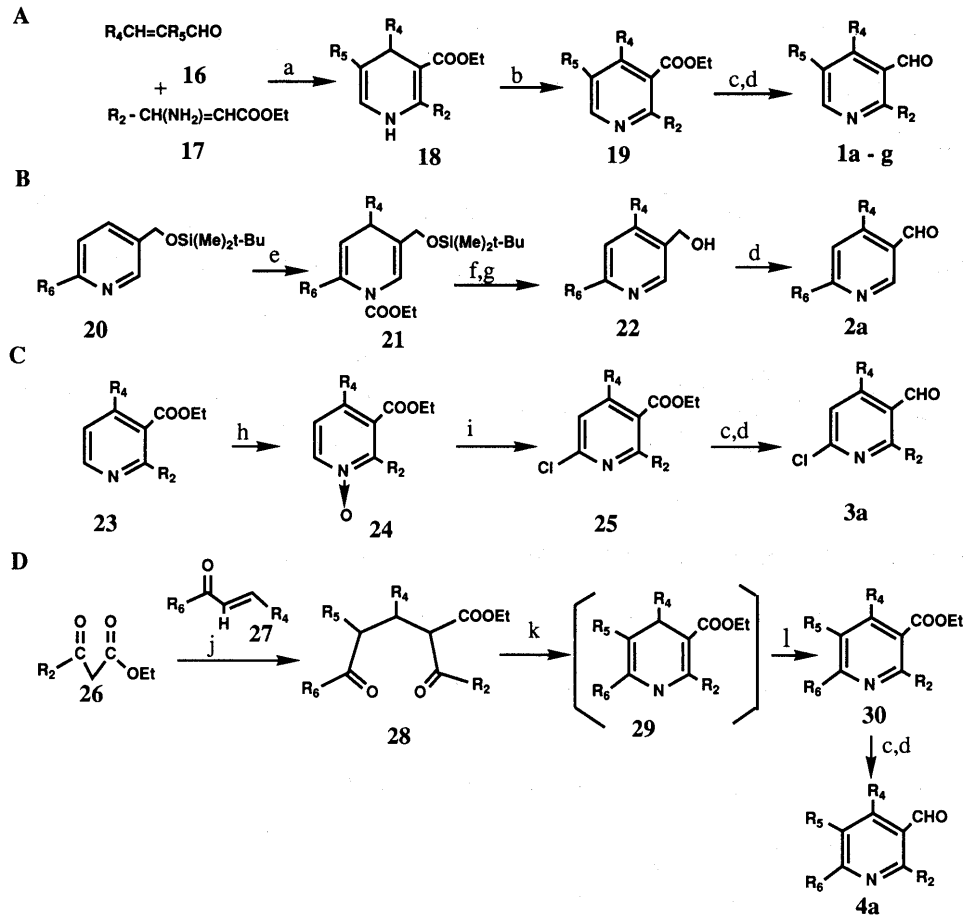


a, $NH_2C(CH_3)CH_2CH_2OH$, CH_2Cl_2,r.t.; b, $SOCl_2$, reflux; c, BrC_6H_5MgBr (**12**) / THF, reflux; d, $POCl_3$, pyridine, reflux; e, 4.5 N HCl, reflux; f, $SOCl_2$, reflux then NH_4OH, r.t.; g, $POCl_3$, 130°C; h, Me_3SnN_3 / toluene, reflux; i, NaOH-H_2O then Ph_3CCl / Et_3N

Chart 2

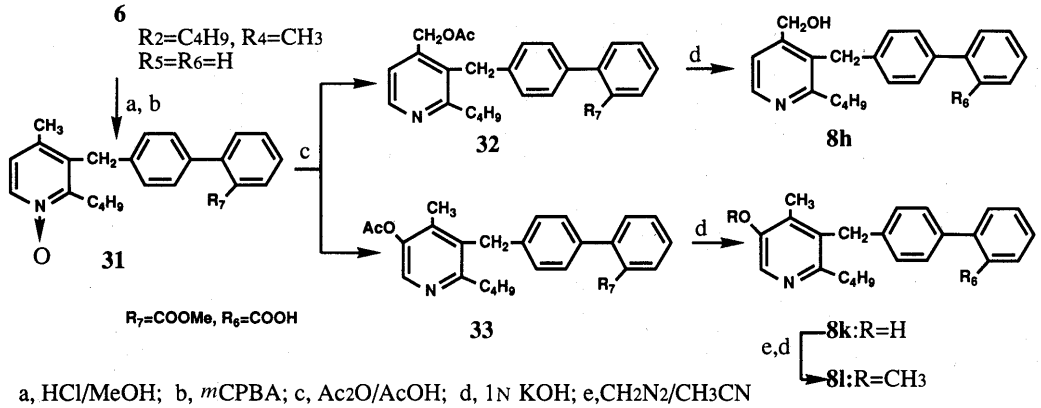
tion of the appropriate acrylaldehyde **16** and ethyl 2-aminoacrylate **17** to give the 1,4-dihydropyridine **18**,[12] followed by oxidation with ceric ammonium nitrate to give the corresponding ethylpyridine-3-carboxylate **19**,[13] which was reduced to the alcohol with $LiAlH_4$, and subsequently oxidized to the aldehyde **1a—g** (route A). 4,6-Disubstituted pyridine-3-carbaldehyde **2a** was prepared by regioselective 1,4 addition of the 6-substituted pyridine-3-methanol silylether **20** *via* pyridinium chloride as outlined in route B.[14] The resulting dihydropyridine **21** was aromatized with O_2, and desilylated to give the alcohol **22**, which was oxidized with MnO_2 to give the aldehyde **2a**.[15] 2,4-Disubstituted-6-chloropyridine-3-carbaldehyde **3a** was prepared as shown in route C.[16] The N-oxide **24**, prepared from **23** with *m*-chloroperbenzoic acid (*m*CPBA), was treated with phosphorus oxychloride ($POCl_3$) to give **25** and then converted to **3a** using the standard procedure. 2,4,6-Trisubstituted pyridine-3-carbaldehyde **4a** was prepared as shown in route D.[17]

Sodium ethoxide-catalyzed Michael addition of the ketoester **26** to **27** gave the desired adduct **28**. Treatment of **28** with ammonium acetate (NH_4OAc) in hot acetic acid (HOAc) afforded the intermediate dihydropyridine **29**, which was oxidized with cupric acetate ($Cu(OAc)_2$) *in situ* to afford the pyridyl ester **30**. The compounds having



a, EtOH, reflux; b, $(NH_4)_2Ce(NO_3)_6$; c, LAH/THF; d, MnO_2/Me_2CO; e, $ClCO_2Et$ then R_1MgBr/CuI; f, O_2; g, F^-; h, *m*CPBA; i, $POCl_3$; j, EtONa/EtOH; k, $NH_4OAc/HOAc$; l, $Cu(OAc)_2/AcOH$

Chart 3



a, HCl/MeOH; b, *m*CPBA; c, $Ac_2O/AcOH$; d, 1N KOH; e, CH_2N_2/CH_3CN

Chart 4

hydroxymethyl group at **8h** and a hydroxy group at **8k** were prepared by means of the Takada reaction as shown in Chart 4.[18] The N-oxide **31**, prepared from **6** ($R_2=C_4H_9$, $R_4=CH_3$ and $R_5=R_6=H$) with *m*CPBA, was treated with HOAc–Ac_2O to obtain a mixture of **32** and **33**, which were chromatographically separated and these compounds were deacetylated to give **8h** and **8k**. Compound **8k** was further methylated to give **8l**. Compound **9e**—**h** and **9j** were obtained in the same manner. In the case of **9e**, the hydroxymethyl group was cyanated to give the cyanomethyl compound **9i**. The 4-hydroxymethyl-6-chloro compound was prepared *via tandem* oxidation of the chloro compound. The synthetic procedure for **9h** (KT 3-579) is illustrated in Chart 5.

