New Fluorine-Substituted Analogue of Eticlopride with High Affinity toward Dopamine D2 Receptors

Kiyoko WATANABE,[a] Toshimitsu FUKUMURA,[b] Shigeki SASAKI,[a] Minoru MAEDA,*[,a] and Shuzo TAKEHARA[c]

Faculty of Pharmaceutical Sciences,[a] Faculty of Medicine,[b] Kyushu University, 3-1-1 Maidashi, Higashi-ku, Fukuoka 812, Japan, and Central Research Laboratories, Yoshitomi Pharmaceutical Industries Ltd.,[c] 955 Koiwai Yoshitomi-cho, Chikujo-gun, Fukuoka 871, Japan. *Received May 30, 1991*

Aiming at the development of positron-emitting ligands with specific and high affinity toward dopamine D2 receptors in the central nervous system, we synthesized a new fluorinated eticlopride derivative. A fluorine atom was introduced at the C-4 position of the pyrrolidine ring of eticlopride, a dopamine D2 antagonist of the benzamide series. The *in vitro* binding affinity of this ligand toward the D2 receptor was found to be as potent as eticlopride, suggesting that the corresponding ^{18}F-labelled compound may be useful as an *in vivo* radioligand for positron emission tomography.

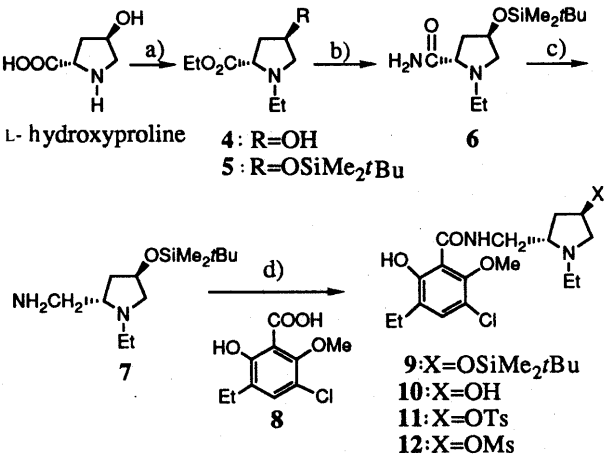
Keywords fluoroeticlopride; antagonist; dopamine D2 receptor; *in vitro* binding affinity

Much effort has been pursued to develop positron-emitting ligands for *in vivo* imaging of the D2 dopamine receptors in the central nervous system (CNS), expecting such radioligands to be clinically useful for monitoring the function of the dopaminergic system in the living human brain by positron emission tomography (PET).[1] Recently, benzamide neuroleptics, *i.e.* D2 antagonists have been chosen as parent compounds in designing a radioligand,[2] since they have been known to show specific, high affinity and reversible binding to D2 dopamine receptors.[3] In addition, among the positron-emitting nuclides, fluorine-18 with a relatively long half-life (110 min) has been accepted as a favorable nuclide for PET studies of slow ligand-receptor binding.[4] However, previous studies on fluorinated ligands by us and other groups have pointed out some problems, for example, eticlopride (**1**)[5] or raclopride (**2**)[6] derivatives with *N*-fluoroalkylated pyrrolidine showed only a low affinity towards D2 receptors. In the present study we wish to report that a new fluorinated eticlopride (**3**), in which a fluorine atom is introduced at the C-4 position of the pyrrolidine skeleton, has retained a high affinity toward D2 receptors as evidenced by *in vitro* binding experimentation.

The synthetic route for the new eticlopride analogue having the fluorinated pyrrolidine ring is summarized in Charts 1 and 2. The substituted pyrrolidine moiety was prepared from commercially available (*S*)-hydroxyproline (Chart 1). Esterification of (*S*)-hydroxyproline with ethanol–HCl, and subsequent *N*-ethylation using ethyl iodide in dioxane–water in the presence of triethylamine gave *N*-ethyl-4-hydroxy-(*S*)-proline ethyl ester **4** in 53% yield. The hydroxy group was protected by *tert*-butyldimethylsilyl group to give **5** (99% yield), then the ethoxycarbonyl group was transformed to the carboxamide (**6**) by the reaction with NH_3 in anhydrous methanol in the presence of a catalytic amount of KI, in 92% overall yield. The reduction of **6** with $LiAlH_4$ in ether afforded the diamine **7** in 82% yield.

