Discovery of Novel *N*-Phenylglycine Derivatives as Potent and Selective β_3-Adrenoceptor Agonists for the Treatment of Frequent Urination and Urinary Incontinence

Nobuyuki Tanaka,* Tetsuro Tamai, Harunobu Mukaiyama, Akihito Hirabayashi, Hideyuki Muranaka, Satoshi Akahane, Hiroshi Miyata, and Masuo Akahane

Central Research Laboratory, Kissei Pharmaceutical Company Ltd., 4365-1, Hotaka, Nagano 399-8304, Japan

Received October 24, 2000

With a novel assay using isolated ferret detrusor to estimate β_3-adrenoceptor agonistic activity, we found that a series of glycine derivatives of ritodrine, a β_2-adrenoceptor agonist, are potent β_3-adrenoceptor agonists, with excellent selectivity versus β_1 and β_2 subtypes. Substitution of halogens in the phenyl ring increased potency and selectivity for the β_3-adrenoceptor, and this was dependent upon the position of the halogens. The chlorine-substituted derivatives **3f**−**i** exhibited potent β_3-adrenoceptor-mediated relaxation of ferret detrusor (EC_{50} = 0.93, 11, 14, and 160 nM) and higher potency at β_3-adrenoceptors than at β_1 or β_2. The intravenous administration of **3h** significantly reduced the urinary bladder pressure in anesthetized male rats (ED_{50} = 48 μg/kg) without cardiovascular side effects. This article is the first report of structure−activity relationships (SAR) concerning β_3-adrenoceptor agonists as agents for the treatment of urinary frequency and incontinence.

Introduction

In 1984, β_3-adrernergic receptors (AR) were proposed as a third group of β-ARs in addition to the β_1- and β_2-ARs found in rat adipose tissue utilizing various β-agonists.[1−4] In 1989, the primary structure of the β_3-AR was identified and characterized using cloning and molecular pharmacological techniques.[5−7] β_3-ARs are G-protein-linked receptors. Like other β-ARs, it has a seven transmembrane-spanning structure and plays a significant role in regulating lipolysis and thermogenesis in rodent and human adipose tissue. Moreover, studies of β_3-AR mRNA demonstrated that β_3-ARs exist in the human heart, gall bladder, gastrointestinal (GI) tract, and prostate in addition to adipocytes.[8] These findings encouraged us to look for potent β_3-AR agonists to treat various metabolic and gastrointestinal diseases, such as obesity, diabetes, and irritable bowel syndrome.[2] Recently, it has been suggested that β_3-ARs exist in the human detrusor and its relaxation is mediated mainly via β_3-ARs.[9−12] In the urinary bladder, urine storage on a peripheral receptor level is controlled by β_3-ARs, which accept signals from sympathetic nerves in the human detrusor. Therefore, we thought that potent, selective β_3-AR agonists might provide a new approach for the treatment of urinary bladder dysfunction, such as urinary frequency and incontinence. Although a number of antimuscarinic drugs are widely prescribed for such diseases,[13] their antimuscarinic activity has unacceptable side effects, such as dry mouth and constipation. In addition, antimuscarinic drugs are contraindicated in patients with urinary retention, because they inhibit the micturation pressure. It is noteworthy that β_3-AR agonists do not possess such adverse effects. However, if β_3-AR activity cannot be separated from β_1- and β_2-AR activity, β_3-AR agonists would have unfavorable effects, such as tachycardia or tremor. To find drugs without such adverse effects, this study evaluated the β_3-AR activity of compounds using the ferret detrusor in a novel assay.[14] We also examined β_1- and β_2-AR agonistic activity using rat atria and uteri.

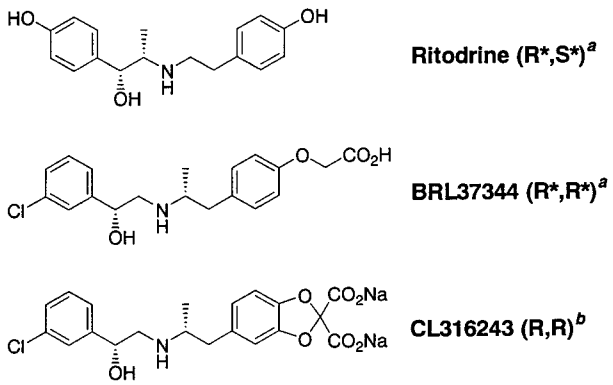


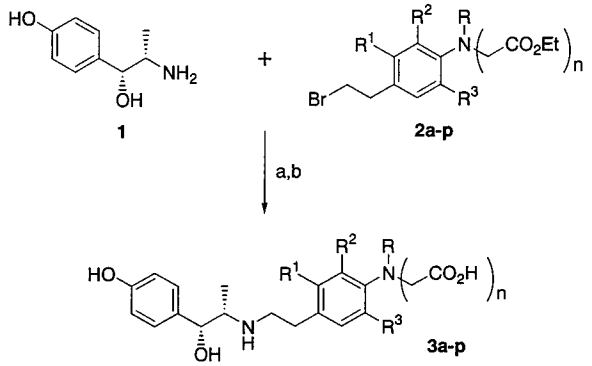
Figure 1. β_2- and β_3-AR agonists for clinical or research use. [a]The R* and S* in parentheses indicate the relative configuration of the hydroxy and methyl groups. [b]The R in parentheses indicates the absolute configuration of the hydroxy and methyl groups.

First, we evaluated some β-AR agonists that are used clinically or preclinically. As shown in Table 2, isoproterenol (a nonselective β-AR agonist), ritodrine[15] (a β_2-AR agonist), and CL316243[16] (a β_3-AR agonist) relax the ferret detrusor via β_3-AR activation.[14] BRL37344[3,17] (a β_3-AR agonist) produced the greatest relaxation and full agonistic activity. In comparison, the *N*-phenylglycine derivatives (**3**) produced sufficient detrusor-relaxation mediated via β_3-AR and exhibited β_3-AR selectivity against effects mediated via β_1- and β_2-AR. Moreover, most of them behaved as full agonists (80−100% maximum response relative to isoproterenol). Thus, the *N*-phenylglycine derivatives **3** appear to be a new class

* To whom correspondence should be addressed. Tel: +81-263-82-8820. Fax: +81-263-82-8827. E-mail: nobuyuki_tanaka@pharm.kissei.co.jp.

10.1021/jm000455z CCC: $20.00
Published on Web 04/03/2001

Scheme 1. General Method for Compounds **3**[a]



[a] Reagents: (a) DIPEA; (b) aq NaOH.

of β_3-AR agonist that might be useful for the treatment of urinary diseases, such as urinary frequency and incontinence, without undesirable side effects. The synthesis of these compounds (**3**) and the results of in vitro and in vivo assays are described in detail in the following sections.

Chemistry

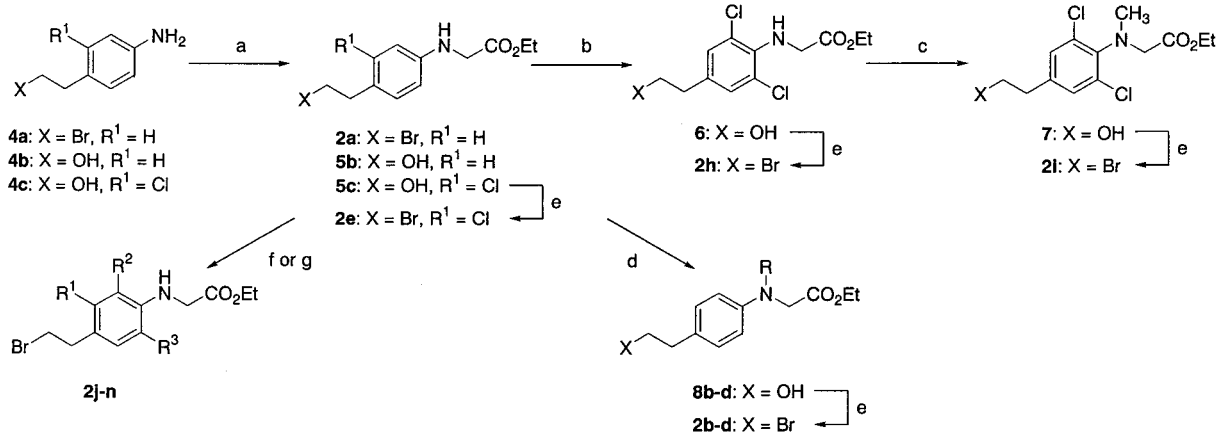
The *N*-phenylglycine derivatives **3** described herein (Table 1) were prepared from 4′-hydroxynorephedrine (**1**) and the correctly substituted aminophenethyl bromide (**2**) via the corresponding esters, followed by saponification, as illustrated in Scheme 1. Optically active 4′-hydroxynorephedrine (**1**) was obtained by optical resolution of its commercially available racemate using L-tartaric acid.[18]

The aminophenethyl bromide (**2**) incorporated glycine moieties were generally synthesized as shown in Scheme 2 except for the *N*-(2-chlorophenyl)glycine derivatives **2f,g**. The 4′-aminophenethyl derivatives **4a**−**c** were converted into the corresponding glycine ester analogues **2a** and **5b,c** by *N*-alkylation using ethyl 2-bromoacetate. These were then halogenated using *N*-halosuccinimide or hypochlorite esters to give the dichloroaniline derivatives (**6** from **5b**; **2j,k** from **2e** following bromination of **5c**), mono- and dibromo derivatives (**2l,m** from **2a**), and monoiodo derivative (**2n** from **2a**). *N*-Phenylglycine analogues (**7**, **8b**−**d**) bearing methyl or benzyl groups, or acetate on nitrogen, were derived using alkylating agents, such as iodomethane, benzyl bromide, and ethyl 2-bromoacetate. The hydroxy groups of all of the phenethyl alcohols described above were substituted by bromine using Ph_3P and CBr_4 to give the phenethyl bromide intermediates.

As mentioned above, *N*-(2-chlorophenyl)glycine derivatives **2f,g** cannot be synthesized in the same manner as the others, since selective *N*-monoalkylation of the 2-chloroaniline derivatives **9**[19] with ethyl 2-bromoacetate was not possible. This failure was assumed to be caused by similar alkylation rates for *N*-monoalkylation and *N*,*N*-dialkylation leading to *N*,*N*-dialkylation. In fact, this reaction consumed few 2-chloroaniline derivatives while using ethyl 2-bromoacetate in an amount equivalent to **9** and afforded only *N*,*N*-dialkylated aniline derivatives when using ethyl 2-bromoacetate in an amount exceeding twice the equivalent to **9**. Nagaraj et al. reported that 2-substituted anilines, particularly 2-chloro or 2-nitroaniline, are more liable to undergo *N*,*N*-diacetylation to give the *N*-phenyldiacetamide derivative than unsubstituted ones.[20] They assumed that the facility of *N*,*N*-dialkylation is determined by the inductive and steric effects of the substituents at the *ortho*-position on the aniline. This hypothesis might also hold in our case of *N*-alkylation, although the reaction type is quite distinct from *N*-acylation. Therefore, we planned to improve the *N*-monoalkylation via trifluoroacetoanilide (**10**) as shown in Scheme 3. Acidic protection of the hydroxy group of **9** with dihydro-2*H*-pyran (DHP) followed by regioselective acylation with trifluoroacetoanhydride gave the trifluoroacetoanilide (**10**). *N*-Alkylation via a metal salt of the amide **10** with sodium hydride gave **11**. Cleavage of the trifluoroacetyl group under alkaline conditions and cleavage of the THP group with *p*-TsOH followed this. The hydroxy group was finally replaced by bromine using Ph_3P and CBr_4 to give the desired intermediates **2f,g**.