Results and Discussion

Compounds **8a**—**l** and **9a**—**j** were tested for AII receptor antagonistic activity in isolated rabbit aorta. Physico-

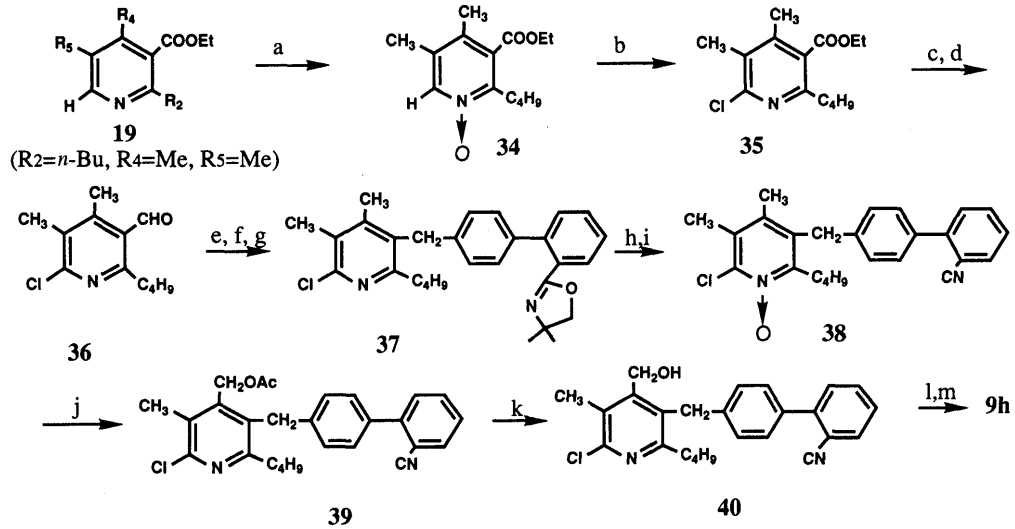
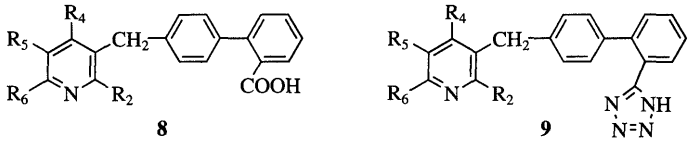


Chart 5

TABLE I. Physical Properties and AII Receptor Antagonistic Activities of Compounds **8** and **9**



No.	R_2	R_4	R_5	R_6	mp (°C)	Formula	Analysis (%) Calcd C	Calcd H	Calcd N	Found C	Found H	Found N	pA_2
8a	CH_3	H	H	H	185—187	$C_{20}H_{17}NO_2$	79.19	5.65	4.62	78.92	5.53	4.81	5.25
8b	CH_3	CH_3	H	H	264—265	$C_{21}H_{19}NO_2$	79.47	6.03	4.41	79.19	6.00	4.79	4.20
8c	CH_3	C_2H_5	H	H	247—249	$C_{22}H_{21}NO_2$	79.73	6.39	4.23	79.39	4.59	6.63	5.90
8d	CH_3	C_3H_7	H	H	193—195	$C_{23}H_{23}NO_2$	79.97	6.71	4.05	79.61	6.96	4.31	6.05
8e	C_3H_7	CH_3	H	H	176—178	$C_{23}H_{23}NO_2$	79.97	6.71	4.05	80.08	6.85	4.11	6.85
8f	C_4H_9	CH_3	H	H	171—173	$C_{24}H_{25}NO_2$	80.19	7.01	3.90	79.85	7.21	4.05	7.03
8g	C_5H_{11}	CH_3	H	H	187—189	$C_{25}H_{27}NO_2$	80.40	7.29	3.75	80.07	7.60	3.93	6.52
8h	C_4H_9	CH_2OH	H	H	117—118	$C_{24}H_{25}NO_3$	76.77	6.71	3.73	76.67	6.93	3.77	7.43
8i	H	C_3H_7	H	CH_3	109—112	$C_{23}H_{23}NO_2$	79.61	6.96	4.31	79.97	6.71	4.05	5.69
8j	C_4H_9	CH_3	H	Cl	120—122	$C_{24}H_{24}ClNO_3$	70.32	5.90	3.42	70.08	6.05	3.72	7.30
8k	C_4H_9	CH_3	OH	H	242—243	$C_{24}H_{25}NO_3$	76.77	6.71	3.73	76.78	6.84	4.02	6.63
8l	C_4H_9	CH_3	OCH_3	H	81—83	$C_{25}H_{27}NO_3$	77.09	6.99	3.60	76.83	7.12	3.92	7.27
9a	C_4H_9	CH_3	H	H	189—190	$C_{24}H_{25}N_5$	75.17	6.57	18.26	75.35	6.79	18.35	7.75
9b	C_4H_9	CH_3	H	Cl	112—113	$C_{24}H_{24}ClN_5$	68.97	5.79	16.76	68.85	5.72	16.95	8.15
9c	C_4H_9	CH_3	CH_3	H	193—194	$C_{24}H_{27}N_5$	75.54	6.85	17.62	75.30	6.70	17.61	7.92
9d	C_4H_9	CH_3	CH_3	Cl	87—89	$C_{25}H_{26}ClN_5$	69.51	6.07	16.21	69.40	6.06	14.49	8.03
9e	C_4H_9	CH_2OH	H	H	194—196	$C_{24}H_{25}N_5O$	72.16	6.31	17.53	71.95	6.55	17.26	8.87
9f	C_4H_9	CH_2OH	H	Cl	114—116	$C_{24}H_{24}ClN_5O \cdot H_2O$	63.78	5.75	15.49	64.08	5.80	15.62	8.68
9g	C_4H_9	CH_2OH	CH_3	H	182—184	$C_{25}H_{27}N_5O$	72.61	6.58	16.94	72.32	6.70	17.12	8.70
9h	C_4H_9	CH_2OH	CH_3	Cl	119—120	$C_{25}H_{26}ClN_5O$	67.03	5.85	15.63	66.91	5.91	15.85	9.31
9i	C_4H_9	CH_2CN	H	H	152—153	$C_{25}H_{24}N_6$	73.51	5.92	20.57	73.24	5.93	20.37	8.15
9j	C_4H_9	CH_2OH	H	CH_3	154—155	$C_{25}H_{27}N_5O$	72.61	6.58	16.94	72.32	6.70	17.12	8.35
Dup753													8.32

chemical properties and pA_2 values are shown in Tables I and II. In the carboxylic acid analogues **8a**—**l**, elongation of the alkyl group at R_4 from H (**8a**) to C_3H_7 (**8d**) did not cause a significant increase in the activity but elongation of the alkyl group at R_2 from C_3H_7 (**8e**) to C_5H_{11} (**8g**) did affect on AII receptor activity. The optimal alkyl length at the 2 position was C_4H_9 (**8f**), with a pA_2 value of 7.03. Introduction of a hydroxymethyl group in-

TABLE II

	(IC_{50}, nM)	
	AT_1 [a]	AT_2 [b]
a. Characterization of **9h** in radioligand binding assay		
9h	0.8	>10000
Dup 753	5.0	>10000
b. Selectivity of **9h** to the other agonists [c]		
KCl	10^{-6}	N.E.
Norepinephrine	10^{-6}	N.E.
5-Hydroxytryptamine	10^{-6}	N.E.

a) Calculated from inhibition of the binding of [^{125}I]Sar^1, Ile^8-AII using rat liver AT_1 receptor membrane. *b*) Calculated from inhibition of the binding of [^{125}I]AII. *c*) Effect on the contraction in rabbit aorta caused by the agonists.

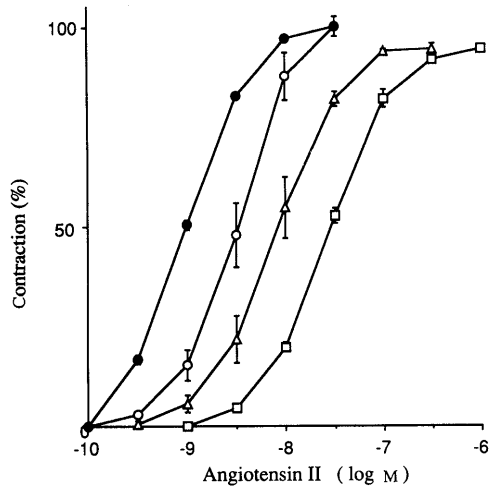


Fig. 1. The Effect of **9h** (KT3-579) on the Concentration–Response Curve for AII in Isolated Rabbit Aorta. Symbols Represent Mean Values ± S.E. ($n=6$)

$pA_2 = 9.31 \pm 0.17$, slope $= 1.06 \pm 0.11$. —●—, Vehicle; —○—, KT3-579 10^{-9} M; —△—, KT3-579 3×10^{-9} M; —□—, KT3-579 10 M.

stead of methyl at the 4 position increased AII receptor antagonistic activity from pA_2 7.03 to 7.43 (**8f** *versus* **8h**).