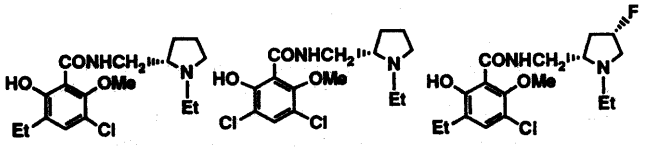
Coupling between the amine **7** and the carboxylic acid **8** was performed using 1-ethyl-3-(3-dimethylaminopropyl)-carbodiimide hydrochloride (WSC) in the presence of 1-hydroxybenzotriazole as a catalyst to give the desired eticlopride derivative **9** in 84% yield. Deprotection of **9** with *n*-Bu_4NF in tetrahydrofuran (THF) (94% yield), and following sulfonylation of the resulting alcohol **10** with *p*-toluenesulfonyl chloride or methanesulfonyl chloride yielded the sulfonate precursors **11** (29% yield) or **12** (84% yield).

The fluoroeticlopride (**3**) was synthesized by three



(a) i) HCl–EtOH; ii) EtI, Et_3N, in dioxane–H_2O; iii) *tert*-butyldimethylchlorosilane (TBDMSCl), imidazole, in dimethylformamide (DMF). (b) NH_3–MeOH, KI. (c) $LiAlH_4$ in ether. (d) **9**; **8**, 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride, 1-hydroxybenzotriazole, in CH_2Cl_2, **10**: *n*-Bu_4NF in THF; **11**: TsCl in pyridine; **12**: MsCl, Et_3N in CH_2Cl_2. *t*Bu = *tert*-butyl.

Chart 1. Synthesis of (2*S*,4*R*)-(−)-2-Aminomethyl-1-ethyl-4-*tert*-butyldimethylsilyloxypyrrolidine Unit and the Intermediates for Fluorination



eticlopride (**1**) raclopride (**2**) fluoroeticlopride (**3**)

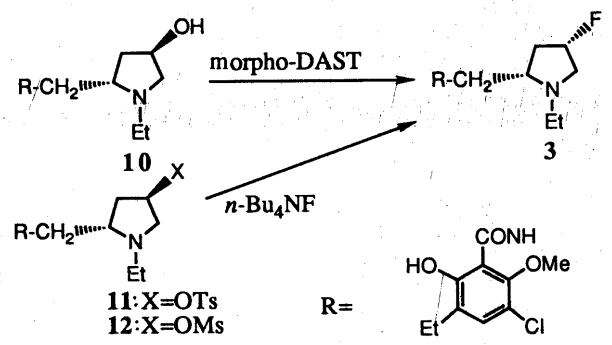
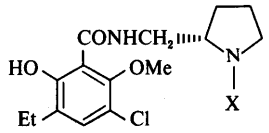


Chart 2. Synthesis of Fluoroeticlopride

TABLE I. Inhibition of [^{3}H]Spiperone Binding in Striatal Homogenates of Rat Brain[a]

Run	Compound	[^{3}H]Spiperone binding IC_{50} (nM)
1	Eticlopride	2.9
2	**9**	35
3	**10**	4.5
4	**3**	1.9
5[b]	**13**	23
6[b]	**14**	17
7[b]	**15**	4800



	X
13	CH_2CH_2F
14	$CH_2CH_2CH_2F$
15	$CH_2C_6H_4$-F(*p*)

a) Homogenates of the rat striatal tissue were incubated with 0.2 nM [^{3}H]spiperone and various concentrations of the compounds. Nonspecific binding was determined in the presence of 10^{-4} M sulpiride. IC_{50} represents the concentration of the compound required to inhibit 50% of specifically bound [^{3}H]spiperone. Values represent means of three determinations. *b*) Data taken from the literature in which homogenates of the bovine striatal tissue *in vitro* were used.[5]

approaches as shown in Chart 2. Fluorination of **11** by treatment with *n*-Bu_4NF in refluxing THF (25 min) gave **3** in 47% yield, whereas a longer reaction time (6.5 h) was needed in the case of the reaction using the mesylate **12** as a substrate (18% yield). The fluorination reaction could also be achieved by using the alcohol **10** and morpholinosulfur trifluoride (morpho DAST)[7] in 16% yield. The fluorinated compounds obtained by the above three approaches were identical in their spectral data, indicating that all the fluorinations proceeded in a manner of SN_2 inversion, and that the configuration of the fluoroeticlopride (**3**) should be as shown in Chart 2.

In order to evaluate the binding affinity of new eticlopride derivatives toward D2 receptors, IC_{50} values against [^{3}H]-spiperone binding in rat striatal tissue preparation were measured.[8] Table I summarizes the results, together with the reported data for comparison. It should be noted that the affinity of the fluoroeticlopride to the D2 receptor is as high as the parent eticlopride (run 1 *vs.* 4). It was also found that the hydroxy group had almost no effect in the binding affinity (run 3), whereas the bulky protecting group caused a slight decrease in the affinity (run 2).