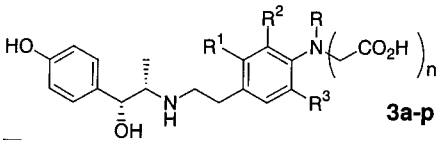
The synthesis of the isobutyrate intermediates **2o,p** was investigated in a manner similar to the preparation of 2-phenoxyisobutyrate such as fibrates,[21] which are agents used to treat hyperlipidemia (Scheme 4). Following protection of the hydroxyl group of the nitrobenzene derivatives **13a,b**, the *O*-protected compounds **14a,b** were obtained by reduction with zinc followed by treatment with 1,1,1-trichloro-2-methyl-2-propanol (Chloreton) to carry out the conversion to *N*-phenylaminoisobutyric acid.[22] Esterification with simultaneous cleav-

Scheme 2. Synthesis of Intermediates **2**[a]



[a] Reagents: (a) ethyl 2-bromoacetate, K_2CO_3; (b) NCS; (c) MeI, K_2CO_3; (d) alkyl halide, K_2CO_3; (e) Ph_3P, CBr_4; (f) NBS or NIS; (g) *tert*-butyl hypochlorite.

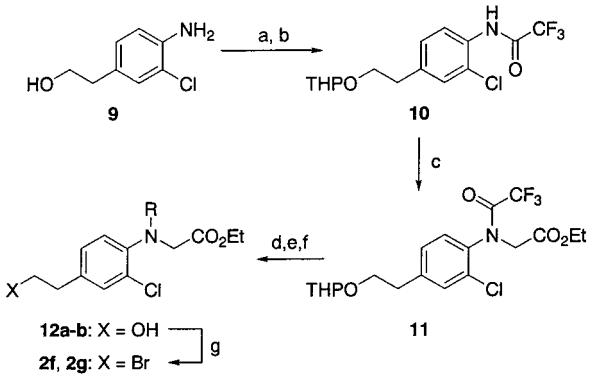
Table 1. Structures and Physical Data of the *N*-Phenylglycine Derivatives **3**



compd	R^1	R^2	R^3	R	*n*	formula	MW	anal.[a]
3a	H	H	H	H	1	$C_{19}H_{24}N_2O_4 \cdot 1.3H_2O$	367.8	C, H, N
3b	H	H	H	CH_3	1	$C_{20}H_{26}N_2O_4 \cdot 0.3H_2O$	363.8	C, H, N
3c	H	H	H		2	$C_{21}H_{26}N_2O_6 \cdot 0.2H_2O$	406.1	C, H, N
3d	H	H	H	Bn	1	$C_{26}H_{30}N_2O_4 \cdot 0.5H_2O$	443.5	C, H, N
3e	Cl	H	H	H	1	$C_{19}H_{23}ClN_2O_4 \cdot 1.5H_2O$	405.9	C, H, N
3f	H	H	Cl	H	1	$C_{19}H_{23}ClN_2O_4$	378.9	C, H, N
3g	H	H	Cl	CH_3	1	$C_{20}H_{25}ClN_2O_4 \cdot 2.4H_2O$	436.1	*b*, *d*
3h	H	Cl	Cl	H	1	$C_{19}H_{22}Cl_2N_2O_4 \cdot H_2O$	431.3	C, H, N
3i	H	Cl	Cl	CH_3	1	$C_{20}H_{24}Cl_2N_2O_4 \cdot 1.5H_2O$	454.4	C, H, N
3j	Cl	Cl	H	H	1	$C_{19}H_{22}Cl_2N_2O_4$	413.3	*c*, *d*
3k	Cl	H	Cl	H	1	$C_{19}H_{22}Cl_2N_2O_4 \cdot 0.5H_2O$	422.3	C, H, N
3l	H	Br	Br	H	1	$C_{19}H_{22}Br_2N_2O_4 \cdot 1.5H_2O$	529.2	C, H, N
3m	H	H	Br	H	1	$C_{19}H_{23}BrN_2O_4 \cdot 1.5H_2O$	450.3	C, H, N
3n	H	H	I	H	1	$C_{19}H_{23}IN_2O_4 \cdot H_2O$	488.3	C, H, N
3o	H	H	H	$C(CH_3)_2CO_2H$	0	$C_{21}H_{28}N_2O_4 \cdot H_2O$	390.5	C, H, N
3p	Cl	H	H	$C(CH_3)_2CO_2H$	0	$C_{21}H_{27}ClN_2O_4$	406.9	*c*, *d*

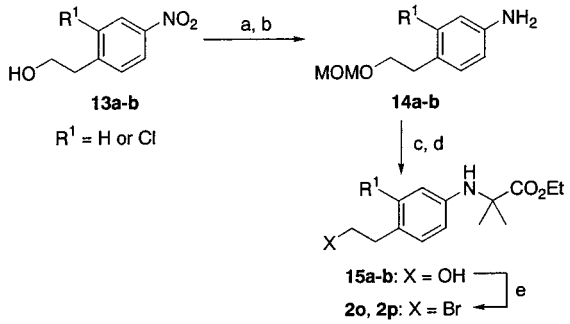
[a] Elemental analyses were within ±0.4% of the theoretical values. [b] Anal. C, N; H: found 6.31, calcd 6.89. [c] Elemental analysis not performed. [d] The chemical purity determined by HPLC was more than 96%.

Scheme 3. Synthesis of Intermediates **2**[a]



[a] Reagents: (a) TFAA, TEA; (b) DHP, PPTS; (c) ethyl 2-bromoacetate, NaH; (d) TsOH; (e) K_2CO_3; (f) MeI, K_2CO_3; (g) Ph_3P, CBr_4.

Scheme 4. Synthesis of Intermediates **2**[a]



[a] Reagents: (a) MOMCl, DIPEA; (b) 3% Pt-C, H_2; (c) chloretone, KOH; (d) $SOCl_2$, EtOH; (e) Ph_3P, CBr_4.

age of the protective group, and further bromination of **15a,b** as described above, gave the phenethyl bromide bearing the aminoisobutyrate moiety (**2o,p**).

Results and Discussion

β_3-AR agonists are reported to produce concentration-dependent relaxation of rat, canine, and human detrusor.[23] Moreover, we found that relaxation of the ferret detrusor is mediated via β_3-AR, as in the human detrusor.[14] Therefore, in this study, we used a ferret detrusor preparation to estimate the β_3-AR agonistic activity. The activities of β_1- and β_2-AR were evaluated using rat atria or uteri, respectively, according to the usual method. Table 2 summarizes the results of the in vitro assays of all of our compounds.

The glycine derivative **3a** of ritodrine possessed potent β_3-AR agonistic activity (EC_{50} = 24 nM) and selectivity over the β_1- and β_2-AR agonist activity (β_1/β_3 = 54-fold and β_2/β_3 = 27-fold). Introduction of a methyl group on the nitrogen of the glycine moiety of **3a** slightly decreased the efficacy for all β-ARs (**3b**) and also slightly altered the selectivity for β_3-AR (β_1/β_3 = 65-fold and β_2/β_3 = 31-fold), even though the β_3 potency was reduced. In contrast, the *N*-benzylglycine derivative **3d** had marked enhancement of all β-AR agonistic activity. Thus, **3d** had twice the β_3-AR agonistic activity of **3a** (EC_{50} = 13 nM) but had low selectivity over the β_1- and β_2-AR agonistic activity (β_1/β_3 = 12-fold and β_2/β_3 = 19-fold). However, replacement of the *N*-benzyl group with a carboxymethyl group (**3c**) significantly lowered all β-AR agonistic activity. The 2-aminoisobutyric acid derivative **3o**, with a geminal methyl group on the α carbon atom of the glycine moiety, had similar β_3-AR potency (23 nM) and higher selectivity (β_1/β_3 and β_2/β_3 = 478-fold) in comparison with **3a**. These initial studies suggested that substitution of methyl groups on the glycine moiety significantly increased the selectivity for β_3-AR, due to a decrease in the β_1- and β_2-AR agonistic activity, even when these compounds produced a modest agonistic effect on β_3-AR.

Next, we investigated the effect of halogen substitution on the phenyl ring for β_3-AR agonistic activity and selectivity. The substitution of halogens, electron-withdrawing groups, changed the potency and selectivity significantly. These changes were dependent upon the position of the halogen substituents in the phenyl ring, i.e., *ortho*- or *meta*-sites or both. The best rationale for the SAR for the halogen substituent position is found

Table 2. β-AR Agonistic Activity and Selectivity of the *N*-Phenylglycine Derivatives **3**

	β_1	β_2	β_3		β_3-AR selectivity[e]	
compd	$-\log EC_{20} \pm SE$ (EC_{20}: nM)[a]	$-\log IC_{50} \pm SE$ (IC_{50}: nM)[b]	$-\log EC_{50} \pm SE$ (EC_{50}: nM)[c]	IA[d]	β_1/β_3	β_2/β_3
3a	5.89 ± 0.21 (1300)	6.19 ± 0.14 (650)	7.62 ± 0.23 (24)	0.82	54	27
3b	5.47 ± 0.14 (3400)	5.80 ± 0.23 (1600)	7.28 ± 0.22 (52)	0.88	65	31
3c	4.57 ± 0.11 (27000)	6.55 ± 0.19 (280)	6.08 ± 0.12 (840)	0.79	32	0.33
3d	6.82 ± 0.06 (150)	6.60 ± 0.02 (250)	7.89 ± 0.25 (13)	0.92	12	19
3e	6.96 ± 0.06 (110)	7.21 ± 0.16 (62)	8.14 ± 0.20 (7.2)	0.97	15	8.6
3f	5.57 ± 0.05 (2700)	5.48 ± 0.13 (3300)	9.03 ± 0.13 (0.93)	0.88	2903	3548
3g	4.60 ± 0.19 (25000)	4 > (>100000)	7.96 ± 0.18 (11)	0.99	2273	>9091
3h	4.54 ± 0.13 (29000)	4.85± 0.20 (14000)	7.85 ± 0.10 (14)	0.85	2071	1000
3i	4 > (>100000)	4 > (>100000)	6.80 ± 0.23 (160)	0.84	>625	>625
3j	6.82 ± 0.05 (150)	5.77 ± 0.33 (1700)	6.34 ± 1.04 (460)	0.84	0.33	3.7
3k	5.89 ± 0.03 (1300)	5.89 ± 0.20 (1300)	7.21 ± 0.17 (62)	0.83	21	21
3l	4.72 ± 0.06 (19000)	5.59 ± 0.06 (2600)	7.09 ± 0.35 (81)	0.83	235	32
3m	5.10 ± 0.09 (8000)	5.85 ± 0.06 (1400)	6.24 ± 0.14 (570)	0.76	14	2.5
3n	5.27 ± 0.19 (5400)	5.66 ± 0.15 (2200)	6.09 ± 0.15 (820)	0.70	6.6	2.7
3o	4.96 ± 0.16 (11000)	4.96 ± 0.01 (11000)	7.64 ± 0.10 (23)	0.74	478	478
3p	6.59 ± 0.0.06 (260)	5.85 ± 0.20 (1400)	8.25 ± 0.13 (5.6)	0.95	46	250
ritodrine	6.80 ± 0.10 (160)	7.62 ± 0.07 (24)	6.72 ± 0.10 (190)	0.83	0.84	0.13
CL316243	4.77 ± 0.07 (17000)	5.01 ± 0.09 (9700)	7.60 ± 0.17 (25)	0.77	680	388
BRL37344	6.92 ± 0.04 (120)	8.04 ± 0.10 (9.1)	8.66 ± 0.08 (2.2)	0.96	55	4.14
isoproterenol	9.82 ± 0.08 (0.15)	10.0 ± 0.03 (0.1)	7.06 ± 0.11 (87)	0.99	0.002	0.001