Introduction of a hydroxy group at the 5 position decreased the activity (**8f** *versus* **8k**) and the methoxy compound **8l** had nearly the same activity as **8i**. In the tetrazole analogues **9a—j**, replacement of carboxylic acid **8f** with tetrazole **9a** increased the potency from pA_2 7.03 to 7.75. Conversion of the methyl group to a hydroxymethyl group at the 4 position increased the activity from pA_2 7.75 to 8.87 (**9a** *versus* **9e**). Chlorination at the 6 position (**9a**, **9c** *versus* **9b**, **9d**) also increased the pA_2 value by 0.11—0.40. However, no increase in AII receptor antagonistic activity was elicited by chlorination of the 4-hydroxy analogue (**9e** *versus* **9f**). In contrast, the introduction of a chlorine atom at the 6 position of the 5-methyl compound increased the pA_2 value by about 0.6 (**9g** *versus* **9h**). These studies suggest that, in the AII receptor, there is a limited lipophilic binding site to which the 6-chloro and 5-methyl groups of **9h** bind, since a chlorine atom and a methyl group are reported to increase the lipophilicity of **9h**.[19] Conversion of 4-hydroxymethyl **9e** to cyanomethyl **9i** lowered the activity. The inhibitory pattern and the potency of **9h** *in vitro* are shown in Fig. 1, using contractile responses to AII in isolated rabbit aorta; **9h** showed competitive antagonism in this model.[20] The concentration–response curve for AII was shifted in parallel to the right and there was no depression of the maximal contractile response in the presence of 1, 3 or 10 nM **9h**. For further characterization, **9h** was tested in a radioligand binding assay for AT_1 receptor using rat liver membrane preparations. It inhibited the binding of [^{125}I]Sar^1,Ile^8-angiotensin to AT_1 receptor with IC_{50} value of 3.09 nM and no AT_2 antagonist activity of **9h** was found at concentrations up to 10^{-5} M in radioligand binding assay with bovine cerebellum (Table II, section a). As shown in Table II, section b, **9h** was selective, having no effect on KCl, norepinephrine or serotonin actions up to 10^{-6} M.

In conclusion, we have developed a new class of potent non peptide AII receptor antagonists with high selectivity for AT_1. Introduction of a methyl group and a chlorine atom at positions 5 and 6 in 2-butyl 4-hydroxymethyl pyridine derivatives increased the AII receptor antagonistic activity and **9h** (KT3-579) was about 10 times more potent than Du Pont 753 in terms of pA_2 value. *In vivo* pharmacological studies of KT3-579 are planned and will be reported elsewhere.

Experimental

Melting points were determined on a Yanagimoto melting point apparatus and are uncorrected. Infrared (IR) spectra were recorded on a Hitachi 270-30. Proton nuclear magnetic resonance (^{1}H-NMR) spectra were measured at 90 MHz on a Hitachi R-90H Fourier-transform NMR spectrometer. Chemical shifts are quoted in parts per million (ppm) with tetramethylsilane as an internal standard. Coupling constants (J) are given in Hz. The following abbreviations are used: s, singlet; d, doublet; t, triplet; q, quartet; brs, broad singlet; dd, doublet of doublet; m, multiplet. Mass spectra (MS) were taken on a Hitachi M-80B spectrometer. Elemental analyses were within ±0.4% of theoretical values and were determined by a Hitachi 026 CHN analyzer. For column chromatography, silica gel (Merck, Kieselgel 60, 70—230 mesh) was used.

2-Butyl-6-chloro-4-methyl-3-[[2′-(triphenylmethyltetrazol-5-yl)biphenyl-4-yl]hydroxymethyl]pyridine (4) (R_2 = Butyl, R_4 = Methyl, R_5 = H and R_6 = Chlorine Grignard reagent (prepared from Mg (5.6 mmol) and 5-(4′-bromobiphenyl-2-yl)-*N*-(triphenylmethyl)tetrazole **3**, 3.6 mmol) was added to a solution of **1** (2.0 g, 1.8 mmol) (R_2 = butyl, R_4 = methyl, R_5 = H and R_6 = chlorine) in dry tetrahydrofuran (THF) (20 ml) at −78 °C under the Ar atmosphere, and the mixture was stirred at −78 °C for 1.5 h. The reaction mixture was poured into a saturated aqueous ammonium chloride (NH_4Cl) solution and extracted with ethyl acetate (AcOEt). The extract was washed with brine, dried (Na_2SO_4) and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (1 : 3)] to give 0.82 g (63.8%) of **4** as a colorless oil. ^{1}H-NMR ($CDCl_3$) δ: 0.85 (3H, t, J = 6.2 Hz), 1.05—1.80 (4H, m), 2.08 (3H, s), 2.33 (2H, m), 3.97 (2H, s), 6.78—7.90 (9H, m). MS (m/z): 620 ($M^+ - 56$).

2-Butyl-6-chloro-4-methyl-3-[[2′-(triphenylmethyltetrazol-5-yl)biphenyl-4-yl]chloromethyl]pyridine (5) (R_2 = Butyl, R_4 = Methyl, R_5 = H and R_6 = Chlorine) A mixture of **4** (2.8 g, 4.0 mmol) and $SOCl_2$ (10 ml) was stirred at 60 °C for 0.5 h. The excess thionyl chloride ($SOCl_2$) was removed under vacuum and the residue was concentrated twice from toluene. The resulting residue was dissolved in H_2O and the solution was basified with $NaHCO_3$ and extracted with AcOEt. The extracts were dried (Na_2SO_4), filtered and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (1 : 3)] to give 2.6 g (85.0%) of **5**. ^{1}H-NMR ($CDCl_3$) δ: 0.89 (3H, t, J = 6.2 Hz), 1.12—1.85 (4H, m), 2.03 (3H, s), 2.80 (2H, m), 6.41 (1H, s), 6.80—8.10 (24H, m). MS (m/z): 637 ($M - 56$).

2-Butyl-6-chloro-4-methyl-3-[[2′-(1*H*-tetrazol-5-yl)biphenyl-4-yl]-methyl]pyridine (9) (R_2 = Butyl, R_4 = Methyl, R_5 = H and R_6 = Chlorine) Zinc dust (1.8 g) was added to a solution of **5** in AcOH (15 ml). After 5 min, the same amount of zinc dust was added to the suspension

and the mixture was stirred for 0.5 h. The mixture was filtered and the filtrate was evaporated *in vacuo*. A saturated aqueous $NaHCO_3$ solution was added to the residue and the mixture was extracted with Et_2O. The extract was washed with brine, dried (Na_2SO_4), filtered, and evaporated *in vacuo* to give 2.2 g (100%) of crude **7**. A solution of **7** (2.2 g, 3.4 mmol) in 75% AcOH (50 ml) and THF (100 ml) was stirred at room temperature for 4 h. An excess 10% NaOH was added and the solvents were removed *in vacuo*. The resulting residue was dissolved in water and the mixture was filtered to removed the triphenylmethanol. The filtrate was adjusted to pH 4.5 using concentrated HCl. The precipitate was recovered by filtration and dried and the solvent was evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [$CHCl_3$] to give 1.1 g (75%) of **9b**. mp 112—113 °C. ^{1}H-NMR ($CDCl_3$) δ: 0.85 (3H, t, J=6.2 Hz), 1.05—1.80 (4H, m), 2.08 (3H, s), 2.33 (2H, m), 3.97 (2H, s), 6.78—7.90 (9H, m). MS (*m/z*): 417 (M^+). IR (KBr): 3004, 2860, 2672, 1581 cm^{-1}.

4,4-Dimethyl-2-(4-methoxyphenyl)-2-oxazoline (11) The title compound was obtained by the reaction of **10** and 2-amino-2-methyl-1-propanol according to the method of Meyers *et al.*[8]

2-(4′-Methylbiphenyl-2-yl)-4,4-dimethyloxazoline (2) 1,4-Dibromobenzene (23.5 g, 106 mmol) was added to dropwise to a suspension of magnesium metal (2.5 g, 103 mmol) in anhydrous THF (200 ml) at 20 °C. Following the addition the mixture was stirred at 20 °C until the magnesium had entirely dissolved (1 h). The resulting reagent was added to a solution of **11** (10.0 g, 48.7 mmol) in anhydrous THF (100 ml) at 20 °C, and the reaction mixture was stirred at 20 °C for 2 h. It was then poured into saturated aqueous NH_4Cl solution, and extracted with AcOEt. The combined organic phases were dried (Na_2SO_4), then filtered, and the solvent was evaporated off *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (1 : 1)] to give 9.6 g (60.0%) of **2** as an oil. ^{1}H-NMR ($CDCl_3$) δ: 1.26 (6H, s), 3.76 (2H, s), 7.12—7.80 (8H, m). MS (*m/z*): 330 (M^+). IR (neat): 2956, 1656, 1473 cm^{-1}.

4′-Bromobiphenyl-2-carboxylic Acid (13) Compound **2** (23 g, 0.07 mol) was taken up in 50 ml of 4.5 N HCl. After 12 h at reflux, the reaction mixture was cooled and extracted with Et_2O. The organic solution was dried (Na_2SO_4), and evaporated to give 15 g (78.7%) of **13** as a colorless solid. MS (*m/z*): 277 (M^+).

4′-Bromo-2-cyanobiphenyl (14) (Route A): 4′-Bromobiphenyl-2-carboxylic acid (**13**) (2.7 g, 0.01 mmol) was added to 11.9 g of $SOCl_2$ and the mixture was refluxed for 2 h. The excess $SOCl_2$ was removed by distillation and the residue was concentrated to yield 2.9 g (100%) of the acid chloride. This was added dropwise manner to magnetically stirred ammonia solution (50 ml) at 0 °C. The mixture was stirred at 0 °C for 15 min, then H_2O (100 ml) was added. The slurry was filtered and washed with water to give 2.3 g of the crude amide. The amide (2.3 g) was dissolved in $SOCl_2$ (3.0 ml), and the resulting solution was stirred at 25 °C for 1 h, then evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (3 : 1)] to give 2.0 g (90%) of **14** as a colorless liquid. MS (*m/z*): 258 (M^+).