Recent studies on the structure–affinity relationship of benzamide antagonists toward D2 receptors have suggested that the hydrogen bonding between the protonated pyrrolidine and the complementary receptor site plays an important role in the ligand affinity.[9] It is known that substitution by fluorine atoms of the methylene hydrogens in the β or γ-position of aliphatic amines causes a marked decrease in the amine basicity.[10] The fact that the benzamide ligands with the *N*-fluoroalkyl-substituted pyrrolidine showed only diminished affinities toward the D2 receptor (run 5 and 6) was explained partly on the basis of a large electronegativity of the fluorine atom.[6a] That is, the electron-withdrawing effect of the fluorine introduced may decrease protonation of the pyrrolidine nitrogen, and therefore, the hydrogen bonding with the receptor may be weakened. Interestingly, despite the fluorine atom introduced at the β position to the pyrrolidine nitrogen, the fluoroeticlopride **3** has retained a high affinity toward the

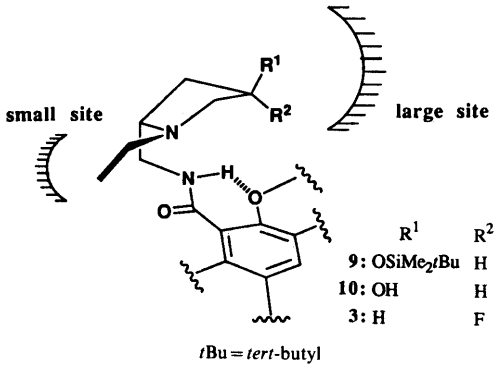


Fig. 1. A Model for the Binding of the Pyrrolidine Moiety of the Eticlopride Derivatives

D2 receptors. These findings may be of help in better understanding of the electronic effect in the ligand-receptor binding and the influence of the fluorine atom introduced.

Steric interactions between the ligands and the receptor surface are another important aspect in determining the affinity. The slight influence of the C-4 substituents (run 2, 3, and 4) on the affinity to the receptor, together with the affinity-reducing effect of *N*-fluorobenzyl derivative (**15**) (run 7), lead us to assume a receptor model for the binding sites in which pyrrolidine *N*-substituents are thought to be directed to a sterically hindered site, whereas a relatively large cleft appears to be accommodated to the C-4 substituents. Independently of us, a new spatial model for the D2 binding sites has been recently proposed based on the difference in affinities of enantiomeric benzamide derivatives, in which the D2 binding sites are thought to involve both a small and a large site for the interaction of the pyrrolidine moiety.[11] Figure 1 was drawn so that the new eticlopride derivatives described here can be applied to such a model, indicating that the substituents at the C-4 position of the pyrrolidine ring seem to be located in the larger site.

Thus, this study indicates that the new fluoroeticlopride **3** with a fluorine atom at the pyrrolidine C-4 position is a promising ligand for the investigation of dopamine D2 receptors. *In vivo* biodistribution in rats using the ^{18}F-labelled **3** showed a specific uptake in the striatum, and the detailed *in vivo* results will soon be reported.

Experimental

Melting points were determined on a Yanagimoto micro-melting point apparatus and are not corrected. Optical rotations were determined by a JASCO DIP-360 digital polarimeter. Proton nuclear magnetic resonance (^{1}H-NMR) spectra were taken at either 100 MHz (JEOL FX-100) or at 270 MHz (JEOL GX-270), and chemical shifts are reported in ppm (δ) downfield from tetramethylsilane (δ 0.0). Infrared (IR) spectra were taken on a JASCO IRA-1 spectrometer. Low resolution electron impact (EI), field desorption (FD), and fast atom bombardment (FAB) mass spectra (respective abbreviations are as follows: EIMS, FDMS, and FABMS) as well as high resolution EI mass spectra (HRMS) were obtained on a JEOL TMS-D300 spectrometer. Low resolution secondary ion (SI) and high resolution SI mass spectra (SIMS and HRSIMS) were obtained on a HITACHI M-2000 spectrometer. Kieselgel 60 (70—230 mesh, Merck) was used for column chromatography. Elemental analyses were performed by the staff of the microanalytical section of Kyushu University. [^{3}H]-Spiperone (1.48 TBq/mmol) was purchased from New England Nuclear. Eticlopride, (*S*)-(−)-5-chloro-3-ethyl-*N*-[(1-ethyl-2-pyrrolidinyl)methyl]-6-methoxysalicylamide, was prepared by the published procedure.[12]