[a] In parentheses is the EC_{20} value (nM), the mean concentration required to increase 20 beats/minute in the rat atria ($n \geq 3$). [b] In parentheses is the IC_{50} value (nM), the mean concentration required to produce 50% inhibition of uterine contraction in the rat uterus ($n \geq 3$). [c] In parentheses is the EC_{50} value (nM), the mean concentration required to produce 50% relaxation of detrusor before the addition of the compound in the ferret detrusor ($n \geq 3$). [d] Intrinsic activity (IA) given as a ratio of the maximum stimulation with forskolin (10^{-5} M). [e] The selectivity is the concentration ratio of β_3 (EC_{50}) to β_1 (EC_{20}) or β_2 (IC_{50}) for each drug.

in the chlorine-substituted compounds. *Ortho*-substitution of chlorine in **3a** (**3f**) enhanced the β_3-AR potency (EC_{50} = 0.93 nM) and selectivity (β_1/β_3 = 2900-fold and β_2/β_3 = 3540-fold). On the other hand, the *meta*-substituted compound **3e** was less selective for β_3-AR than the *ortho*-chloride **3f**, since *meta*-substitution enhanced the agonistic activities for all the β-ARs. Substitution of chlorine into the phenyl ring of **3a** at both *ortho*-positions (**3h**), i.e., 2,6-dichloro substitution, also increased the potency and selectivity, while **3h** had a 15-fold lower β_3-AR potency (EC_{50} = 14 nM) and lower selectivity over the β_1- and β_2-AR agonistic activity by 1.4- and 3.6-fold, respectively, than the monochloride **3f**. In contrast, the 2,5-dichloro substitution (**3k**) only slightly altered the β-AR agonistic activity of **3a**, presumably due to the counteracting effect of the chlorine at each position. In fact, the selectivity of **3k** was similar to that of **3a**. The 2,3-dichloro substitution (**3j**) unexpectedly increased the β_1-AR agonistic activity rather than that of β_2- or β_3-AR. The results of substituting chlorines in the phenyl ring indicated that to improve the potency and selectivity of a β_3-AR agonist requires mono- or disubstitution of a halogen at the *ortho*-position rather than at the *meta*-position.

The bromine- and iodine-substituted compounds **3l**−**n** were also β_3-AR agonists that reduced the β_1- and β_2-AR agonistic activities of **3a**. It should be noted that the dibromo-substituted compound **3l** retained a high selectivity (β_1/β_3 = 235-fold and β_2/β_3 = 32-fold) for β_3-AR. Thus, their profiles had a similar trend to those of the chlorine-substituted compounds. The rank order of β_3-AR agonistic potency of monohalogen-substituted compounds was chlorine **3f** (EC_{50} = 0.93 nM) > bromine **3m** (EC_{50} = 570 nM) > iodine **3n** (EC_{50} = 820 nM). This suggested that the larger the atomic nucleus of the halogen substituted in the phenyl ring of **3a**, the less potent the β_3-AR agonistic activity induced.

Based on this evidence, particularly the enhancement of the β_3-AR agonistic activity by appropriate chlorine substitution and of β_3-AR selectivity by methyl substitution on the glycine moiety, further optimization was carried out by introducing chlorine into the methyl-substituted glycine analogues (**3b**,**o**). Mono- and dichloro substituents at the *ortho*-positions (**3g**,**i**) altered the β_3-

Table 3. Comparing **3h** and Isoproterenol Application in the Rat

	in vitro		in vivo
compd	rat detrusor $-\log EC_{50} \pm SE$[a]	ferret detrusor $-\log EC_{50} \pm SE$[a]	rat (iv) ED_{50} (μg/kg)[b]
3h	6.72 ± 0.02 (190)	7.85 ± 0.10 (14)	48
isoproterenol	8.00 ± 0.01 (10)	7.06 ± 0.11 (87)	0.60

[a] See footnote *c* in Table 2. [b] ED_{50} is the mean dose required to lower the intrabladder pressure of an anesthetized rat by 50% over that before drug administration ($n \geq 3$). Since the ED_{50} value was read from a plot of the dose−response curve that is multiplicate, it is not meaningful to assign standard error to it.

AR agonist activity of **3b** 2.2- and 0.15-fold, respectively. Interestingly, both compounds showed weak potency for both β_1- and β_2-ARs. Therefore, the selectivity for β_3-AR was greatly improved by the chloro substituent as compared to **3b**. Second, monochloro substituents at the *meta*-position (**3p**) increased the agonistic activities of all the β-ARs (β_3: EC_{50} = 5.6 nM) as compared to **3o**, so that there was little improvement in selectivity for β_3-AR activity over β_1- and β_2-AR activity. Simple monochloro substitution at the *ortho*-position improved both β_3-AR activity and selectivity.

Next, we attempted to evaluate the effect of these compounds on intrabladder pressure in anesthetized rats. Compound **3h**, which had one of the best profiles overall, was selected for this examination, because the synthesis of **3h** was comparatively facile. Relaxation of the rat urinary bladder is mediated via both β_2- and β_3-ARs.[23] Therefore, we confirmed the potency of compound **3h** in isolated rat detrusor preparations before conducting in vivo experiments. As shown in Table 3 and Figure 2a, the EC_{50} of **3h** in rat detrusor was 190 nM, and isoproterenol exhibited 19 times more potent relaxing activity than **3h**. In vivo, **3h** sufficiently lowered the intrabladder pressure (ED_{50} = 48 μg/kg) and the potency of isoproterenol was 80 times higher than that of **3h**. On the other hand, **3h** had no effects on heart rate or blood pressure, while isoproterenol elicited a significant increase in heart rate and lowering of blood pressure, as shown in Figure 2b,c. These results were due to high selectivity for β_3-AR, indicating that **3h** was a potent, selective β_3-AR agonist that reduced the urinary bladder pressure without cardiovascular side effects, which are mediated by β_1- and β_2-AR.

In conclusion, a series of glycine-modified ritodrine was synthesized, and their β_3-AR activity was estimated in a novel functional assay using the ferret detrusor. Replacement of the phenolic hydroxy group on ritodrine with glycine enhanced both β_3-AR potency for ferret detrusor relaxation and selectivity over β_1- and β_2-AR. Additionally, mono- or disubstitution of chlorine at the *ortho*-position improved both the β_3-AR activity and selectivity. In vitro results demonstrated that *ortho*-chloro-substituted compounds with or without *N*-methyl substituents (**3f**−**i**) exhibited the best full agonistic activity and selectivity for β_3-AR. The in vivo result for the glycine analogue **3h**, a representative of this series, reflected the in vitro result; namely, **3h** lowered the intrabladder pressure of anesthetized rats without cardiovascular side effects due to β_1- or β_2-AR mediation. Moreover, **3h** showed nearly full agonistic activity to Chinese hamster ovary cells expressing the cloned human β_3-AR (data not shown). The discovery of these

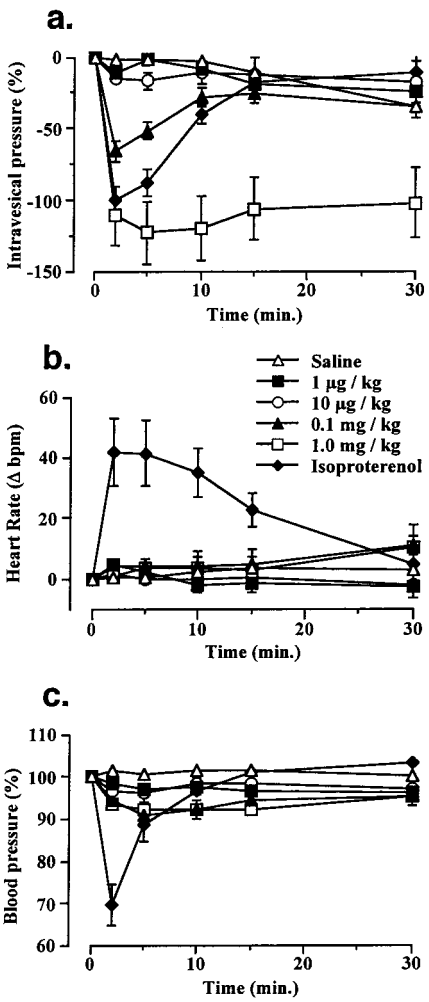


Figure 2. Time course of intravesical pressure (a), heart rate (b), and blood pressure (c) in anesthetized rats after administration of iv saline (1 mL/kg), isoproterenol (10 μg/g), and **3h** (1 μg, 10 μg, 0.1 mg, 1.0 mg) (n = 3). Intravesical pressure (a) is expressed as a percentage of the maximal relaxation with isoproterenol at 2 min. Heart rate (b) is expressed as the difference from the value before drug administration. Blood pressure (c) is expressed as a percentage of the value before drug administration.

compounds (**3**), which are novel selective β_3-AR agonists, is expected to provide impetus for developing a new approach in the field of urology. Encouraged by these findings, we are continuing to examine the toxicology, pharmacodynamics, and pharmacokinetics of these compounds in several species in the hope of finding useful drugs for the treatment of frequent urination and urinary incontinence.

Experimental Section

General Methods. Melting points were taken on a Yanaco MP-3S Micro melting point apparatus and are uncorrected. Infrared spectra were measured on a Nicolet 510 FT-IR spectrophotometer and are reported in reciprocal centimeters. Proton NMR spectra were recorded at 400 or 500 MHz with a Bruker AMX 400 or DRX 500 instrument, and chemical shifts are reported in parts per million (δ) downfield from tetramethylsilane as the internal standard. The peak patterns are shown as the following abbreviations: br = broad, d = doublet, m = multiplet, s = singlet, t = triplet, q = quartet. The mass spectra (MS) and high-resolution mass spectra (HRMS) were carried out with a JEOL JMS-SX102A mass spectrometer with capabilities for fast atom bombardment. Elemental analyses

were performed by the Yanaco CHN corder MT-5 analyzer. The analytical results obtained were within ±0.4% of the theoretical values, unless otherwise stated. Silica gel 60 F_{254} precoated plates on glass from Merck KGaA or aminopropyl silica gel (APS) precoated NH TLC from Fuji Silysia Chemical Ltd. was used for thin-layer chromatography (TLC). Flash or medium-pressure liquid column chromatography (MPLC) was performed on silica gel BW-350 (particle size 25−44 μm) from Fuji Silysia Chemical Ltd. or APS Daisogel IR-60 (particle size 25−40 μm) from Daiso Co., Ltd. Analytical HPLC was run on a Shimadzu LC-VP instrument, equipped with an Inertsil ODS-3, 5 μm, 4.6 × 250 mm column under two elution conditions: (a) a gradient (MeCN in 0.02 M H_3PO_4): 10 min (17.0%), 10−30 min (17−50%), 30−40 min (50%); (b) an isocratic condition in 0.02 M H_3PO_4 containing a specified concentration of MeCN, flow rate = 1.0 mL/min, λ = 225 nm. The column temperature was maintained at 40 °C. All of the reported purities were over 96%, unless otherwise stated. All reagents and solvents were commercially available unless otherwise indicated. Yields were not optimized.