(Route B): A solution of **2** (33.0 g, 0.1 mmol) in 100 ml of pyridine at 10 °C was treated dropwise with $POCl_3$ (172 ml, 0.2 mmol) such that the reaction temperature did not exceed 15 °C. The reaction mixture was stirred at 100 °C for 3 h, then cooled to room temperature and the emulsion was extracted with AcOEt. The combined organic phases were washed with water and brine, dried (Na_2SO_4), filtered and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (3 : 1)] to give 23.6 g (92%) of **14** as a colorless liquid. MS (*m/z*): 258 (M^+).

5-(4′-Bromobiphenyl-2-yl)-*N*-(triphenylmethyl)tetrazole (3) A solution of **14** (3.3 g, 10 mmol) and trimethyltin azide (8.2 g, 40 mmol) in 35 ml of toluene was refluxed for 72 h, then cooled to 20 °C, and 10 N aqueous NaOH solution (2.9 g, 10.5 mmol) and triphenylmethyl chloride were added at 20 °C. The resulting mixture was stirred at 20 °C for 3 h. Then, 100 ml of water was added and the whole was extracted with benzene (50 ml). The combined organic phases were dried (Na_2SO_4), filtered and evaporated *in vacuo*. Recrystallization of the residue from Et_2O gave **3** (4.3 g, 80.0%) as a colorless solid. mp 132—134 °C. ^{1}H-NMR ($CDCl_3$) δ: 6.70—8.10 (23H, m). MS (*m/z*): 488 ($M^+ - 56$).

General Method A for the Synthesis of 2,4,5-Trisubstituted 3-Pyridinecarbaldehyde. 2-Methyl-3-pyridinecarbaldehyde (1a) Acrolein **16** (6 ml, 0.09 mol) was added over a period of 2 h to a stirred solution of ethyl-3-aminocrotonate **17** (10.5 g, 0.08 mol) and piperidine in 250 ml of anhydrous EtOH at 40—50 °C. After the addition was complete, the solution was heated to reflux for 3 h, during which time the color changed from yellow to brown. The solution was concentrated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (1 : 1)] to give 14 g (94.0%) of **18**. MS (*m/z*): 167 (M^+).

A solution of 1.0 N ceric ammonium nitrate/H_2O (100 ml) was added dropwise to a solution of **18** (4.2 g, 25.0 mmol) in 200 ml of acetone at 0 °C to maintain the temperature at 0—5 °C for 40 min. The resulting solution was stirred at 0° for 20 min. The solvent was removed by evaporation in vacuo and the residue was dissolved in $CHCl_3$ (300 ml). This solution was washed with saturated $NaHCO_3$ (150 ml), and saturated NH_4Cl, dried (Na_2SO_4), and evaporated *in vacuo*. The residue was distilled to give 1.1 g (26.8%) of **19** as a yellow oil (bp 68—70 °C/3 mmHg). (R_2 = Me, R_4 = R_5 = H). ^{1}H-NMR ($CDCl_3$) δ: 1.40 (3H, t, J=7.2 Hz), 2.83 (3H, s), 4.38 (2H, q, J=7.2 Hz), 7.19 (1H, q), 8.18 (1H, dd), 8.60 (1H, q). MS (*m/z*): 165 (M^+).

Lithium aluminum hydride ($LiAlH_4$) (0.46 g) was dissolved in 50 ml of absolute THF and added in a dropwise manner to a stirred solution of **19** (1.0 g, 6.05 mmol) in 10 ml of THF at 0 °C. The mixture was stirred at 0 °C for 2 h. Excess $LiAlH_4$ was destroyed with AcOEt, 10 ml of a mixture of ice and water was added, and the THF was removed by distillation. The precipitate of $Al(OH)_3$ was filtered off and the resulting emulsion was extracted with AcOEt. The combined organic phases were dried (Na_2SO_4), filtered, and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (1 : 1)] to give 0.71 g (74.6%) of 3-hydroxymethylpyridine. ^{1}H-NMR ($CDCl_3$) δ: 2.49 (3H, s), 4.00 (1H, s), 4.70 (2H, s) ,7.18 (1H, q), 7.7 (1H, dd), 8.30 (1H, dd). MS (*m/z*): 123 (M^+).

A suspension of 61.6 g (0.71 mol) of freshly prepared MnO_2 in a solution of 4.85 g (39.4 mmol) of 3-hydroxymethylpyridine in 50 ml of $CHCl_3$ was stirred at reflux for 5 h. The mixture was filtered, and the oxide was washed with five 10 ml portion of Et_2O. The combined filtrate and washing were evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [$CHCl_3$–MeOH (15 : 1)] to give 3.6 g (66.7%) of **1a** as a colorless oil. ^{1}H-NMR ($CDCl_3$) δ: 2.90 (3H, s), 7.33 (1H, q), 8.13 (1H, dd), 8.65 (1H, dd), 10.35 (1H, s). MS (*m/z*): 121 (M^+).

2,4-Dimethyl-3-pyridinecarbaldehyde (1b) ^{1}H-NMR ($CDCl_3$) δ: 2.63 (3H, s), 2.84 (3H, s), 7.10 (1H, d), 8.50 (1H, d), 10.65 (1H, s). MS (*m/z*): 151 (M^+).

4-Ethyl-2-methyl-3-pyridinecarbaldehyde (1c) ^{1}H-NMR ($CDCl_3$) δ: 1.25 (3H, t, J=6.8 Hz), 2.81(3H, s), 3.05 (2H, q, J=6.8 Hz), 7.16 (1H, d), 8.49 (1H, d), 10.62 (1H, s). MS (*m/z*): 149 (M^+).

2-Methyl-4-propyl-3-pyridinecarbaldehyde (1d) ^{1}H-NMR ($CDCl_3$) δ: 0.83 (3H, t, J=6.2 Hz), 1.40—1.91 (2H, m), 2.65—3.10 (5H, m), 7.10 (1H, d), 8.49 (1H, d), 10.60 (1H, s). MS (*m/z*): 179 (M^+).

4-Methyl-2-propyl-3-pyridinecarbaldehyde (1e) ^{1}H-NMR ($CDCl_3$) δ: 1.01 (3H, t, J=6.2 Hz), 1.57—1.95 (2H, m), 2.60 (3H, s), 3.12 (2H, t, J=6.2 Hz), 7.07 (1H, d), 8.51 (1H, d), 10.60 (1H, s). MS (*m/z*): 163 (M^+).

2-Butyl-4-methyl-3-pyridinecarbaldehyde (1f) ^{1}H-NMR ($CDCl_3$) δ: 0.91 (3H, t, J=6.1 Hz), 0.99—1.80 (4H, m), 2.60 (3H, s), 3.13 (2H, q, J=6.1 Hz), 7.06 (1H, d), 8.50 (1H, d), 10.61 (1H, s). MS (*m/z*): 177 (M^+).

4-Methyl-2-pentyl-3-pyridinecarbaldehyde (1g) ^{1}H-NMR ($CDCl_3$) δ: 0.90 (3H, t, J=6.3 Hz), 1.17—1.90 (6H, m), 2.60 (3H, s), 8.50 (1H, d), 10.60 (1H, s). MS (*m/z*): 191 (M^+).

General Method B for the Synthesis of 4,6-Trisubstituted Pyridine-3-carbaldehyde. 6-Methyl-4-propyl-3-pyridinecarbaldehyde (2a) (R_4 = *n*-Propyl, R_6 = Methyl) *n*-Propylmagnesium bromide (*n*-PropylMgBr) prepared from 1-bromopropane (2.1 ml, 23.0 mmol) and magnesium (0.61 g) in THF, was added to CuI (0.12 g) and dry THF (40 ml) in a 100 ml flask at −20 °C under Ar, and the mixture was stirred for 0.5 h at this temperature. It was then cooled to −78 °C and transferred to a suspension of 3-[(*tert*-butyl-dimethylsilyl)oxy]methyl-6-methylpyridine **20** (5.0 g, 21.0 mmol) and ethyl chloroformate (2.0 ml, 21.0 mmol) in THF. The mixture was allowed to warm slowly to room temperature with stirred. After the reaction mixture was stirring. Stirring was continued for 3 h at room temperature, then aqueous 5% $NaHCO_3$ was added and the THF was evaporated off under reduced pressure. The product was extracted with Et_2O and the solution was dried (Na_2SO_4), filtered, and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (1 : 10)] to give 3.5 g (47.1%) of **21**.