(2*S*,4*R*)-(−)-1-Ethyl-4-*tert*-butyldimethylsilyloxy-2-ethoxycarbonylpyrrolidine (5) A solution of 4-hydroxy-L-proline ethyl ester hydro-

chloride (1.5 g, 7.69 mmol), triethylamine (1.8 g, 17.8 mmol), and ethyl iodide (0.195 g, 12.5 mmol) in water–dioxane (7.5 ml–7.5 ml) was stirred at room temperature for 5 d. The reaction mixture was extracted with chloroform, and the extract was dried over anhydrous Na_2SO_4 and filtered, then evaporated. The residue was purified by column chromatography (silica gel, chloroform : methanol = 15 : 1) to give a colorless oil (0.77 g, 54%). This oil was used for the next reaction without further purification. IR (neat): 3400, 1740 cm^{-1}. ^{1}H-NMR ($CDCl_3$) (100 MHz) δ: 4.50 (1H, m), 4.19 (2H, q, J = 7.0 Hz), 3.50 (1H, dd, J = 10.0, 7.8 Hz), 3.46 (1H, dd, J = 10.0, 5.8 Hz), 2.66 (2H, q, J = 7.0 Hz), 2.41 (1H, m), 2.14 (2H, m), 1.27 (3H, t, J = 7.1 Hz), 1.10 (3H, t, J = 7.3 Hz).

A solution of the above oil (0.49 g, 2.60 mmol), *tert*-butyldimethylsilyl chloride (0.56 g, 3.72 mmol), and imidazole (0.46 g, 6.76 mmol) was stirred at room temperature for 3 d. The reaction mixture was extracted with pentane (65 ml), and the extract was washed successively with water and brine, then dried over anhydrous Na_2SO_4. Filtration and evaporation of the solvent gave an oil (0.78 g, 99%). IR (neat): 1740 cm^{-1}. ^{1}H-NMR ($CDCl_3$) (100 MHz) δ: 4.44 (1H, qui, J = 5.6 Hz), 4.19 (2H, q, J = 7.2 Hz), 3.41 (1H, dd, J = 9.5, 5.8 Hz), 3.38 (1H, dd, J = 8.3, 7.4 Hz), 2.77 (1H, dq, J = 12.0, 7.2 Hz), 2.52 (1H, dq, J = 12.0, 7.2 Hz), 2.29 (1H, dd, J = 9.5, 5.3 Hz), 2.08 (2H, m), 1.27 (3H, t, J = 7.2 Hz), 1.08 (3H, t, J = 7.2 Hz), 0.88 (9H, s), 0.04 (6H, s). EIMS m/z: 301 (M^+). HRMS m/z: Calcd for $C_{15}H_{31}NO_3Si$: 301.2071. Found: 301.2074.

(2*S*,4*R*)-(−)-1-Ethyl-4-*tert*-butyldimethylsilyloxy-2-pyrrolidinecarboxamide (6) A solution of **5** (0.47 g, 1.56 mmol) and KI (30 mg) in methanol saturated with NH_3 (20 ml) was heated at 40 °C for 7 d. Evaporation of the solvent gave a residue, which was purified by column chromatography (silica gel, chloroform) to yield colorless needles (0.40 g, 93%). mp 56 °C. $[\alpha]_D^{26} = -68.3°$ (c = 1.2, $CHCl_3$). IR (Nujol): 3400, 1680 cm^{-1}. ^{1}H-NMR ($CDCl_3$) (100 MHz) δ: 7.21 (2H, br), 4.31 (1H, qui, J = 5.5 Hz), 3.31 (1H, dd, J = 9.5, 4.8 Hz), 3.26 (1H, dd, J = 7.3, 7.3 Hz), 2.59 (2H, q, J = 7.1 Hz), 2.30 (1H, ddd, J = 9.5, 5.5, 4.7 Hz), 2.06 (2H, m), 1.07 (3H, t, J = 7.1 Hz), 0.87 (9H, s), 0.05 (6H, s). FABMS m/z: 273 (MH^+). *Anal.* Calcd for $C_{13}H_{28}N_2O_2Si$: C, 57.31; H, 10.36; N, 10.28. Found: C, 57.54; H, 10.30; N, 10.22.

(2*S*,4*R*)-(−)-2-Aminomethyl-1-ethyl-4-*tert*-butyldimethylsilyloxypyrrolidine(7) A mixture of **6** (1.00 g, 3.68 mmol) and $LiAlH_4$ (0.60 g, 15.5 mmol) in ether (10 ml) was heated under reflux for 2 d. The reaction mixture was quenched with aqueous saturated Na_2SO_4, and extracted with ether. The extract was dried over anhydrous Na_2SO_4, and evaporated to give a crude oil, which was purified by column chromatography (silica gel, chloroform : methanol = 5 : 1—methanol) to yield a pure oil (0.78 g, 82%). $[\alpha]_D^{30} = -41.3°$ (c = 0.8, $CHCl_3$). IR (neat): 3350 cm^{-1}. ^{1}H-NMR ($CDCl_3$) (100 MHz) δ: 4.32 (1H, qui, J = 6.0 Hz), 3.33 (1H, dd, J = 9.3, 6.1 Hz), 2.82 (1H, q, J = 7.1 Hz), 2.50—2.80 (3H, m), 2.29 (1H, q, J = 7.1 Hz), 2.20 (1H, dd, J = 9.5, 6.0 Hz), 1.90—1.75 (4H, m), 1.07 (3H, t, J = 7.2 Hz), 0.88 (9H, s), 0.05 (6H, s). FDMS m/z: 259 (MH^+). HRSIMS m/z: Calcd for $C_{13}H_{31}N_2OSi$: 259.2204. Found: 259.2270.