4-(2-Bromoethyl)aniline Hydrobromide (4a). 4-Aminophenethyl alcohol (**4b**) (25 g, 0.18 mol) was dissolved in 48% HBr (250 mL), and the mixture was heated under reflux for 4 h with stirring. After cooling, collection of the resulting precipitates by filtration gave 30.3 g (59%) of **4a** as a white solid: mp 204−206 °C; IR (KBr) 2849, 2564, 1916, 1614, 1570, 1503; ^{1}H NMR (DMSO-d_6) δ 3.15 (2H, t, J = 7.0 Hz), 3.74 (2H, t, J = 7.0 Hz), 7.25 (2H, d, J = 8.0 Hz), 7.38 (2H, d, J = 8.0 Hz), 9.70 (2H, br).

4′-Amino-2′-chlorophenethyl Alcohol (4c). To a solution of 2-chloro-4-nitrophenethyl alcohol[24] (770 mg, 3.82 mmol) in HOAc (40 mL) was added concentrated HCl (0.67 mL, 8.0 mmol) at room temperature. While the mixture was vigorously stirred, zinc powder (2.62 g, 40.0 mmol) was added. The resulting suspension was stirred for 5min and diluted with water (40 mL) followed by filtration to remove the zinc powder. The filtrate was concentrated in vacuo and the residue was basified with saturated aqueous $NaHCO_3$, and extracted with EtOAc. The extract was washed with brine, dried over anhydrous $MgSO_4$, and concentrated in vacuo to give 530 mg (81%) of **4c** as a light brown oil: IR (neat) 3343, 2953, 2884, 1724, 1620, 1501; ^{1}H NMR ($CDCl_3$) δ 1.36 (1H, br), 2.90 (2H, t, J = 6.7 Hz), 3.64 (2H, br), 3.82 (2H, t, J = 6.7 Hz), 6.53 (1H, dd, J = 8.1, 2.5 Hz), 6.71 (1H, d, J = 2.5 Hz), 7.03 (1H, d, J = 8.1 Hz).

Ethyl *N*-[4-(2-Bromoethyl)phenyl]aminoacetate (2a). To a solution of **4a** (9.15 g, 32.6 mmol) in DMF (65 mL) were added K_2CO_3 (4.95 g, 35.8 mmol) and ethyl bromoacetate (3.97 mL, 35.8 mmol), and the mixture was stirred for 36 h at room temperature. The reaction mixture was poured into ice water, and collection of the resulting precipitates by filtration gave 8.39 g (90%) of **2a** as a yellow solid: mp 66−68 °C; IR (KBr) 3384, 2983, 1727, 1611, 1521; ^{1}H NMR ($CDCl_3$) δ 1.29 (3H, t, J = 7.1 Hz), 3.04 (2H, t, J = 7.8 Hz), 3.49 (2H, t, J = 7.8 Hz), 3.88 (2H, s), 4.15−4.35 (3H, m), 6.56 (2H, d, J = 8.5 Hz), 7.02 (2H, d, J = 8.5 Hz).

Ethyl *N*-[4-(2-Hydroxyethyl)phenyl]aminoacetate (5b). To a solution of **4b** (25 g, 0.18 mol) in DMF (500 mL) were added K_2CO_3 (30.0 g, 0.22 mol) and ethyl bromoacetate (24.0 mL, 0.22 mol), and the mixture was stirred for 16 h at room temperature. Et_2NH (38.0 mL, 0.37 mol) was added to the reaction mixture, and the resulting mixture was stirred for 1 h. The insoluble material was filtered off and the filtrate was concentrated in vacuo. The residue was diluted with diethyl ether, and the resulting insoluble material was filtered off. The filtrate was washed with 10% aqueous citric acid solution, saturated aqueous $NaHCO_3$ and brine subsequently, and dried over anhydrous $MgSO_4$. Removal of the solvent in vacuo gave 35.5 g (87%) of **5b** as an off-white solid: mp 54−55 °C; IR (KBr) 3413, 3378, 2953, 1730, 1617, 1521; ^{1}H NMR ($CDCl_3$) δ 1.29 (3H, t, J = 7.2 Hz), 1.42 (1H, t, J = 6.3 Hz), 2.76 (2H, t, J = 6.3 Hz), 2.76 (2H, q, J = 6.3 Hz), 3.88 (2H, s), 4.22 (1H, br), 4.24 (2H, q, J = 7.2 Hz), 6.58 (2H, d, J = 8.4 Hz), 7.05 (2H, d, J = 8.4 Hz).

Ethyl *N*-[3-Chloro-4-(2-hydroxyethyl)phenyl]aminoacetate (5c). The title compound was prepared from **4c** by similar method described in **5b** and purification by MPLC on silica gel (eluent: hexane/EtOAc = 2/1) to give **5c** as a white solid: mp 69−71 °C; IR (KBr) 3384, 3331, 2936, 1730, 1614, 1515; ^{1}H NMR ($CDCl_3$) δ 1.30 (3H, t, J = 7.2 Hz), 1.40 (1H, t, J = 6.3 Hz), 2.90 (2H, t, J = 6.3 Hz), 3.81 (2H, q, J = 6.3 Hz), 3.86 (2H, d, J = 5.2 Hz), 4.25 (2H, q, J = 7.2 Hz), 4.28−4.32 (1H, m), 6.47 (1H, dd, J = 8.1, 2.5 Hz), 6.62 (1H, d, J = 2.5 Hz), 7.06 (1H, d, J = 8.1 Hz).

Ethyl *N*-[2,6-Dibromo-4-(2-bromoethyl)phenyl]aminoacetate (2l). To a stirred solution of **2a** (515 mg, 1.80 mmol) in MeCN (3.6 mL) were added concentrated HCl (150 μL) and *N*-bromosuccinimide (641 mg, 3.60 mmol) under ice-cooling, and the mixture was stirred for 1 h. EtOH (3.6 mL) was added to the reaction mixture, and the mixture was stirred for 30 min under ice-cooling. 0.4 M aqueous $Na_2S_2O_3$ (9.0 mL) was added, and the mixture was stirred for 1 h under ice-cooling. The reaction mixture was diluted with water and extracted with EtOAc. The extract was washed with brine and dried over anhydrous $MgSO_4$, and the solvent was removed in vacuo. Purification of the residue by MPLC on silica gel (eluent: hexane/EtOAc = 6/1) gave 799 mg (100%) of **2l** as a pale yellow oil: ^{1}H NMR ($CDCl_3$) δ 1.29 (3H, t, J = 7.1 Hz), 3.03 (2H, t, J = 7.4 Hz), 3.50 (2H, t, J = 7.4 Hz), 4.12 (2H, d, J = 5.7 Hz), 4.24 (2H, q, J = 7.1 Hz), 4.78 (1H, t, J = 5.7 Hz), 7.32 (2H, s).

Ethyl *N*-[2-Bromo-4-(2-bromoethyl)phenyl]aminoacetate (2m). The title compound was prepared by similar method described in **2l** using a molar equivalent of *N*-bromosuccinimide to **2a** and purification by MPLC on silica gel (eluent: hexane/EtOAc = 6/1) as a pale yellow oil: ^{1}H NMR ($CDCl_3$) δ 1.30 (3H, t, J = 7.1 Hz), 3.03 (2H, t, J = 7.6 Hz), 3.49 (2H, t, J = 7.6 Hz), 3.93 (2H, s), 4.26 (2H, q, J = 7.1 Hz), 4.90 (1H, br s), 6.46 (1H, d, J = 8.2 Hz), 7.03 (1H, dd, J = 2.0, 8.2 Hz), 7.31 (1H, d, J = 2.0 Hz).

Ethyl *N*-[4-(2-Bromoethyl)-2-iodophenyl]aminoacetate (2n). The title compound was prepared by similar method described in **2l** using a molar equivalent of *N*-iodosuccinimide to **2a** and purification by MPLC on silica gel (eluent: hexane/EtOAc = 9/1) as a white solid: mp 70−72 °C; ^{1}H NMR ($CDCl_3$) δ 1.31 (3H, t, J = 7.1 Hz), 3.01 (2H, t, J = 7.6 Hz), 3.48 (2H, t, J = 7.6 Hz), 3.92 (2H, d, J = 5.3 Hz), 4.26 (2H, q, J = 7.1 Hz), 4.75−4.85 (1H, m), 6.39 (1H, d, J = 8.2 Hz), 7.06 (1H, dd, J = 1.9, 8.2 Hz), 7.54 (1H, d, J = 1.9 Hz).

Ethyl *N*-[2,6-Dichloro-4-(2-hydroxyethyl)phenyl]aminoacetate (6). The title compound was prepared from **5b** instead of **2a** by similar method described in **2l** using *tert*-butyl hypochlorite instead of *N*-bromosuccinimide and purification by MPLC on silica gel (eluent: hexane/EtOAc = 3/1) as an oil: IR (neat) 3356, 2937, 1736, 1562; ^{1}H NMR ($CDCl_3$) δ 1.28 (3H, t, J = 7.1 Hz), 2.74 (2H, t, J = 6.4 Hz), 3.82 (2H, t, J = 6.4 Hz), 4.14 (2H, s), 4.23 (2H, q, J = 7.1 Hz), 7.12 (2H, s).

Ethyl *N*-Ethyloxycarbonylmethyl-*N*-[4-(2-hydroxyethyl)phenyl]aminoacetate (8c). A mixture of **4b** (1.37 g, 10.0 mmol), ethyl bromoacetate (11.1 mL, 0.10 mol), and K_2CO_3 (3.46 g, 25.0 mmol) in DMF (30 mL) was stirred overnight at room temperature and then for 1 h at 70 °C. EtOH (30 mL) was added and the mixture was stirred for 1 h at 70 °C followed by cooling to room temperature. After addition of diethylamine (20.7 mL, 0.20 mol) at 10 °C and stirring for 1 h, the reaction mixture was concentrated to remove EtOH and diethylamine, diluted with diethyl ether, and washed with water. The organic layer was washed with 1 N HCl, saturated aqueous $NaHCO_3$, and brine successively, and dried over anhydrous $MgSO_4$. The solvent was removed in vacuo to give 3.05 g (99%) of **8c** as an oil: IR (neat) 3448, 2983, 2936, 1744, 1617, 1524; ^{1}H NMR ($CDCl_3$) δ 1.28 (6H, t, J = 7.2 Hz), 1.35 (1H, t, J = 6.4 Hz), 2.76 (2H, t, J = 6.4 Hz), 3.79 (2H, q, J = 6.4 Hz), 4.12 (4H, s), 4.21 (4H, q, J = 7.2 Hz), 6.58 (2H, d, J = 8.9 Hz), 7.08 (2H, d, J = 8.9 Hz).