A solution of **21** (3.5 g, 9.9 mmol), (n-Bu)$_4$NF/THF in 50 ml of THF

was stirred at 20 °C for 3 h. THF was evaporated off *in vacuo*. The product was extracted with AcOEt and the solution was dried (Na_2SO_4), filtered and evaporated to provide the crude product. This was placed in a flask and stirred for 6 h under a stream of oxygen. Then the product was extracted with CH_2Cl_2 and the combined extracts were dried (Na_2SO_4), filtered and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (1 : 1)] to give 440 mg of **22**. ^{1}H-NMR ($CDCl_3$) δ: 1.0 (3H, t, J=6.8Hz), 1.52—1.77 (2H, m), 2.11 (1H, br s), 4.69 (2H, s), 6.98 (1H, s), 8.32 (1H, s). MS (*m/z*): 165 (M^+).

A suspension of 2.5 g (29.0 mmol) of freshly prepared MnO_2 in a solution of 0.48 g (2.90 mmol) of 3-hydroxymethylpyridine (**22**) in 30 ml of $CHCl_3$ was stirred at reflux for 2.5 h. The mixture was filtered, and the oxide was washed with five 10 ml portion of Et_2O. The combined filtrate and washing were evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (3 : 1)] to give 0.38 mg (80.1%) of **2a** as a colorless oil. ^{1}H-NMR ($CDCl_3$) δ: 1.01 (3H, t, J=6.8 Hz), 2.60 (3H, s), 2.91 (2H, t, J=6.8 Hz), 7.07 (1H, s), 8.82 (1H, s), 10.22 (1H, s). MS (*m/z*): 163 (M^+).

General Method C for the Synthesis of 2,4,6-Trisubstituted Pyridine-3-carbaldehyde. 2-Butyl-6-chloro-4-methyl-3-pyridinecarbaldehyde (3a) (R_2 = Butyl, R_4 = Methyl, R_6 = Chlorine) A solution of *m*-chloroperbenzoic acid (1.7 g, 9.9 mmol) in $CHCl_3$ (23 ml) was added dropwise to a solution of **23** (2.0 g, 9.0 mmol) in $CHCl_3$ (15 ml) at room temperature. After the addition was completed, the reaction mixture was stirred at the same temperature for 4 h. The resulting mixture was washed with saturated $NaHCO_3$, and the aqueous layer was further extracted with $CHCl_3$. The organic extracts were combined, washed with brine, dried (Na_2SO_4) and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [$CHCl_3$–MeOH (30 : 1)] to give 1.50 g (73.7%) of **24** as a colorless oil. ^{1}H-NMR ($CDCl_3$) δ: 1.03 (3H, t, J=6.8 Hz), 1.42 (3H, t, J=7.2 Hz), 1.21—1.78 (4H, m), 2.28 (3H, s), 2.84 (2H, q, J=6.8 Hz), 4.47 (2H, q, J=7.2 Hz), 7.00 (1H, d), 8.18 (1H, d). MS (*m/z*): 273 (M^+).

Compound **24** (200 mg, 0.84 mmol) was added to 3.0 ml of $POCl_3$ and the mixture was refluxed for 2 h. Excess $POCl_3$ was removed by distillation, and the residue was concentrated and extracted with AcOEt. The organic extracts were combined, washed with brine, dried (Na_2SO_4) and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt] to give 42 mg (42.7%) of **25**. ^{1}H-NMR ($CDCl_3$) δ: 1.01 (3H, t, J=6.8 Hz), 1.40 (3H, t, J=7.2 Hz), 1.24—1.70 (4H, m), 2.30 (3H, s), 2.69 (2H, q, J=6.8 Hz), 4.46 (2H, q, J=7.2 Hz), 7.02 (1H, s). MS (*m/z*): 254 (M^+).

A solution of $LiAlH_4$ (0.34 g, 9.0 mmol) in 20 ml of absolute THF was added dropwise with stirring to a solution of **25** (1.1 g, 4.5 mmol) in 10 ml of THF at 0 °C. The mixture was stirred at 20 °C for 2 h. Excess $LiAlH_4$ was destroyed with AcOEt, then 10 ml of a mixture of ice and water was added, and the THF was removed by distillation. The precipitate of $Al(OH)_3$ was filtered off and the resulting emulsion was extracted with AcOEt. The combined organic phases were dried (Na_2SO_4), filtered and evaporated to provide 0.79 mg (82.9%) of the crude 3-hydroxymethyl pyridine.

A suspension of freshly prepared MnO_2 (5.8 g) in a solution of the above product (460 mg, 2.2 mmol) in 50 ml of $CHCl_3$ was stirred at reflux for 5 h. The mixture was filtered, and the oxide was washed with five 10 ml portion of Et_2O. The combined filtrate and washing were evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (1 : 20)] to give 0.4 g (87.8%) of **3a** as an oil. ^{1}H-NMR ($CDCl_3$) δ: 0.92 (3H, t, J=6.8 Hz), 1.29—1.71 (4H, m), 2.57 (3H, s), 3.10 (2H, t, J=6.8 Hz), 7.08 (1H, s), 10.53 (1H, s). MS (*m/z*): 211 (M^+).

General Method D for the Synthesis of 2,4,5,6-Tetrasubstituted Pyridine-3-carbaldehyde. 2-Butyl-4-benzyloxymethyl-6-methyl-3-pyridine-carbaldehyde (4a) (R_2 = Butyl, R_4 = Benzyloxymethyl, R_6 = Methyl) A slurry of the enone **27** (6.37 g, 33.5 mmol) and ethyl valeryl acetate (11.5 g, 67.0 mmol) in absolute EtOH (100 ml) was treated with a solution of EtONa in EtOH. The mixture was stirred at room temperature for 4 h, concentrated to 50 ml and partitioned between 50% saturated NH_4Cl and Et_2O. The layers were separated and the Et_2O layer was washed with H_2O, and brine, dried (Na_2SO_4), filtered and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (1 : 10)] to give 10.2 g (88.7%) of **28**.

A mixture of **28** (8.7 g, 24.1 mmol), $AcONH_4$ (5.7 g, 72.2 mmol), and $Cu(OAc)_2$ (12.0 g, 60.2 mmol) in glacial AcOH (100 ml) was refluxed for 6 h. The solution was cooled to room temperature and subsequently poured into an ice-cold mixture of concentrated NH_4OH (110 ml). The mixture was extracted with Et_2O (300 ml), and Et_2O solution was washed with brine, dried (Na_2SO_4) and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (1 : 10)] to give 2.0 g (25.0%) of **30** as a colorless oil. ^{1}H-NMR ($CDCl_3$) δ: 0.92 (3H, t, J=6.8 Hz), 1.32 (3H, t, J=7.0 Hz), 1.20—1.90 (4H, m), 2.54 (3H, s), 4.31 (2H, q, J=7.0 Hz), 4.54 (2H, s), 7.11 (1H, s), 7.33 (5H, s). MS (*m/z*): 341 (M^+).

Compound (**1d**) was prepared in a similar manner (C, D) from **30**. ^{1}H-NMR ($CDCl_3$) δ: 0.96 (3H, t, J=6.8 Hz), 1.10—2.00 (4H, m), 2.61 (3H, s), 3.16 (2H, t, J=6.8 Hz), 4.68 (2H, s), 4.93 (2H, d), 7.37 (5H, s), 10.5 (1H, s). MS (*m/z*): 397 (M^+).

4-Acetoxymethyl-2-butyl-3-[(2′-carbmethoxybiphenyl-4-yl)methyl]-pyridine(32), 5-Acetoxy-2-butyl-4-methyl-3-[(2′-carbmethoxybiphenyl-4-yl)methyl]pyridine(33) A mixture of the N-oxide **31** (0.35 mmol), Ac_2O (3.6 ml) and AcOH (15 ml) was heated at 120 °C for 2 h. The excess Ac_2O and other volatile materials were removed *in vacuo*. The residue was extracted with AcOEt. The organic extracts were combined, washed with brine, dried (Na_2SO_4) and evaporated *in vacuo* to give a mixture of **32** and **33**. The mixture was separated by silica gel column chromatography [AcOEt–hexane (1 : 3)] to give 70 mg (48%) of the 4-acetyl methyl product **32** and 50 mg (35%) of the 5-acetoxy product **33**. **32**: ^{1}H-NMR ($CDCl_3$) δ: 0.92 (3H, t, J=6.2 Hz), 1.15—1.90 (4H, m), 2.11 (3H, s), 2.38 (3H, s) 2.75 (2H, t, J=6.2 Hz), 3.65 (3H, s), 4.16 (2H, s), 6.85—8.30 (9H, m). **33**: ^{1}H-NMR ($CDCl_3$) δ: 0.89 (3H, t, J=6.2 Hz), 1.15—2.00 (4H, m), 2.02 (3H, s), 2.76 (2H, t, J=6.2 Hz), 3.62 (3H, s), 4.14 (2H, s), 5.06 (2H, s), 6.95—8.53 (10H, m).