(2′*S*,4′*R*)-(−)-5-Chloro-3-ethyl-*N*-[(1-ethyl-4-*tert*-butyldimethylsilyloxy-2-pyrrolidinyl)methyl]-6-methoxysalicylamide (9) 1-Hydroxybenzotriazole (56 mg, 0.415 mmol) and 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (78 mg, 0.405 mmol) were added to a solution of 5-chloro-3-ethyl-6-methoxysalicylic acid (**8**)[12] (89 mg, 0.39 mmol) in anhydrous dichloromethane (2 ml) at −10 °C, and the whole was stirred for 100 min at −10 °C and for 30 min at room temperature. A solution of **7** (96 mg, 0.371 mmol) in dichloromethane (2 ml) was added dropwise to the above mixture, and the mixture was stirred overnight at room temperature. The solvent was removed and the residue was purified by column chromatography (silica gel, chloroform : methanol = 15 : 1) to give a pure oil (0.15 g, 84%). $[\alpha]_D^{25} = -34.2°$ (c = 0.95, $CHCl_3$). IR (neat): 3350, 1635 cm^{-1}. ^{1}H-NMR ($CDCl_3$) (270 MHz) δ: 13.8 (1H, br), 8.83 (1H, br), 7.22 (1H, s), 4.30 (1H, qui, J = 5.5 Hz), 3.87 (3H, s), 3.75 (1H, ddd, J = 14.1, 7.6, 2.3 Hz), 3.40 (1H, dd, J = 9.8, 5.9 Hz), 3.30 (1H, dd, J = 14.1, 2.6 Hz), 3.01 (1H, br), 2.86 (1H, dq, J = 12.1, 7.3 Hz), 2.62 (2H, q, J = 7.6 Hz), 2.37 (1H, dq, J = 12.1, 7.3 Hz), 2.27 (1H, dd, J = 9.8, 5.5 Hz), 1.83 (2H, m), 1.20 (3H, t, J = 7.6 Hz), 1.11 (3H, t, J = 7.3 Hz), 0.87 (9H, s), 0.04 (6H, s). SIMS m/z: 471 (MH^+). HRSIMS m/z: Calcd for $C_{23}H_{40}ClN_2O_4Si$: 471.2469. Found: 471.2472.

(2′*S*,4′*R*)-(−)-5-Chloro-3-ethyl-*N*-[(1-ethyl-4-hydroxy-2-pyrrolidinyl)methyl]-6-methoxysalicylamide (10) A solution of n-Bu_4NF (1.3 mmol) and **9** (0.19 g, 0.403 mmol) in THF (2.8 ml) was stirred at room temperature for 3 d. The reaction mixture was quenched with aqueous saturated NH_4Cl, and extracted with ether, then the extract was dried over anhydrous Na_2SO_4. Evaporation of the solvent gave a crude product, which was purified by column chromatography (silica gel, chloroform : methanol = 10 : 1 or ethyl acetate : n-hexane = 3 : 1) to afford a pure oil (0.14 g, 94%). $[\alpha]_D^{28} = -52.0°$ (c = 1.0, $CHCl_3$). IR (neat): 3350, 1635 cm^{-1}. ^{1}H-NMR ($CDCl_3$) (270 MHz) δ: 13.7 (1H, br), 8.85 (1H, br), 7.22 (1H, s), 4.40 (1H, br), 3.87 (3H, s), 3.79 (1H, ddd, J = 14.2, 7.6, 2.0 Hz), 3.54 (1H, dd, J = 9.9, 5.6 Hz), 3.33 (1H, ddd, J = 14.2, 4.6, 2.6 Hz), 3.06 (1H, br), 2.89 (1H, dq, J = 12.1, 7.3 Hz), 2.62 (2H, q, J = 7.5 Hz), 2.37 (1H, dq, J = 12.1, 7.3 Hz), 2.31 (1H, dd, J = 9.9, 4.6 Hz), 1.91 (2H, m), 1.20 (3H, t, J = 7.5 Hz), 1.13 (3H, t, J = 7.3 Hz). FABMS m/z: 357 (MH^+). HRSIMS m/z: Calcd for $C_{17}H_{26}ClN_2O_4$: 357.1580. Found: 357.1579.