2′-Chloro-4′-[2-((*RS*)-tetrahydropyran-2-yloxy)ethyl]-2,2,2-trifluoroacetanilide (10). To a stirred solution of **9** (950 mg, 5.54 mmol) and Et_3N (2.3 mL, 16.5 mmol) in THF (5.5

mL) was added a solution of trifluoroacetic anhydride (1.6 mL, 11.3 mmol) in THF (2.0 mL) under ice-cooling, and the mixture was stirred for 10 min. To the reaction mixture was added MeOH (2.0 mL), and the resulting mixture was stirred for 5 min. 1 N HCl (10 mL) was added to the stirred mixture, and the resulting mixture was extracted with EtOAc. The extract was washed with a saturated aqueous $NaHCO_3$ and brine subsequently, and dried over anhydrous $MgSO_4$. Removal of the solvent in vacuo gave 1.4 g (94%) of 2′-chloro-4′-(2-hydroxyethyl)-2,2,2-trifluoroacetanilide as a white solid: mp 74−76 °C; IR (KBr) 3436, 3279, 2948, 1724, 1611, 1553; ^{1}H NMR ($CDCl_3$) δ 2.86 (2H, t, J = 6.4 Hz), 3.87 (2H, t, J = 6.4 Hz), 7.22 (1H, dd, J = 8.4, 2.0 Hz), 7.35 (1H, d, J = 2.0 Hz), 8.24 (1H, d, J = 8.4 Hz), 8.36 (1H, br).

To a solution of 2′-chloro-4′-(2-hydroxyethyl)-2,2,2-trifluoroacetanilide (1.4 g, 5.23 mmol) and 3,4-dihydro-2*H*-pyran (1.4 mL, 15.4 mmol) in CH_2Cl_2 (15 mL) was added pyridinium *p*-toluenesulfonate (14 mg, 0.06 mmol), and the mixture was heated under reflux for 1 h with stirring. After concentration of the reaction mixture in vacuo, the residue was dissolved in EtOAc, washed with a saturated aqueous $NaHCO_3$ and brine. The organic layer was dried over anhydrous $MgSO_4$, and the solvent was removed in vacuo. Purification of the residue by MPLC on silica gel (eluent: hexane/diethyl ether = 3/1) gave 1.4 g (76%) of **10** as a brown oil: IR (neat) 3407, 3262, 2948, 1736, 1588, 1535; ^{1}H NMR ($CDCl_3$) δ 1.40−1.90 (6H, m), 2.88 (2H, t, J = 6.7 Hz), 3.40−3.50 (1H, m), 3.60 (1H, dt, J = 9.8, 6.7 Hz), 3.65−3.80 (1H, m), 3.94 (1H, dt, J = 9.8, 6.7 Hz), 4.55−4.60 (1H, m), 7.23 (1H, dd, J = 8.4, 1.9 Hz), 7.36 (1H, d, J = 1.9 Hz), 8.22 (1H, d, J = 8.4 Hz), 8.36 (1H, br).

Ethyl *N*-[2-Chloro-4-[2-((*RS*)-tetrahydropyran-2-yloxy)ethyl]phenyl]-*N*-trifluoroacetylaminoacetate (11). To a stirred solution of **10** (352 mg, 1.0 mmol) in DMF (3 mL) was added 60% NaH in mineral oil (48 mg, 2.0 mmol) under ice-cooling, and the mixture was stirred for 30 min at room temperature. Ethyl bromoacetate (133 μL, 1.2 mmol) was added to the reaction mixture, and the mixture was stirred for 16 h. The reaction mixture was diluted with EtOAc, washed with brine, and dried over anhydrous $MgSO_4$. After the solvent was removed in vacuo, purification of the residue by MPLC on silica gel (eluent: hexane/diethyl ether = 2/1) gave 250 mg (57%) of **11** as a clear oil: IR (neat) 2948, 1750, 1710, 1498; ^{1}H NMR ($CDCl_3$) δ 1.28 (3H, t, J = 7.1 Hz), 1.40−1.85 (6H, m), 2.91 (2H, t, J = 6.6 Hz), 3.40−3.50 (1H, m), 3.55−3.70 (2H, m), 3.72 (1H, d, J = 17.2 Hz), 3.90−4.00 (1H, m), 4.15−4.30 (2H, m), 4.55−4.65 (1H, m), 4.98 (1H, d, J = 17.2 Hz), 7.15−7.25 (1H, m), 7.35−7.45 (1H, m), 7.56 (1H, d, J = 8.1 Hz).

Ethyl *N*-[2-Chloro-4-(2-hydroxyethyl)phenyl]aminoacetate (12a; R = H). A solution of **11** (830 mg, 1.90 mmol) and *p*-TsOH monohydrate (80 mg, 0.42 mmol) in EtOH (9.0 mL) was stirred for 2 h at 40 °C. K_2CO_3 (314 mg, 2.27 mmol) was added, and the mixture was heated under reflux for 5 h with stirring. The insoluble material was filtered off, the filtrate was concentrated in vacuo, and the residue was dissolved in EtOAc. The solution was washed with saturated aqueous $NaHCO_3$ and brine, and dried over anhydrous $MgSO_4$, and the solvent was removed in vacuo. Purification of the residue by MPLC on silica gel (eluent: hexane/EtOAc = 2/1) gave 315 mg (65%) of **12** as an oil: IR (neat) 3407, 2942, 1739, 1614, 1521; ^{1}H NMR ($CDCl_3$) δ 1.30 (3H, t, J = 7.1 Hz), 1.37 (1H, br s), 2.74 (2H, t, J = 6.5 Hz), 3.75−3.85 (2H, m), 3.93 (2H, d, J = 5.5 Hz), 4.26 (2H, q, J = 7.1 Hz), 4.80−4.90 (1H, m), 6.49 (1H, d, J = 8.2 Hz), 7.00 (1H, dd, J = 8.2, 2.0 Hz), 7.17 (1H, d, J = 2.0 Hz).

Ethyl *N*-[4-(2-Hydroxyethyl)phenyl]-*N*-methylaminoacetate (8b; R = Me). To a solution of **5b** (1.15 g, 5.15 mmol) in DMF (10 mL) were added K_2CO_3 (1.17 g, 8.47 mmol) and iodomethane (420 μL, 6.75 mmol), and the mixture was stirred for 9 h at room temperature. The reaction mixture was poured into water and extracted with EtOAc. The extract was washed with brine, dried over anhydrous $MgSO_4$, and concentrated in vacuo. Purification of the residue by flash column chromatography on silica gel (eluent: CH_2Cl_2/diethyl ether = 10/1) gave 820 mg (67%) of **8b** as a brown oil: IR (neat) 3395, 2936, 1742, 1617, 1521; ^{1}H NMR ($CDCl_3$) δ 1.25 (3H, t, J = 7.1 Hz), 1.51 (1H, br), 2.77 (2H, t, J = 6.5 Hz), 3.05 (3H, s), 3.80 (2H, t, J = 6.5 Hz), 4.04 (2H, s), 4.18 (2H, q, J = 7.1 Hz), 6.65 (2H, d, J = 8.8 Hz), 7.09 (2H, d, J = 8.8 Hz).

The following compounds (**7**, **8d**, and **12b**) were prepared from the corresponding compounds (**6**, **5b**, and **12**) and an alkyl halide such as iodomethane or benzyl bromide by a similar method as described above.

Ethyl *N*-[2,6-dichloro-4-(2-hydroxyethyl)phenyl]-*N*-methylaminoacetate (7): ^{1}H NMR ($CDCl_3$) δ 1.26 (3H, t, J = 7.1 Hz), 1.41 (1H, br), 2.70−2.80 (2H, m), 2.94 (3H, s), 3.80−3.90 (4H, m), 4.17 (2H, q, J = 7.1 Hz), 7.10−7.20 (2H, m).

Ethyl *N*-benzyl-*N*-[4-(2-hydroxyethyl)phenyl]aminoacetate (8d; R = Bn): IR (neat) 3404, 2935, 1743, 1616, 1522; ^{1}H NMR ($CDCl_3$) δ 1.26 (3H, t, J = 7.1 Hz), 1.36 (1H, t, J = 6.3 Hz), 2.76 (2H, t, J = 6.3 Hz), 3.79 (2H, q, J = 6.3 Hz), 4.06 (2H, s), 4.20 (2H, q, J = 7.1 Hz), 4.63 (2H, s), 6.64 (2H, d, J = 8.8 Hz), 7.06 (2H, d, J = 8.8 Hz), 7.20−7.40 (5H, m).

Ethyl *N*-[2-chloro-4-(2-hydroxyethyl)phenyl]-*N*-methylaminoacetate (12b; R = Me): IR (neat) 3413, 2942, 1742, 1501; ^{1}H NMR ($CDCl_3$) δ 1.24 (3H, t, J = 7.1 Hz), 1.35−1.50 (1H, m), 2.78 (2H, t, J = 6.5 Hz), 2.97 (3H, s), 3.75−3.90 (2H, m), 3.96 (2H, s), 4.15 (2H, q, J = 7.1 Hz), 7.07 (1H, dd, J = 8.2, 1.9 Hz), 7.14 (1H, d, J = 8.2 Hz), 7.21 (1H, d, J = 1.9 Hz).

4-[2-(Methoxymethoxy)ethyl]aniline (14a; R^1 = H). To a solution of 4-nitrophenethyl alcohol (5.0 g, 29.9 mmol) and *N*,*N*-diisopropylethylamine (DIPEA) (6.3 mL, 35.9 mmol) in THF (100 mL) was added chloromethyl methyl ether (MOMCl) (2.7 mL, 35.9 mmol) at room temperature and the mixture was stirred for 5 h. DIPEA (6.3 mL) and MOMCl (2.7 mL) were added to the reaction mixture, and the mixture was stirred overnight. Moreover, DIPEA (6.3 mL) and MOMCl (2.7 mL) were added to the reaction mixture. After stirred for 6 h, 28% aqueous ammonia solution (7.3 mL, 0.12 mol) was added at room temperature and the mixture was stirred for 3 h. The resulting mixture was diluted with hexane and vigorously stirred for 15 min. The organic layer was separated and washed successively with water, 10% aqueous citric acid solution, saturated aqueous $NaHCO_3$, and brine. The hexane layer was dried over anhydrous $MgSO_4$ and concentrated in vacuo to give 6.0 g (96%) of methoxymethyl 4′-nitrophenethyl ether as a light yellow oil: IR (neat) 3076, 2948, 2878, 2459, 1927, 1602, 1518; ^{1}H NMR ($CDCl_3$) δ 3.01 (2H, t, J = 6.5 Hz), 3.27 (3H, s), 3.80 (2H, t, J = 6.5 Hz), 4.60 (2H, s), 7.41 (2H, d, J = 8.7 Hz), 8.16 (2H, d, J = 8.7 Hz).