2-Butyl-4-hydroxymethyl-3-[(2′-carbbiphenyl-4-yl)methyl]pyridine (8h) A solution of **32** (70 mg, 0.16 mmol) in 5 ml of MeOH and 2 ml of 10% NaOH was refluxed for 5 h. After cooling, the reaction mixture was filtered, and evaporated *in vacuo*. The residue was dissolved in water, and the solution was acidified to pH 4.5 with 10% HCl. The precipitated solid was recovered by filtration and recrystallized from iso-propyl alcohol (IPA) to afforded 8 h (40 mg, 66.0%). mp 178—179.5 °C. ^{1}H-NMR ($CDCl_3$) δ: 0.75 (3H, t, J=6.2 Hz), 0.89—1.60 (4H, m), 2.16—2.30 (2H, t, J=6.2 Hz), 4.01 (2H, s), 4.58 (2H, s), 6.94—8.50 (12H, m). MS (*m/z*): 375 (M^+). IR (KBr): 2854, 2722, 1446 cm^{-1}.

5-Acetoxy-2-butyl-4-methyl-3-[(2′-carbbiphenyl-4-yl)methyl]pyridine (8j) The title compound was obtained from **32** by a procedure similar to that described for **8h**. A colorless solid, 30 mg, (68.9%), mp 242—244 °C. ^{1}H-NMR ($CDCl_3$) δ: 0.92 (3H, t, J=6.2 Hz), 1.10—1.90 (4H, m), 2.18 (3H, s), 2.75 (2H, t, J=6.2 Hz), 4.12 (2H, s), 7.00—7.95 (8H, m), 8.06 (1H, s). MS (*m/z*): 375 (M^+). IR (KBr): 2944, 1548, 1461, 1390 cm^{-1}.

2-Butyl-5-methoxy-4-methyl-3-[(2′-carbbiphenyl-4-yl)methyl]pyridine (8l) A solution of CH_2N_2 (240 mg, 5.7 mmol) in Et_2O (30 ml) was added dropwise to a solution of **8k** (0.7 g, 1.9 mmol) in THF (30 ml) at 0 °C. After the addition was completed, the reaction mixture was stirred at room temperature for 4 h. The solvent was removed *in vacuo* and the residue was subjected to column chromatography on silica gel [hexane–AcOEt (1 : 1)] to give 0.4 g (54.8%) of the 5-methoxy pyridine.

A solution of the 5-methoxy pyridine (60 mg, 0.15 mmol) in MeOH (5 ml) and 10% NaOH (2 ml) was refluxed for 5 h. After cooling, the reaction mixture was filtered and the solvent was removed *in vacuo*. The residue was dissolved in water and the solution was acidified to pH 4.5 with 10% HCl. The residue was extracted with $CHCl_3$. The organic extracts were combined, washed with brine, dried (Na_2SO_4) and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt] to give 55 mg (94.9%) of **8l**. ^{1}H-NMR ($CDCl_3$) δ: 0.78 (3H, t, J=6.2 Hz), 1.13—1.70 (4H, m), 2.21 (3H, s), 3.87 (3H, s) 2.75 (2H, t, J=6.2 Hz), 4.12 (2H, s), 6.78—7.98 (8H, m), 8.24 (1H, s). MS (*m/z*): 389 (M^+) IR (KBr): 2920, 1707, 1460, 1290 cm^{-1}.

2-Methyl-3-[(2′-carbbiphenyl-4-yl)methyl]pyridine (8a) ^{1}H-NMR ($CDCl_3$) δ: 2.21 (3H, s), 4.0 (2H, s), 6.90—8.59 (11H, m), 9.18 (1H,d). MS (*m/z*): 303 (M^+). IR (KBr): 1686, 1851, 1446, 1290 cm^{-1}.

2,4-Dimethyl-3-[(2′-carbbiphenyl-4-yl)methyl]pyridine (8b) ^{1}H-NMR ($CDCl_3$) δ: 2.26 (3H, s), 2.42 (3H, s), 4.09 (2H, s), 6.93—8.27 (10H, m). MS (*m/z*): 316 (M^+). IR (KBr): 3418, 1941, 1689, 1599, 1443 cm^{-1}.

4-Ethyl-2-methyl-3-[(2′-carbbiphenyl-4-yl)methyl]pyridine (8c) ^{1}H-NMR ($CDCl_3$) δ: 1.15 (3H, t, J=7.2 Hz), 2.42 (3H, s), 2.80 (2H, t, J=7.2 Hz), 4.12 (2H, s), 6.92—8.33 (10H, s). MS (*m/z*) 330 (M^+). IR (KBr): 3448, 1677, 1602 cm^{-1}.

2-Methyl-4-propyl-3-[(2′-carbbiphenyl-4-yl)methyl]pyridine (8d) ^{1}H-

NMR ($CDCl_3$) δ: 1.02 (3H, t, J=7.0 Hz), 1.50—1.83 (2H, m), 2.52 (3H, s), 2.81 (2H, t, J=7.0 Hz), 4.26 (2H, s), 6.99—8.34 (10H, m). MS (*m/z*): 345 (M^+). IR (KBr): 2950, 1689 cm^{-1}.

4-Methyl-2-propyl-3-[(2′-carbbiphenyl-4-yl)methyl]pyridine (8e) ^{1}H-NMR ($CDCl_3$) δ:0.79 (3H, t, J=7.0 Hz), 1.25—1.70 (2H, m), 2.51 (2H, t, J=7.0 Hz), 4.09 (2H, s), 6.87—8.52 (10H, m). MS (*m/z*): 345 (M^+). IR (KBr): 3004, 2956, 2866, 1704 cm^{-1}.

4-Methyl-2-pentyl-3-[(2′-carbbiphenyl-4-yl)methyl]pyridine (8g) ^{1}H-NMR ($CDCl_3$) δ: 0.85 (3H, t, J=6.8 Hz), 1.21—1.78 (4H, m), 2.46 (3H, s), 3.21 (2H, t, J=6.8 Hz), 4.19 (2H, s), 7.19—8.53 (9H, m). MS (*m/z*): 373 (M^+). IR (KBr): 3032, 2916, 2688, 1710, 1620 cm^{-1}.

6-Methyl-4-propyl-3-[(2′-carbbiphenyl-4-yl)methyl]pyridine (8i) ^{1}H-NMR ($CDCl_3$) δ: 1.01 (3H, t, J=7.0 Hz), 1.36—1.70 (2H, m), 2.51 (3H, s), 2.59 (2H, t, J=7.0 Hz), 4.04 (2H, s), 7.00—7.71 (10H, m). MS (*m/z*): 345 (M^+). IR (KBr): 2950, 1602, 1548 cm^{-1}.

Ethyl 2-Butyl-4,5-dimethylnicotinate-*N*-oxide (34) A solution of *m*CPBA (3.00 g, 16.91 mmol) in $CHCl_3$ (50 ml) was added dropwise to a solution of **19** (2.60 g, 11.34 mmol) in $CHCl_3$ (10 ml) at room temperature. After the addition was completed, the reaction mixture was stirred at the same temperature for 4 h. The resulting mixture was washed with saturated $NaHCO_3$, and the aqueous layer was further extracted with $CHCl_3$. The organic extracts were combined, washed with brine, dried (Na_2SO_4) and evaporated in vacuo. The resulting product was purified by silica gel column chromatography [$CHCl_3$–MeOH (1 : 10)] to give 3.05 g (98.3%) of **34**. ^{1}H-NMR ($CDCl_3$) δ: 0.80—1.10 (3H, m), 1.41 (3H, t, J=7.3 Hz), 1.10—1.80 (4H, m), 2.16 (3H, s), 2.19 (3H, s), 2.70—2.76 (2H, t, J=7.3 Hz), 4.43 (2H, q, J=7.3 Hz), 8.09 (1H, s). MS (*m/z*): 251 (M^+).

Ethyl 2-Butyl-6-chloro-4,5-dimethylnicotinate (35) The title compound was obtained from **34** by a procedure similar to that described for **23**. ^{1}H-NMR ($CDCl_3$) δ: 0.80—1.10 (3H, m), 1.45 (3H, t, J=7.3 Hz), 1.15—1.85 (4H, m), 2.22 (3H, s), 2.35 (3H, s), 2.56 (2H, t, J=7.3 Hz), 4.56 (2H, q, J=7.3 Hz). MS (*m/z*): 271 (M^+).

2-Butyl-6-chloro-4,5-dimethyl-3-pyridinecarbaldehyde (36) The title compound was obtained from **35** by a procedure similar to that described for **25**. ^{1}H-NMR ($CDCl_3$) δ: 0.80—1.03 (3H, t, J=7.0 Hz), 1.15—1.85 (4H, m), 2.38 (3H, s), 2.91 (3H, s) 2.87—3.10 (3H, t, J=7.0 Hz), 10.54 (1H, s). MS (*m/z*): 227 (M^+).