(2′*S*,4′*R*)-(−)-5-Chloro-3-ethyl-*N*-[(1-ethyl-4-*p*-toluenesulfonyloxy-2-pyrrolidinyl)methyl]-6-methoxysalicylamide (11) *p*-Toluenesulfonyl chloride (57 mg, 0.295 mmol) was added to a solution of **10** (105 mg, 0.295 mmol) in pyridine (0.76 g, 9.5 mmol) at 0 °C, and the whole was stirred for 4 h at 0 °C and for 26 h at room temperature. The reaction mixture was quenched with ice-water, then extracted with ether. The water layer was separated and alkalinized with concentrated aqueous NH_3, then again extracted with ethyl acetate. The combined organic layers were dried over anhydrous Na_2SO_4, and evaporated to give a crude oil, which was purified by column chromatography (silica gel, chloroform : methanol = 10 : 1) to afford a pure oil (44 mg, 29%). $[\alpha]_D^{25} = -9.3°$ (c = 1.0, $CHCl_3$). IR (neat): 3350, 1635 cm^{-1}: ^{1}H-NMR ($CDCl_3$) (270 MHz) δ: 13.67 (1H, s), 8.72 (1H, br d, J = 6.3 Hz), 7.76 (2H, dt, J = 8.5, 1.98 Hz), 7.33 (2H, dd, J = 8.5, 0.7 Hz), 7.23 (1H, s), 4.87 (1H, m), 3.82 (3H, s), 3.75 (1H, ddd, J = 14.5, 7.9, 1.98 Hz), 3.51 (1H, dd, J = 10.9, 5.9 Hz), 3.27 (1H, dm, J = 14.2 Hz), 2.97 (1H, br), 2.85 (1H, dq, J = 12.2, 7.5 Hz), 2.61 (2H, q, J = 7.3 Hz), 2.55 (1H, dd, J = 10.9, 5.0 Hz), 2.44 (3H, s), 2.37 (1H, dq, J = 12.2, 7.5 Hz), 1.99 (1H, ddd, J = 14.5, 7.3, 3.3 Hz), 1.83 (1H, ddd, J = 14.5, 9.2, 7.3 Hz), 1.19 (3H, t, J = 7.5 Hz), 1.08 (3H, t, J = 7.3 Hz). FDMS m/z: 510 (M^+), 512 $(M+2)^+$. HRSIMS m/z: Calcd for $C_{24}H_{32}ClN_2O_6S$: 511.1695. Found: 511.1667.

(2′*S*,4′*R*)-(−)-5-Chloro-3-ethyl-*N*-[(1-ethyl-4-methanesulfonyloxy-2-pyrrolidinyl)methyl]-6-methoxysalicylamide (12) Methanesulfonyl chloride (45 mg, 0.389 mmol) and triethylamine (41 mg, 0.405 mmol) were added to a solution of **10** (97 mg, 0.27 mmol) in dichloromethane at 0 °C, and the whole was stirred for 2 h at 0 °C. The reaction mixture was quenched with ice-water, then extracted with dichloromethane. The extract was dried over anhydrous Na_2SO_4, and evaporated to give a crude oil, which was purified by column chromatography (silica gel, chloroform : methanol = 10 : 1) to afford a pure oil (99 mg, 84%). $[\alpha]_D^{24} = -20.7°$ (c = 0.9, $CHCl_3$). IR (neat): 3350, 1635 cm^{-1}. ^{1}H-NMR ($CDCl_3$) (100 MHz) δ: 13.67 (1H, br), 8.77 (1H, br), 7.23 (1H, s), 5.08 (1H, br), 3.87 (3H, s), 3.81—3.13 (3H, m), 3.01 (3H, s), 2.97—1.96 (8H, m), 1.20 (3H, t, J = 7.3 Hz), 1.12 (3H, t, J = 7.1 Hz). FABMS m/z: 435 (MH^+). HRSIMS m/z: Calcd for $C_{18}H_{28}ClN_2O_6S$: 435.1332. Found: 435.1354.