3% Platinum on carbon, sulfided and moistened with water (63%), was suspended in the solution of methoxymethyl 4′-nitrophenethyl ether (1.0 g, 4.74 mmol) in MeOH (10 mL) and stirred for 18 h at room temperature under hydrogen. The reaction mixture was filtrated to remove the catalyst and concentrated in vacuo to give 850 mg (99%) of **14a** as a colorless oil: IR (neat) 3444, 3363, 2935, 2885, 1628, 1520; ^{1}H NMR ($CDCl_3$) δ 2.80 (2H, t, J = 7.2 Hz), 3.31 (3H, s), 3.56 (2H, br), 3.70 (2H, t, J = 7.2 Hz), 4.61 (2H, s), 6.63 (2H, d, J = 8.2 Hz), 7.19 (2H, d, J = 8.2 Hz).

3-Chloro-4-[2-(methoxymethoxy)ethyl]aniline (14b; R^1 = Cl). The title compound as an oil was prepared from 2′-chloro-4′-nitrophenethyl alcohol[21] by similar method described in **14a**: ^{1}H NMR ($CDCl_3$) δ 2.92 (2H, t, J = 7.1 Hz), 3.31 (3H, s), 3.71 (2H, t, J = 7.1 Hz), 3.70−3.90 (2H, m), 4.61 (2H, s), 6.54 (1H, dd, J = 8.2, 2.4 Hz), 6.72 (1H, d, J = 2.4 Hz), 7.04 (1H, d, J = 8.2 Hz).

Ethyl 2-[[4-(2-Hydroxyethyl)phenyl]amino]-2-methylpropionate (15a; R^1 = H). To the mixture of **14a** (725 mg, 4.0 mmol) and 1,1,1-trichloro-2-methyl-2-propanol 0.5 hydrate (1.47 g, 4.0 mmol) in acetone (12 mL) was added potassium hydroxide (6.40 g, 114 mmol) at 0 °C, and the mixture was stirred for 1 h at 0 °C and overnight at room temperature. The reaction mixture was concentrated in vacuo, diluted with water, and washed with diethyl ether. Ammonium chloride (3.57 g, 66.7 mmol) was dissolved in the aqueous solution, and the mixture was acidified with 10% aqueous citric acid solution and extracted with EtOAc. The organic layer was washed with brine, dried over anhydrous $MgSO_4$, and concentrated in vacuo.

The residue was dissolved in EtOH (40 mL), and thionyl chloride (875 μL, 12.0 mmol) was carefully added dropwise to the solution at 0 °C. The mixture was heated under reflux for 5 h and concentrated to remove the solvent. The residual oil was dissolved in EtOAc, basified with saturated aqueous $NaHCO_3$, and washed with brine. The organic layer was dried over anhydrous $MgSO_4$ and concentrated in vacuo. The residue was purified by MPLC on silica gel (eluent: hexane/EtOAc = 1/1) to give 155 mg (15.4%) of **15a** as a dark brown oil: IR (neat) 3419, 2988, 2936, 1733, 1620, 1521; ^{1}H NMR ($CDCl_3$) δ 1.19 (3H, t, J = 7.2 Hz), 1.34 (1H, br), 1.54 (6H, s), 2.75 (2H, t, J = 6.7 Hz), 3.76−3.82 (2H, m), 4.00 (1H, br), 4.17 (2H, q, J = 7.2 Hz), 6.56 (2H, d, J = 8.6 Hz), 7.00 (2H, d, J = 8.6 Hz).

Ethyl 2-[[3-Chloro-4-(2-hydroxyethyl)phenyl]amino]-2-methylpropionate (15b). The title compound as an oil was prepared from **14b** by similar method described in **15a**: ^{1}H NMR ($CDCl_3$) δ 1.21 (3H, t, J = 7.1 Hz), 1.54 (6H, s), 2.89 (2H, t, J = 6.7 Hz), 3.81 (2H, t, J = 6.7 Hz), 4.19 (2H, q, J = 7.1 Hz), 6.43 (1H, dd, J = 8.3, 2.5 Hz), 6.61 (1H, d, J = 2.5 Hz), 7.01 (1H, d, J = 8.3 Hz).

Ethyl *N*-[4-(2-Bromoethyl)-2,6-dichlorophenyl]aminoacetate (2h). To a stirred solution of **6** (650 mg, 2.23 mmol) and triphenylphosphine (700 mg, 2.67 mmol) in CH_2Cl_2 (10 mL) was added carbon tetrabromide (886 mg, 2.67 mmol) under ice-cooling, and the mixture was stirred for 1 h. Simply purification of the reaction mixture by flash column chromatography on silica gel (eluent: hexane/EtOAc = 3/1) and further purification of the fraction by MPLC on silica gel (eluent: hexane/CH_2Cl_2 = 1/1) gave 708 mg (89%) of **2h** as an oil: ^{1}H NMR ($CDCl_3$) δ 1.28 (3H, t, J = 7.2 Hz), 3.03 (2H, t, J = 7.4 Hz), 3.50 (2H, t, J = 7.4 Hz), 4.16 (2H, d, J = 5.7 Hz), 4.23 (2H, q, J = 7.2 Hz), 4.80−4.90 (1H, m), 7.10 (2H, s).

The following phenethyl bromide analogues were prepared by a similar method as described here using the corresponding phenethyl alcohol derivatives. All of the compounds were obtained as an oil except **2e**.

Ethyl *N*-[4-(2-bromoethyl)phenyl]-*N*-methylaminoacetate (2b): ^{1}H NMR ($CDCl_3$) δ 1.24 (3H, t, J = 7.1 Hz), 3.05 (3H, s), 3.05 (2H, t, J = 7.9 Hz), 3.50 (2H, t, J = 7.9 Hz), 4.04 (2H, s), 4.17 (2H, q, J = 7.1 Hz), 6.64 (2H, d, J = 8.8 Hz), 7.07 (2H, d, J = 8.8 Hz).

Ethyl *N*-[4-(2-bromoethyl)phenyl]-*N*-ethyloxycarbonylmethylaminoacetate (2c): ^{1}H NMR ($CDCl_3$) δ 1.27 (3H, t, J = 7.1 Hz), 3.05 (2H, t, J = 7.8 Hz), 3.48 (2H, t, J = 7.8 Hz), 4.12 (2H, s), 4.21 (2H, q, J = 7.1 Hz), 6.57 (2H, d, J = 8.6 Hz), 7.05 (2H, d, J = 8.6 Hz).

Ethyl *N*-benzyl-*N*-[4-(2-bromoethyl)phenyl]aminoacetate (2d): ^{1}H NMR ($CDCl_3$) δ 1.26 (3H, t, J = 7.1 Hz), 3.04 (2H, t, J = 7.8 Hz), 3.48 (2H, t, J = 7.8 Hz), 4.06 (2H, s), 4.20 (2H, q, J = 7.1 Hz), 4.63 (2H, s), 6.63 (2H, d, J = 8.5 Hz), 7.03 (2H, d, J = 8.5 Hz), 7.20−7.40 (5H, m).

Ethyl *N*-[4-(2-bromoethyl)-3-chlorophenyl]aminoacetate (2e): mp 85−86 °C; ^{1}H NMR ($CDCl_3$) δ 1.31 (3H, t, J = 7.1 Hz), 3.00−3.20 (2H, m), 3.45−3.7 (2H, m), 3.86 (2H, d, J = 5.3 Hz), 4.25 (2H, q, J = 7.1 Hz), 4.34 (1H, br), 6.45−6.50 (1H, m), 6.60 (1H, d, J = 1.6 Hz), 7.00−7.10 (1H, m).

Ethyl *N*-[4-(2-bromoethyl)-2-chlorophenyl]aminoacetate (2f): ^{1}H NMR ($CDCl_3$) δ 1.30 (3H, t, J = 7.1 Hz), 3.03 (2H, t, J = 7.6 Hz), 3.49 (2H, t, J = 7.6 Hz), 3.93 (2H, d, J = 5.5 Hz), 4.26 (2H, q, J = 7.1 Hz), 4.85−4.95 (1H, m), 6.49 (1H, d, J = 8.3 Hz), 6.98 (1H, dd, J = 8.3, 2.0 Hz), 7.14 (1H, d, J = 2.0 Hz).

Ethyl *N*-[4-(2-bromoethyl)-2-chlorophenyl]-*N*-methylaminoacetate (2g): ^{1}H NMR ($CDCl_3$) δ 1.24 (3H, t, J = 7.1 Hz), 2.97 (3H, s), 3.07 (2H, t, J = 7.6 Hz), 3.52 (2H, t, J = 7.6 Hz), 3.97 (2H, s), 4.15 (2H, q, J = 7.1 Hz), 7.04 (1H, dd, J = 8.2, 2.0 Hz), 7.13 (1H, d, J = 8.2 Hz), 7.18 (1H, d, J = 2.0 Hz).

Ethyl *N*-[4-(2-bromoethyl)-2,6-dichlorophenyl]-*N*-methylaminoacetate (2i): ^{1}H NMR ($CDCl_3$) δ 1.26 (3H, t, J = 7.1 Hz), 2.95 (3H, s), 3.06 (2H, t, J = 7.3 Hz), 3.52 (2H, t, J = 7.3 Hz), 3.88 (2H, s), 4.17 (2H, q, J = 7.1 Hz), 7.14 (2H, s).

Ethyl 2-[[4-(2-bromoethyl)phenyl]amino]-2-methylpropionate (2o): ^{1}H NMR ($CDCl_3$) δ 1.18 (3H, t, J = 7.1 Hz), 1.54 (6H, s), 3.03 (2H, t, J = 7.9 Hz), 3.48 (2H, t, J = 7.9 Hz), 4.04 (1H, br), 4.16 (2H, q, J = 7.1 Hz), 6.54 (2H, d, J = 8.6 Hz), 6.98 (2H, d, J = 8.6 Hz).

Ethyl 2-[[4-(2-bromoethyl)-3-chlorophenyl]amino]-2-methylpropionate (2p): ^{1}H NMR ($CDCl_3$) δ 1.20 (3H, t, J = 7.1 Hz), 1.55 (6H, s), 3.14 (2H, t, J = 7.8 Hz), 3.51 (2H, t, J = 7.8 Hz), 4.13 (1H, br s), 4.18 (2H, q, J = 7.1 Hz), 6.42 (1H, dd, J = 8.3, 2.5 Hz), 6.58 (1H, d, J = 2.5 Hz), 7.00 (1H, d, J = 8.3 Hz).

Ethyl *N*-[4-(2-Bromoethyl)-2,3-dichlorophenyl]aminoacetate (2j), Ethyl *N*-[4-(2-Bromoethyl)-2,5-dichlorophenyl]aminoacetate (2k). To a stirred solution of **2e** (79 mg, 0.25 mmol) in CH_2Cl_2 (2.0 mL) was added *tert*-butyl hypochlorite (31 μL, 0.27 mmol) under ice-cooling, and the mixture was stirred for 6 h at room temperature. The solvent was removed in vacuo, and purification of the residue by flash column chromatography on silica gel (eluent: hexane/EtOAc = 15/1) gave 31 mg (36%) of **2j** as a high-polar regioisomer and 34 mg (39%) of **2k** as a low-polar regioisomer. **2j**: ^{1}H NMR ($CDCl_3$) δ 1.30 (3H, t, J = 7.1 Hz), 3.05−3.25 (2H, m), 3.50−3.70 (2H, m), 3.94 (2H, d, J = 5.4 Hz), 4.26 (2H, q, J = 7.1 Hz), 5.04 (1H, br), 6.41 (1H, d, J = 8.4 Hz), 7.05 (1H, d, J = 8.4 Hz). **2k**: ^{1}H NMR ($CDCl_3$) δ 1.31 (3H, t, J = 7.1 Hz), 3.00−3.20 (2H, m), 3.45−3.70 (2H, m), 3.90 (2H, d, J = 5.3 Hz), 4.27 (2H, q, J = 7.1 Hz), 4.95 (1H, br), 6.53 (1H, s), 7.17 (1H, s).