2-Butyl-6-chloro-4,5-dimethyl-3-[4,4-dimethyl-2-(4′-methylbiphenyl-2-yl)oxazolyl]pyridine (37) The title compound was obtained from **36** by a procedure similar to that described for **6**. ^{1}H-NMR ($CDCl_3$) δ: 0.91 (3H, t, J=6.2 Hz), 1.25 (6H, s), 1.10—1.85 (4H, m), 2.10 (3H, s) 2.42 (3H, s), 3.04 (2H, t, J=8.1 Hz), 4.15 (2H, s), 6.95—7.80 (8H, m). MS (*m/z*): 404 (M^+).

2-Butyl-6-chloro-4,5-dimethyl-3-[(2′-cyanobiphenyl-4-yl)methyl]-pyridine-*N*-oxide (38) A solution of **37** (3.18 g, 6.90 mmol) in 10 ml of pyridine at 10 °C was treated dropwise with $POCl_3$ (1.3 ml) such that the reaction temperature did not exceed 15 °C. The reaction mixture was stirred at 100 °C for 3 h, cooled to room temperature. The reaction was quenched by addition of water, and the resulting emulsion was extracted with AcOEt. The extract was washed with brine, dried (Na_2SO_4) and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (1 : 3)] to give 2.27 g (93.1%) of the 2′-cyanobiphenyl.

A solution of *m*CPBA (0.80 g, 4.64 mmol) in $CHCl_3$ (40 ml) was added dropwise to a solution of the 2′-cyanobiphenyl (1.20 g, 3.04 mmol) in $CHCl_3$ (15ml) at room temperature. After the addition was completed, the reaction mixture was stirred at the same temperature for 4 h. The resulting mixture was washed with saturated $NaHCO_3$, and the aqueous layer was further extracted with $CHCl_3$. The organic extracts were combined, washed with brine, dried (Na_2SO_4) and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [$CHCl_3$–MeOH (30 : 1)] to give 0.92 g (73.7%) of **38**. ^{1}H-NMR ($CDCl_3$) δ: 0.91 (3H, t, J=6.2 Hz), 1.00—1.85 (4H, m), 2.20 (3H, s), 2.42 (3H, s), 3.04 (2H, t, J=8.1 Hz), 4.15 (2H, s), 6.95—7.80 (8H, m). MS (*m/z*): 404 (M^+).

4-Acetoxymethyl-2-butyl-6-chloro-5-methyl-3-[(2′-cyanobiphenyl-4-yl)methyl]pyridine (39) A mixture of the N-oxide **38** (0.92 g, 2.28 mmol), Ac_2O (20 ml) and AcOH (20 ml) was stirred at 120 °C for 2 h. The excess Ac_2O and other volatile materials were removed by rotary evaporation and the residue was extracted with AcOEt. The organic extracts were combined, washed with brine, dried (Na_2SO_4) and evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (1 : 8)] to give 0.45 g (44.3%) of **39** as a colorless oil. ^{1}H-NMR ($CDCl_3$) δ: 0.88 (3H, t, J=6.4 Hz), 1.05—1.85 (4H, m), 1.85 (3H, s), 2.44 (3H, s), 2.77 (2H, t, J=9.0 Hz), 4.21 (2H, s), 5.08 (2H, s), 6.95—7.80 (3H, m). MS (*m/z*): 446 (M^+).

2-Butyl-6-chloro-4-hydroxymethyl-5-methyl-3-[(2′-cyanobiphenyl-4-yl)methyl] pyridine (40) A catalytic amount of 10% K_2CO_3 was added to a solution of **39** (0.45 g, 1.00 mmol) in MeOH (10 ml) and the mixture was stirred for 1.5 h. After addition of H_2O to the mixture and neutralization with 10% HCl, the solution was extracted with $CHCl_3$. The organic extracts were combined, washed with brine, dried (Na_2SO_4) and was evaporated *in vacuo*. The resulting product was purified by silica gel column chromatography [AcOEt–hexane (1 : 2)] to give 0.38 g (94.1%) of **40** as a colorless oil. ^{1}H-NMR ($CDCl_3$) δ: 0.87 (3H, t, J=6.4 Hz), 1.10—1.80 (5H, m), 2.47 (3H, s), 2.76 (2H, t, J=8.6 Hz), 4.24 (2H, s), 4.56—4.70 (2H, br s), 7.00—7.80 (3H, m). MS (*m/z*): 404 (M^+).

2-Butyl-6-chloro-4-hydroxymethyl-5-methyl-3-[[2′-(1*H*-tetrazol-5-yl)-biphenyl-4-yl]methyl]pyridine (9h) A solution of **40** (0.38 g, 0.94 mmol) and trimethyltin azide (0.77 g, 3.76 mmol) in 50 ml of toluene was refluxed for 20 h. The mixture was cooled to room temperature, then filtered and the solvent was evaporated *in vacuo*. The residue was dissolved in CH_2Cl_2–MeOH–NH_3 (10 ml, 5 : 3 : 1, v/v) and the mixture was stirred for 0.5 h. The solvent was removed by a rotary evaporator and column chromatography on silica gel [$CHCl_3$–MeOH (30 : 1)] gave 0.21 g (50.0%) of **9h** as a white solid. ^{1}H-NMR($CDCl_3$ + CD_3OD) δ: 0.87 (3H, t, J=6.4 Hz), 1.10—1.78 (5H, m), 2.47 (3H, s), 2.76 (2H, t, J=6.4 Hz), 3.91 (2H, br s), 4.17 (2H, s), 7.56 (2H, s), 6.82—7.80 (8H, m). MS (*m/z*): 447 (M^+). IR (KBr): 2944, 2854, 2404, 1596 cm^{-1}.

2-Butyl-4-methyl-3-[[2′-(1*H*-tetrazol-5-yl)biphenyl-4-yl]methyl]pyridine (9a) ^{1}H-NMR ($CDCl_3$) δ: 0.85 (3H, t, J=6.4 Hz), 1.00—1.70 (4H, m), 2.00 (3H, s), 2.35 (2H, t, J=6.4 Hz), 3.90 (2H, s), 6.60—7.90 (10H, m). MS (*m/z*): 383 (M^+). IR (KBr): 2950, 2920, 2860, 1596 cm^{-1}.

2-Butyl-6-chloro-4-methyl-3-[[2′-(1*H*-tetrazol-5-yl)biphenyl-4-yl]-methyl]pyridine (9b) ^{1}H-NMR ($CDCl_3$) δ: 0.85 (3H, t, J=6.4 Hz), 1.05—1.80 (4H, m), 2.08 (3H, s), 2.33 (2H, t, J=6.4 Hz), 3.97 (2H, s), 6.78—7.90 (9H, m). MS (*m/z*): 417 (M^+). IR (KBr): 3004, 2950, 2860, 2693 cm^{-1}.

2-Butyl-4,5-dimethyl-3-[[2′-(1*H*-tetrazol-5-yl)biphenyl-4-yl]methyl]-pyridine (9c) ^{1}H-NMR ($CDCl_3$) δ: 0.86 (3H, t, J=6.4 Hz), 1.00—1.80 (4H, m), 2.11 (3H, s), 2.21 (3H, s), 2.68 (2H, t, J=6.4 Hz), 3.45 (1H, s), 4.06 (2H, s), 6.75—7.85 (8H, m), 7.87 (1H, s). MS (*m/z*): 397 (M^+) . IR (KBr): 2944, 2854, 2404, 1596 cm^{-1}.

2-Butyl-6-chloro-4,5-dimethyl-3-[[2′-(1*H*-tetrazol-5-yl)biphenyl-4-yl]-methyl]pyridine (9d) ^{1}H-NMR ($CDCl_3$) δ: 0.88 (3H, t, J=6.4 Hz), 1.00—1.80 (4H, m), 2.16 (3H, s), 2.34 (3H, s), 2.75 (2H, t, J=6.4 Hz), 4.06 (1H, s), 4.18 (2H, s), 6.80—7.75 (8H, m). MS (*m/z*): 431 (M^+). IR (KBr): 2944, 2854, 2718, 1563 cm^{-1}.

2-Butyl-4-hydroxymethyl-3-[[2′-(1*H*-tetrazol-5-yl)biphenyl-4-yl]-methyl]pyridine (9e) ^{1}H-NMR ($CDCl_3$ + CD_3OD) δ: 0.90 (3H, t, J=6.4 Hz), 1.00—1.78 (4H, m), 2.81 (2H, t, J=6.4 Hz), 4.01 (2H, s), 4.50 (2H, s), 6.77—7.76 (9H, m), 8.32 (1H, s). MS (*m/z*): 399 (M^+). IR (KBr): 3352, 2920, 2854, 2434, 1596 cm^{-1}.

2-Butyl-4-hydroxymethyl-6-chloro-3-[[2′-(1*H*-tetrazol-5-yl)biphenyl-4-yl]methyl]pyridine (9f) ^{1}H-NMR ($CDCl_3$) δ:0.87 (3H, t, J=6.4 Hz), 1.10—1.72 (4H, m), 2.52 (2H, t, J=6.4 Hz), 3.95 (2H, s), 4.50 (2H, s), 6.75—7.87 (9H, m). MS (*m/z*): 433 (M^+). IR (KBr): 3330, 2944, 1730, 1578, 1380 cm^{-1}.