(2′*S*,4′*S*)-(−)-5-Chloro-3-ethyl-*N*-[(1-ethyl-4-fluoro-2-pyrrolidinyl)methyl]-6-methoxysalicylamide (3). The Synthesis Using 11 A solution of n-Bu_4NF (0.11 mmol) and **11** (14 mg, 0.027 mmol) in THF (0.6 ml) was heated under reflux for 25 min. The solvent was removed, and the residue was shaken with brine and ether. The ether layer was separated, and dried over anhydrous Na_2SO_4. Evaporation of the solvent gave a crude product, which was purified by column chromatography (silica gel, ethyl acetate : n-hexane = 3 : 1—5 : 1), then by HPLC (Whatman Partisil 5PAC, 9.4 mm × 10 cm, n-hexane : ethyl acetate = 4 : 1, flow rate = 2 ml/min) to afford a pure oil (4.6 mg, 47%). $[\alpha]_D^{24} = -20.9°$ (c = 1,3, $CHCl_3$). IR (neat): 3350, 1635 cm^{-1}. ^{1}H-NMR ($CDCl_3$) (270 MHz) δ: 13.9 (1H, s), 8.99 (1H, br), 7.21, (1H, s), 5.11 (1H, dddd, J = 55.1, 9.5, 4.6, 2.4 Hz), 3.92 (3H, s), 3.84 (1H, ddd, J = 14.4, 7.3, 2.0 Hz), 3.47 (1H, ddd, J = 20.1, 11.5, 1.4 Hz), 3.37 (1H, ddd, J = 14.4, 4.3, 2.6 Hz), 2.91 (1H, dq, J = 12.2, 7.0 Hz), 2.68 (1H, br), 2.62 (2H, q, J = 7.0 Hz), 2.48—2.15 (3H, m), 1.94 (1H, ddddd, J = 33.3, 14.9, 7.6, 1.7, 1.7 Hz), 1.20 (3H, t, J = 7.0 Hz), 1.15 (3H, t, J = 7.0 Hz). FDMS m/z: 359 (MH^+). HRSIMS m/z: Calcd for $C_{17}H_{25}ClFN_2O_3$: 359.1588, Found 359.1563.

The Synthesis Using 12 A solution of n-Bu_4NF (0.6 mmol) and **12** (66 mg, 0.15 mmol) in THF (1.6 ml) was heated under reflux for 6.5 h. The solvent was removed, and the residue was purified by column chromatography (silica gel, chloroform : methanol = 99 : 1) to afford a pure oil (9.6 mg, 18%). The IR, ^{1}H-NMR, and MS spectra of the product were identical with those of the compound obtained using **11**.

The Synthesis Using 10 A solution of morpholinosulfur trifluoride[7] (87 mg, 0.498 mmol) in dichloromethane (1 ml) was added dropwise to a solution of **10** (89 mg, 0.249 mmol) in dichloromethane (1 ml) at −78 °C. The reaction mixture was stirred for 2 h, while the reaction temperature was raised gradually from −78 °C to room temperature. The reaction mixture was poured into aqueous saturated $NaHCO_3$, and extracted with dichloromethane. The extract was washed with brine, dried over anhy-

drous Na_2SO_4, and then evaporated. The residue was purified by column chromatography (silica gel, *n*-hexane : ethyl acetate = 3 : 1) to give a pure oil (15 mg, 16%). The IR, ^{1}H-NMR, and MS spectra of the product were identical with those of the compound obtained using **11**.

***In Vitro* Receptor Binding Assay** The assays were performed in the rat striatal membranes using a previously described method.[8] Briefly, rat striata were homogenized in 100 volumes of ice-cold Tris–HCl buffer (50 mM, pH 7.7) and centrifuged (500 *g*, 10 min, 0 °C). The supernatant was centrifuged at 50000 *g* for 15 min. The pellet was suspended in 100 volumes of ice-cold Tris–HCl buffer (50 mM, pH 7.7) and recentrifuged (50000 *g*, 15 min, 0 °C). The final pellet was resuspended in 150 volumes of ice-cold Tris–HCl buffer (50 mM, pH 7.1) containing 120 mM NaCl, 5 mM KCl, 2 mM $CaCl_2$, 1 mM $MgCl_2$, 1.1 mM ascorbic acid, and 10 mM pargyline and incubated at 37 °C for 10 min. A portion of this membrane suspension (900 μl) was placed in a tube, and 50 μl of either test compound or vehicle solution was added, followed by 50 μl of [^{3}H]spiperone (1.48 TBq/mmol) at a final concentration of 0.2 nM. The tubes were incubated at 37 °C for 20 min and filtered through Whatman GF/B glass filters, which were then washed three times with 3 ml of the Tris–HCl buffer (50 mM, pH 7.7). The nonspecific binding was determined in the presence of 10^{-4} M of (±)-sulpiride. The radioactivity trapped on the filters was measured by liquid-scintillation spectrometry. The IC_{50} values to displace specific [^{3}H]spiperone binding were determined from concentration-inhibition curves.

Acknowledgements This work was supported by a Grant-in-Aid for Scientific Research (B) from the Ministry of Education, Science and Culture of Japan. We are also grateful to Mitsubishi Kasei Corporation for SI mass spectral (SIMS) measurements.