***N*-[2,6-Dichloro-4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]aminoacetic Acid (3h).** To a solution of **1** (495 mg, 2.96 mmol) and **2h** (700 mg, 1.97 mmol) in DMF (6 mL) was added *N*,*N*-diisopropylethylamine (343 μL, 1.97 mmol), and the mixture was stirred for 7 h at 70 °C. The reaction mixture was concentrated in vacuo, and purification of the residue by MPLC on APS (eluent: CH_2Cl_2/EtOH = 20/1) gave 510 mg (64%) of ethyl *N*-[2,6-dichloro-4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]aminoacetate as a glassy oil: ^{1}H NMR ($CDCl_3$) δ 0.95 (3H, d, J = 6.4 Hz), 1.30 (3H, t, J = 7.2 Hz), 2.55−3.05 (5H, m), 4.16 (2H, d, J = 6.0 Hz), 4.25 (2H, q, J = 7.2 Hz), 4.51 (1H, d, J = 5.3 Hz), 4.78 (1H, t, J = 6.0 Hz), 6.75 (2H, d, J = 8.5 Hz), 7.00 (2H, s), 7.10 (2H, d, J = 8.5 Hz).

Ethyl *N*-[2,6-dichloro-4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]aminoacetate (500 mg, 1.13 mmol) was dissolved in 1 N NaOH (5.0 mL), and the solution was stirred for 1 h at room temperature. To the reaction mixture was added 1 N HCl (5.0 mL) under ice-cooling with stirring, and collection of the resulting precipitates by filtration gave 464 mg (99%) of **3h** as a solid: mp 175−180 °C dec; $[\alpha]_D^{30}$ = −3.5° (c = 1.00, HOAc); IR (KBr) 3358, 3017, 1618, 1575, 1516; ^{1}H NMR (DMSO-d_6) δ 0.90 (3H, d, J = 6.6 Hz), 2.65−2.80 (2H, m), 2.95−3.20 (3H, m), 3.76 (2H, s), 4.93 (1H, br s), 5.55 (1H, br), 6.72 (2H, d, J = 8.5 Hz), 7.13 (2H, d, J = 8.5 Hz), 7.16 (2H, s); MS m/z (relative intensity) 415 (0.75), 413 (M + H)$^+$, 289 (1.1), 246 (3.8). Anal. ($C_{19}H_{22}Cl_2N_2O_4 \cdot H_2O$) 431.32: C, H, N.

The following compounds were prepared from the corresponding phenethyl bromide derivatives (**2a**−**p**) by a similar method as described here.

***N*-[4-[2-[[(1*S*,2*R*)-2-Hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]aminoacetic acid (3a):** mp 186−188 °C dec; $[\alpha]_D^{25}$ = −6.7° (c = 0.75, HOAc); IR (KBr) 3411, 3017, 1615, 1572, 1523; ^{1}H NMR (DMSO-d_6) δ 0.87 (3H, d, J = 6.4 Hz), 2.50−3.20 (5H, m), 3.51 (2H, s), 4.86 (1H, br s), 6.45 (2H, d, J = 8.1 Hz), 6.70 (2H, d, J = 8.4 Hz), 6.83 (2H, d, J = 8.1 Hz), 7.11 (2H, d, J = 8.4 Hz); MS m/z (relative intensity) 345 (M + H)$^+$, 221 (0.18), 178 (0.51). Anal. ($C_{19}H_{24}N_2O_4 \cdot 1.3H_2O$) 367.8: C, H, N.

***N*-[4-[2-[[(1*S*,2*R*)-2-Hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]-*N*-methylaminoacetic acid (3b):** amorphous; $[\alpha]_D^{25}$ = −6.9° (c = 0.96, HOAc); IR (KBr) 3377, 2958, 1618, 1579, 1519; ^{1}H NMR (CD_3OD) δ 1.07 (3H, d, J = 6.7 Hz), 2.84 (2H, t, J = 8.1 Hz), 3.03 (3H, s), 3.10−3.25 (2H, m), 3.30−3.40 (1H, m), 3.86 (2H, s), 4.99 (1H, d, J = 3.4 Hz), 6.64 (2H, d, J = 8.8 Hz), 6.78 (2H, d, J = 8.6 Hz), 7.02 (2H, d, J = 8.8 Hz), 7.17 (2H, d, J = 8.6 Hz); MS m/z

(relative intensity) 359 (M + H)$^+$, 192 (0.76). Anal. ($C_{20}H_{26}N_2O_4$·0.3H_2O) 363.8: C, H, N.

***N*-Carboxymethyl-*N*-[4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]aminoacetic acid (3c):** mp 228−230 °C dec; $[\alpha]_D^{25} = -6.9°$ ($c = 0.96$, HOAc); IR (KBr) 3198, 2930, 1695, 1614, 1573, 1524; ^{1}H NMR (DMSO-d_6) δ 0.94 (3H, d, $J = 6.7$ Hz), 2.83 (2H, t, $J = 8.4$ Hz), 3.05−3.17 (2H, m), 3.30−3.37 (2H, m), 3.97 (4H, s), 5.01 (1H, br s), 5.96 (1H, br), 6.41 (2H, d, $J = 8.9$ Hz), 6.74 (2H, d, $J = 8.5$ Hz), 7.03 (2H, d, $J = 8.9$ Hz), 7.15 (2H, d, $J = 8.5$ Hz), 8.71 (2H, br), 9.39 (1H, br); MS *m*/*z* (relative intensity) 403 (M + H)$^+$, 236 (0.99), 176 (0.81). Anal. ($C_{21}H_{26}N_2O_6$·0.2H_2O) 406.1: C, H, N.

***N*-Benzyl-*N*-[4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]aminoacetic acid (3d):** amorphous; $[\alpha]_D^{30} = -6.8°$ ($c = 0.62$, DMSO); IR (KBr) 3429, 2513, 1616, 1576, 1520; ^{1}H NMR (DMSO-d_6) δ 0.90 (3H, d, $J = 6.6$ Hz), 2.60−2.80 (2H, m), 2.90−3.05 (2H, m), 3.15−3.25 (1H, m), 3.98 (2H, s), 4.59 (2H, s), 4.95−5.05 (1H, m), 6.51 (2H, d, $J = 8.6$ Hz), 6.72 (2H, d, $J = 8.5$ Hz), 6.88 (2H, d, $J = 8.6$ Hz), 7.13 (2H, d, $J = 8.5$ Hz), 7.15−7.35 (5H, m), 9.35 (2H, br); MS *m*/*z* (relative intensity) 435 (M + H)$^+$, 268 (0.61). Anal. ($C_{26}H_{30}N_2O_4$·0.5H_2O) 443.5: C, H, N.

***N*-[3-Chloro-4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]aminoacetic acid (3e):** amorphous; $[\alpha]_D^{25} = -1.6°$ ($c = 0.25$, 1 N HCl); IR (KBr) 3413, 1614, 1562, 1512; ^{1}H NMR (DMSO-d_6) δ 0.86 (3H, d, $J = 6.4$ Hz), 2.55−2.85 (5H, m), 3.35 (2H, s), 4.55 (1H, br s), 6.40 (1H, dd, $J = 8.3$, 2.2 Hz), 6.51 (1H, d, $J = 2.2$ Hz), 6.68 (2H, d, $J = 8.4$ Hz), 6.87 (1H, d, $J = 8.3$ Hz), 7.06 (2H, d, $J = 8.4$ Hz); MS *m*/*z* 379 (M + H)$^+$. Anal. ($C_{19}H_{23}ClN_2O_4$·1.5H_2O) 405.9: C, H, N.

***N*-[2-Chloro-4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]aminoacetic acid (3f):** mp 193−195 °C dec; $[\alpha]_D^{30} = -6.7°$ ($c = 1.00$, HOAc); IR (KBr) 3367, 3029, 1615, 2360,1618, 1575, 1519; ^{1}H NMR (DMSO-d_6) δ 0.90 (3H, d, $J = 6.4$ Hz), 2.60−2.80 (2H, m), 2.90−3.20 (3H, m), 3.52 (2H, s), 4.95 (1H, br s), 5.35 (1H, br), 6.46 (1H, d, $J = 8.4$ Hz), 6.72 (2H, d, $J = 8.6$ Hz), 6.91 (1H, dd, $J = 8.4$, 1.8 Hz), 7.05−7.20 (3H, m); MS *m*/*z* (relative intensity) 379 (M + H)$^+$, 212 (6.29). Anal. ($C_{19}H_{23}ClN_2O_4$) 378.9: C, H, N.

***N*-[2-Chloro-4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]-*N*-methylaminoacetic acid (3g):** amorphous; $[\alpha]_D^{25} = -4.5°$ ($c = 0.75$, HOAc); IR (KBr) 3209, 1597, 1516; ^{1}H NMR (DMSO-d_6) δ 0.87 (3H, d, $J = 6.4$ Hz), 2.65−2.90 (5H, m), 2.89 (3H, s), 3.60−3.70 (2H, m), 4.56 (1H, br), 6.65 (2H, d, $J = 8.5$ Hz), 6.81 (1H, dd, $J = 8.3$, 1.9 Hz), 6.95−7.05 (4H, m); MS *m*/*z* (relative intensity) 393 (M + H)$^+$; HPLC $t_R = 14.3$ min (gradient), 7.8 min (20% MeCN, isocratic). Anal. Calcd ($C_{20}H_{25}ClN_2O_4$·2.4H_2O) 436.1: C, 55.08; H, 6.89; N, 6.42. Found: C, 54.76; H, 6.31; N, 6.21.

***N*-[2,6-Dichloro-4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]-*N*-methylaminoacetic acid (3i):** mp 135−138 °C; $[\alpha]_D^{32} = -5.9°$ ($c = 0.54$, MeOH); IR (KBr) 3382, 3193, 1615, 1575, 1516; ^{1}H NMR (DMSO-d_6) δ 0.84 (3H, d, $J = 6.6$ Hz), 2.30−2.95 (8H, m), 3.76 (2H, s), 4.64 (1H, br s), 6.69 (2H, d, $J = 8.5$ Hz), 7.09 (2H, d, $J = 8.5$ Hz), 7.22 (2H, s); MS *m*/*z* (relative intensity) 429 (0.64), 427 (M + H)$^+$, 303 (0.31), 260 (0.23). Anal. ($C_{20}H_{24}Cl_2N_2O_4$·1.5H_2O) 454.4: C, H, N.