2-Butyl-4-hydroxymethyl-5-methyl-3-[[2′-(1*H*-tetrazol-5-yl)biphenyl-4-yl]methyl]pyridine (9g) ^{1}H-NMR ($CDCl_3$ + CD_3OD) δ: 0.85 (3H, t, J=6.4 Hz), 1.00—1.75 (4H, m), 2.40 (3H, s), 2.65 (2H, t, J=6.4 Hz), 4.21 (2H, s), 4.57 (2H, s), 6.70—7.80 (8H, m), 8.18 (1H, s). MS (*m/z*): 413 (M^+). IR (KBr): 2944, 2854, 2080, 1602 cm^{-1}.

2-Butyl-4-cyanomethyl-3-[[2′-(1*H*-tetrazol-5-yl)biphenyl-4-yl]methyl]-pyridine (9i) ^{1}H-NMR ($CDCl_3$ + CD_3OD) δ: 0.95 (3H, t, J=6.4 Hz), 1.10—1.82 (4H, m), 2.85 (2H, t, J=6.4 Hz), 3.68 (2H, s), 4.11 (2H, s), 6.80—7.13 (4H, m), 7.17—8.45 (6H, m). MS (*m/z*): 408 (M^+). IR (KBr): 2914, 2428, 2250, 1941, 1590 cm^{-1}.

2-Butyl-4-hydroxymethyl-6-methyl-3-[[2′-(1*H*-tetrazol-5-yl)biphenyl-4-yl]methyl]pyridine (9j) ^{1}H-NMR (CD_3OD) δ: 0.90 (3H, t, J=6.4 Hz), 1.00—1.70 (4H, m), 2.64 (3H, s), 2.83 (2H, t, J=6.4 Hz), 4.09 (2H, s), 6.80—7.70 (9H, m). MS (*m/z*): 413 (M^+). IR (KBr): 2944, 1653 cm^{-1}.

AII Receptor Binding Assay Binding assay for AT_1 receptor in rat liver membranes or for AT_2 receptor in bovine cerebellar membranes

were performed using a commercial assay Kit (DuPont-NEN, Boston, MA, U.S.A.). [^{125}I]Sar^1,Ile^8-AII (17 pM) and [^{125}I]AII (74 pM) were used for AT_1 and AT_2 receptor binding assay, respectively. The test compounds (25 μl) and the radioligands (25 μl) were added to test tubes. Membrane fractions (200 μl) for each receptor binding assay were then added to initiate the binding reaction. Incubation was performed for 3 h at 27 °C (AT_1 receptor binding assay) or for 1 h at 37 °C (AT_2 receptor binding assay). After incubation, the reaction was terminated by the addition of 3 ml of ice-cold 0.9% saline. To separate the bound and free radioactivity, the reaction mixtures were immediately filtered under pressure through glass-fiber GF/C filters (Whatman Inc., Tokyo, Japan) presoaked with filter soak solution, and each tube and filter was rinsed twice with ice-cold 0.9% saline. The radioactivity trapped on the filters was counted using a gamma counter (ARC-300 ; Aloka, Tokyo, Japan). Nonspecific binding of [^{125}I]Sar^1, Ile^8-AII to the receptor was estimated in the presence of 10^{-5} M unlabeled AII. The inhibitory concentration of test compound that gave a 50% inhibition of the specific binding of AII (IC_{50}) was determined by regression analysis of displacement curves.

Effects on AII Contractile Responses in Isolated Rabbit Aorta Thoracic aorta was isolated from male New Zealand White rabbits (2.7—3.2 kg, Tokyo Laboratory Animals Science Inc., Tokyo, Japan). The aorta was cleaned of connective tissue and adherent fat and cut into 3 mm ring segments. Opened ring preparations were used in the present study. The vascular endothelium was removed by gently rubbing the intimal surface of the blood vessel with cotton wool. Preparations were mounted vertically in organ baths containing 10 ml of Krebs–Ringer solution (NaCl, 120.3; KCl, 4.8; $MgSO_4 \cdot 7H_2O$, 1.3; KH_2PO_4, 1.2; $CaCl_2 \cdot 2H_2O$, 1.2; $NaHCO_3$, 24.2 and glucose, 5.5 mM) maintained at 37 °C and bubbled with a 95% O_2 + 5% CO_2 gas. Under the resting tension of 1.5 g, isometric tension changes were recorded on a polygraph (Nihon Kohden, Tokyo, Japan) through a force displacement transducer (T7-30-240; Orientec, Tokyo, Japan). After a 1.5 h equilibration period, the cumulative concentration–response curve for AII was constructed by the method of stepwise addition of the agonist. Then, AII was washed out repeatedly for 1 h. Tissues were incubated with various concentrations of compounds for 20 min and the concentration–response curve for AII was again obtained. Responses were expressed as a percentage of the maximum response in the first concentration–response curve for AII. To measure the potency of the antagonist, the pA_2 values were determined from Schild plots using the least squares method. The effects of KT3-579 on the contractile responses of rabbit aorta induced by KCl, norepinephrine and serotonin were also examined.

References

1) W. W. Douglas, "Goodman and Gilman's The Pharmacological Basis of Therapeutics 7th Ed.," ed. by A. G. Gilman, L. S. Goodman, T. W. Rall, F. Murad, Macmillan Pub. Co., New York, 1975, pp. 639—659.
2) H. Okunishi, M. Miyazaki, T. Okamura, N. Toda, *Biochem. Biophys. Res. Commun.*, **149**, 1186 (1987).
3) D. J. Carin, J. V. Duncia, P. E. Aldrich, A. T. Chiu, A. L. Johnson, M. E. Pierce, W. A. Price, J. B. Santell, III, G. J. Wells, R. R. Wexler, P. C. Wong, S-E. Yoo, P. B. M. W. M. Timmermans, *J. Med. Chem.*, **34**, 2525 (1991).
4) N. B. Mantlo, P. K. Chakravarty, D. L. Ondyeka, P. K. S. Siegl, R. S. Chang, V. J. Lotti, K. A. Faust, T. W. Schorn, C. S. Sweet, A. A. Patchett, W. J. Greenlee, *J. Med. Chem.*, **34**, 2919 (1991).
5) *a*) C. Bernhart, J-C. Brelier, J. Clement, D. Nisato, P. Perreaut EPA 454511 (1991); *b*) *Idem*, PCT#91/14679 (1991).
6) G. P. Schiemanz, "Organic Syntheses," Coll. Vol. 5, ed. by H. E. Baumgarten, John Wiley and Son, Inc., New York, 1973, p. 496.
7) T. Tanouchi, M. Kawamura, I. Ohyama, I. Kajiwara, Y. Iguchi, T. Okada, K. Iizuka, M. Nakazawa, *J. Med. Chem.*, **24**, 1149 (1981).
8) A. I. Meyers, E. D. Mihelich, *J. Am. Chem. Soc.*, **97**, 7383 (1975).
9) A. I. Meyers, D. L. Temple, D. Haidukewych, E. D. Mihelich, *J. Org. Chem.*, **39**, 2787 (1974).
10) I. M. Dordor, J. M. Mellor, P. D. Kennewell, *Tetrahedron Lett.*, **24**, 1437 (1983).
11) J. V. Duncia, M. E. Piece, J. B. Santella, III, *J. Org. Chem.*, **56**, 2395 (1991).
12) K. Tsuda, Y. Satoh, N. Ikekawa, H. Mishima, *J. Org. Chem.*, **21**, 800 (1956).
13) A. Guzman, M. Romero, M. L. Maddox, J. Muchowski, *J. Org. Chem.*, **55**, 5793 (1990).
14) R. Gosmini, P. Mangeney, A. Alexakis, M. Commerçon, J-F. Normant, *Synlett*, **1990**, 111.
15) E. P. Papadopoulos, A. Jakkar, C. H. Issidorides, *J. Org. Chem.*, **31**, 615 (1966).
16) E. F. V. Scriven, "Comprehensive Heterocyclic Chemistry," ed. by A. R. Katritzy, C. W. Rees, Pergamon Press Ltd., Oxford, 1984, p. 217.
17) E. M. Gordon, C. P. Ciosek, Jr., L. C. Rich, V. C. Dehmel, D. A. Slusarckyk, T. W. Harrity, A. K. Obrien, *J. Med. Chem.*, **34**, 2804 (1991).
18) A. McKillop, M-K. Bhagrath, *Heterocycles*, **23**, 1697 (1985).
19) Y. C. Martin, "Quantitative Drug Design," Japanese edition by T. Esaki, Chijin Shokan Co., Ltd., 1980, p. 58.
20) P. C. Wong, W. A. Price, A. T. Chiu, J. V. Duncia, D. J. Carini, R. R. Wexler, A. L. Johnson, P. B. M. W. M. Timmermans, *J. Pharmacol. Exp. Ther.*, **255**, 211 (1990).