References and Notes

1) T. Suhara, "Atarasii Nou No Reseputa (Receptors in the Brain, New Edition)," ed. by N. Ogawa, Sekai Hoken Tsusinsha, Osaka, 1989, Part III, Chapter 3, p. 608, and references cited therein.
2) For Example, [^{11}C]eticlopride: C, Halldin, L. Farde, T. Högberg, H. Hall, and G. Sedvall, *Appl. Radiat. Isot.*, **41**, 669 (1990); [^{11}C]raclopride: L. Farde, E. Ehrin, L. Eriksson, T. Greitz, P. Johnström, J.-E. Litton, and G. Sedvall. *Proc. Natl. Acad. Sci., U.S.A.*, **82**, 3863 (1985); [^{11}C]YM-09151-2: K. Hatano, K. Ishikawa, K. Kawashima, J. Hatazawa, M. Itoh, and T. Ido, *J. Nucl. Med.*, **30**, 515 (1989).
3) Raclopride: C. Köhler, O.-S. Ögren, and L. Gawell, *Biochem. Pharmacol.*, **34**, 2251 (1985); eticlopride: C. Köhler, H. Hall, and L. Gawell, *Eur. J. Pharmacol.*, **119**, 191 (1985); YM-09151-2: S. Iwanami, M. Takashima, Y. Hirata, O. Hasegawa, and S. Usuda, *J. Med. Chem.*, **24**, 1224 (1981).
4) Following ^{18}F-labelled ligands have been investigated. [^{18}F]spiroperidol: H. N. Wagner, Jr., H. D. Burns, R. F. Dannals, D. F. Wong, G. Langstrom, T. Duelfer, J. J. Frost, H. T. Ravert, J. M. Links, S. B. Rosenbloon, S. E. Lukas, A. V. Kramer, and M. J. Kuhar, *Science*, **221**, 1264 (1983); M. J. Welch, M. R. Kilbourn, C. J. Mathias, M. A. Mintun, and M. E. Raichle, *Life Science*, **33**, 1687 (1983); C. D. Arnett, A. P. Wolf, C. Y. Shiue, J. S. Fowler, R. R. MacGregor, D. R. Christman, and M. R. Smith, *J. Nucl. Med.*, **27**, 1878 (1986); M. J. Welch, D. Y. Chi, C. J. Mathias, M. R. Kilboun, J. W. Brodack, and J. A. Katzenellenbogen, *Nucl. Med. Biol.*, **13**, 523 (1986); [^{18}F]bezamide: J. Mukherjee, K. E. Luh, N. Yasillo, B. D. Perry, D. Levy, C.-T. Chen, C. Ortega, R. N. Beck, and M. Cooper, *Eur. J. Pharmacol.*, **175**, 363 (1990); J. Mukherjee, B. D. Perry, and M. Cooper, *J. Labelled Compd. Radiopharm.*, **28**, 609 (1990).
5) T. Fukumura, H. Dohmoto, M. Maeda, E. Fukuzawa, and M. Kojima, *Chem. Pharm. Bull.*, **38**, 1740 (1990).
6) *a*) G. S. Lannoye, S. M. Moerlein, D. Parkinson, and M. J. Welch, *J. Med. Chem.*, **33**, 2430 (1990); *b*) D. O. Kieselwetter, R. Kawai, M. Chelliah, E. Owens, C. Mclellan, and R. G. Blasberg, *Nucl. Med. Biol.*, **17**, 347 (1990).
7) K. C. Mange and W. J. Middleton, *J. Fluorine Chem.*, **43**, 337 (1989).
8) I. Greese, R. Schneider, and S. H. Snyder, *Eur. J. Pharmacol.*, **46**, 377 (1977).
9) M. W. Harrold, R. A. Wallace, T. Tarooqui, L. J. Wallace, N. Uretsky, and D. D. Miller, *J. Med. Chem.*, **32**, 874 (1989), and references cited therein. General dopamine-D2 pharmacophore has been discussed in U. Norinder and T. Hoegberg, *Acta Pharm. Nord.*, **1**, 75 (1989).
10) J. G. Baillon, P. S. Mamont, J. Wagner, F. Gerhart, and P. Lux, *Eur. J. Biochem*, **176**, 237 (1989); W. G. Reifenrath, E. B. Roche, and W. A. Al-Turk, *J. Med. Chem.*, **23**, 985 (1980).
11) S. Murakami, N. Marubayashi, T. Fukuda, S. Takehara, and T. Tahara, *J. Med. Chem.*, **34**, 261 (1991).
12) T. de Paulis, H. Hall, S.-O. Ögren, A. Wägner. B. Stensland, and I. Csöregh, *Eur. J. Med. Chem. Chim. Ther.*, **20**, 273 (1985).