***N*-[2,3-Dichloro-4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]aminoacetic acid (3j):** mp 199−208 °C dec; $[\alpha]_D^{25} = -4.3°$ ($c = 0.47$, HOAc); IR (KBr) 3355, 3017, 1603, 1578, 1519; ^{1}H NMR (DMSO-d_6) δ 0.89 (3H, d, $J = 6.5$ Hz), 2.75−3.25 (5H, m), 3.53 (2H, s), 4.90 (1H, br s), 5.62 (1H, br s), 6.45 (1H, d, $J = 8.5$ Hz), 6.71 (2H, d, $J = 8.4$ Hz), 7.01 (1H, d, $J = 8.5$ Hz), 7.13 (2H, d, $J = 8.4$ Hz); MS *m*/*z* (relative intensity) 413 (M + H)$^+$, 247 (0.98), HRMS $C_{19}H_{23}Cl_2N_2O_4$; HPLC $t_R = 21.3$ min (gradient), 6.2 min (25% MeCN, isocratic).

***N*-[2,5-Dichloro-4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]aminoacetic acid (3k):** mp 191−195 °C dec; $[\alpha]_D^{25} = -3.4°$ ($c = 0.83$, HOAc); IR (KBr) 3360, 3023, 1608, 1573, 1512; ^{1}H NMR (DMSO-d_6) δ 0.89 (3H, d, $J = 6.5$ Hz), 2.65−3.25 (5H, m), 3.55 (2H, s), 4.93 (1H, br s), 5.58 (1H, br s), 6.53 (1H, s), 6.71 (2H, d, $J = 8.4$ Hz), 7.13 (2H, d, $J = 8.4$ Hz), 7.19 (1H, s), 9.30 (1H, br); MS *m*/*z* (relative intensity) 413 (M + H)$^+$, 246 (0.18). Anal. ($C_{19}H_{22}Cl_2N_2O_4$·0.5H_2O) 422.3: C, H, N.

***N*-[2,6-Dibromo-4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]aminoacetic acid (3l):** mp 188−192 °C dec; $[\alpha]_D^{29} = -3.4°$ ($c = 1.29$, HOAc); IR (KBr) 3327, 3188, 1593, 1516; ^{1}H NMR (DMSO-d_6) δ 0.89 (3H, d, $J = 6.6$ Hz), 2.65−3.20 (5H, m), 3.75 (2H, s), 4.75−4.90 (1H, m), 5.40 (1H, br), 6.71 (2H, d, $J = 8.3$ Hz), 7.11 (2H, d, $J = 8.3$ Hz), 7.38 (2H, s); MS *m*/*z* (relative intensity) 505 (0.45), 503 (M + H)$^+$, 501 (0.53). Anal. ($C_{19}H_{22}Br_2N_2O_4$·1.5H_2O) 529.23: C, H, N.

***N*-[2-Bromo-4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]aminoacetic acid (3m):** amorphous; $[\alpha]_D^{27} = -4.7°$ ($c = 0.51$, HOAc); IR (KBr) 3392, 3191, 1615, 1560, 1519; ^{1}H NMR (DMSO-d_6) δ 0.92 (3H, d, $J = 6.5$ Hz), 2.70−2.90 (2H, m), 3.00−3.40 (3H, m), 3.75 (2H, s), 4.93 (1H, br s), 5.36 (1H, br), 6.50 (1H, d, $J = 8.3$ Hz), 6.74 (2H, d, $J = 8.5$ Hz), 7.04 (1H, dd, $J = 8.3$, 1.8 Hz), 7.14 (2H, d, $J = 8.5$ Hz), 7.34 (1H, d, $J = 1.8$ Hz); MS *m*/*z* (relative intensity) 425 (0.99), 423 (M + H)$^+$, 258 (0.32), 256 (0.38). Anal. ($C_{19}H_{23}BrN_2O_4$·1.5H_2O) 450.3: C, H, N.

***N*-[4-[2-[[(1*S*,2*R*)-2-Hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]-2-iodophenyl]aminoacetic acid (3n):** mp 151−155 °C dec; $[\alpha]_D^{27} = -4.7°$ ($c = 0.51$, HOAc); IR (KBr) 3367, 3200, 1603, 1516; ^{1}H NMR (DMSO-d_6) δ 0.92 (3H, d, $J = 6.6$ Hz), 2.70−2.80 (2H, m), 3.00−3.25 (3H, m), 3.68 (2H, s), 4.93 (1H, br s), 5.15 (1H, br), 6.39 (1H, d, $J = 8.3$ Hz), 6.73 (2H, d, $J = 8.6$ Hz), 7.00−7.10 (1H, m), 7.14 (2H, d, $J = 8.6$ Hz), 7.53 (1H, d, $J = 1.8$ Hz); MS *m*/*z* (relative intensity) 471 (M + H)$^+$, 347 (0.18), 304 (0.51). Anal. ($C_{19}H_{23}IN_2O_4$·H_2O) 488.3: C, H, N.

2-[[4-[2-[[(1*S*,2*R*)-2-Hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]amino]-2-methylpropionic acid (3o): mp 193−200 °C dec; $[\alpha]_D^{29} = -8.2°$ ($c = 1.00$, HOAc); IR (KBr) 3392, 3147, 1618, 1557, 1519; ^{1}H NMR (DMSO-d_6 + D_2O) δ 0.91 (3H, d, $J = 6.6$ Hz), 1.37 (6H, s), 2.55−2.75 (2H, m), 2.85−3.00 (2H, m), 3.10−3.20 (1H, m), 4.92 (1H, d, $J = 2.2$ Hz), 6.47 (2H, d, $J = 8.5$ Hz), 6.70−6.80 (4H, m), 7.13 (2H, d, $J = 8.5$ Hz); MS *m*/*z* (relative intensity) 373 (M + H)$^+$, 249 (0.04), 206 (0.19). Anal. ($C_{21}H_{28}N_2O_4$·H_2O) 390.48: C, H, N.

2-[[3-Chloro-4-[2-[[(1*S*,2*R*)-2-hydroxy-2-(4-hydroxyphenyl)-1-methylethyl]amino]ethyl]phenyl]amino]-2-methylpropionic acid (3p): mp 170−173 °C dec; $[\alpha]_D^{28} = -4.0°$ ($c = 1.00$, 1 N HCl); IR (KBr) 3197, 1615, 1557, 1522; ^{1}H NMR (DMSO-d_6) δ 0.88 (3H, d, $J = 6.6$ Hz), 1.37 (6H, s), 2.60−2.90 (4H, m), 3.00−3.10 (1H, m), 4.83 (1H, br s), 6.40 (1H, dd, $J = 8.3$, 2.4 Hz), 6.54 (1H, d, $J = 2.4$ Hz), 6.71 (2H, d, $J = 8.6$ Hz), 6.76 (1H, d, $J = 8.3$ Hz), 7.11 (2H, d, $J = 8.6$ Hz); MS *m*/*z* (relative intensity) 407 (M + H)$^+$, 361 (0.28), 240 (0.43), 194 (0.62); HPLC $t_R = 19.4$ min (gradient), 5.8 min (25% MeCN, isocratic).

In Vitro Assay for β_3-Adrenoceptor-Stimulating Effects. Urinary bladders of male ferrets (1100−1400 g in body weight) and male rats (200−380 g in body weight) were isolated and longitudinal detrusor strips, approximately 10 mm in length and approximately 2 mm in width were prepared. The experiment was performed according to the organ bath method. The preparations were suspended in a Krebs solution maintained at 37 °C and continuously gassed with a mixture of 95% oxygen and 5% carbon dioxide under 1 g of tension. Detrusor tension were measured isometrically with a force-displacement transducer (SB-1T; Nihon-Kohdan, Tokyo, Japan) and recorded on a rectigram (Rectigraph 8K; NEC San-ei, Tokyo, Japan). The drug was added cumulatively to the organ bath every about 5 min. The drug efficacy was evaluated as the molar concentration of the drug required to produce 50% of detrusor relaxation before the addition of the drug. In this experiment, tension of detrusor before the addition of the drug was expressed as 100% and tension of

maximal relaxation after the addition of 10^{-5} M concentration of forskolin was expressed as 0%.

In Vitro Assay for β_1-Adrenoceptor-Stimulating Effects. Atria of male SD rats (250−400 g in body weight) were isolated and the experiment was performed according to the organ bath method. The preparations were exposed to Krebs solution maintained at 37 °C and gassed with a mixture of 95% oxygen and 5% carbon dioxide under 0.5 g of tension. The cardiac contractility was measured isometrically with a force-displacement transducer (SB-1T; Nihon-Kohdan), and heart rate was recorded on a rectigram via a tachometer (Rectigraph 8K; NEC San-ei). The drug was added cumulatively to the Magnus bath every 3 min. The drug efficacy was evaluated as the molar concentration of the drug required to increase 20 beats/minute.

In Vitro Assay for β_2-Adrenoceptor-Stimulating Effects. Uteri of pregnant SD rats (pregnancy day 21, 200−380 g in body weight) were isolated and longitudinal uterine muscle strips of approximately 15 mm in length and 5 mm in width were prepared. The experiment was performed according to the Magnus method. The preparations were exposed to Locke-Ringer solution maintained at 37 °C and continuously gassed with a mixture of 95% oxygen and 5% carbon dioxide under 0.5 g of tension. Spontaneous contractions of myometrium were measured isometrically with a force-displacement transducer (SB-1T; Nihon-Kohdan) and recorded on a rectigram (Rectigraph 8K; NEC San-ei). The drug was added cumulatively to the organ bath every 5 min. The drug efficacy was evaluated as the molar concentration of the drug required to produce 50% of the inhibition of uterine contraction as comparing the total degree of uterine contraction during 5 min before the addition of the drug (100%) with the total degree of uterine contraction during 5 min after the addition of the drug at various concentrations.

Bladder Pressure in Anesthetized Rats. Male rats (300−350 g in body weight) were anesthetized with urethane (1.5 g kg^{-1}, sc), and the midline abdomen was incised. The ureter on each side was ligated and cut proximal to the ligature so as to allow urine to drain into cotton wads. After the urethra had been ligated, a polyethylene catheter (PE-50; Nihon Becton Dickinson, Tokyo, Japan) was inserted into the urinary bladder via the top of the bladder dome and connected through a three-way connector to a pressure transducer (SPB-108; NEC San-ei) and syringe filled with warmed saline. The initial bladder pressure was adjusted to 6 cm H_2O by instillation of warmed saline (37 °C) in 0.05 mL increments. An arterial catheter was inserted into the left carotid artery (PE-90; Nihon Becton Dickinson) and connected to a pressure transducer (SPB-108; NEC San-ei) for the measurement of blood pressure. Heart rate was measured via tachometer (132l; NEC San-ei) connected to the transducer amplifier (1829; NEC San-ei). Bladder pressure, blood pressure, and heart rate were recorded continuously on a rectigraph (Recti-Horiz-8K; NEC San-ei). Drug effects on bladder pressure, blood pressure, and heart rate were quantified by expressing each post-administration value as a percentage of the value before drug administration. No animal was exposed to more than one of the test drugs. A venous catheter was inserted into the left femoral vein (PE-50; Nihon Becton Dickinson) for drug injection.

Acknowledgment. We thank Dr. Yoshinobu Yamazaki and Mr. Hiroo Takeda for their scientific contributions to this work. We also thank Dr. Hiromu Harada for helpful comments while reviewing the manuscript.

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JM000455Z