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# Syntheses and Adrenergic Agonist Properties of Ring-Fluorinated Isoproterenols

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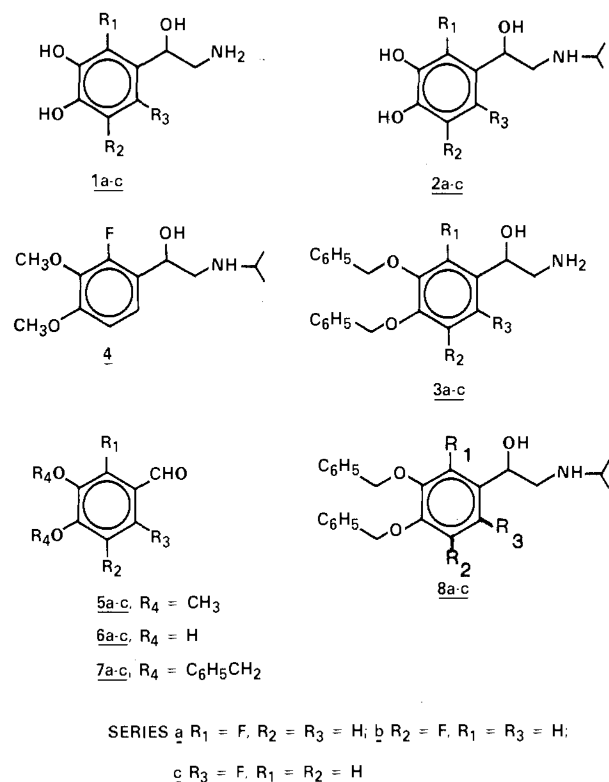
2-Fluoro-, 5-fluoro-, and 6-fluoroisoproterenol were synthesized by reduction of the Schiff base formed between the corresponding ring-fluorinated 3,4-bis(benzyloxy)phenethanolamine and acetone, followed by reductive debenzylation in the presence of oxalic acid to yield crystalline neutral oxalates. The apparent $\beta$-adrenergic potencies were determined in the isolated guinea pig atria. 2-Fluoro- and 5-fluoroisoproterenol were equipotent with (±)-isoproterenol, while 6-fluoroisoproterenol was virtually inactive. No $\alpha$-adrenergic agonist activity (guinea pig aorta) was shown by any of the fluoroisoproterenols. Displacement of $\alpha$- and $\beta$-specific radioligands from isolated membrane preparations from rat brain by the fluoroisoproterenols were in agreement with the responses of the organ preparations. Thus, the apparent fluorine-induced specificity is due to specificity at the receptor binding site. The effects of fluorine substitution are discussed with regard to the apparent *negative* influence of the 6-fluoro substituent on the $\beta$-agonist properties of isoproterenol, the lack of any increase in potency due to the 2-fluoro substituent, and the possibility of fluorine-induced changes in the electron density of the aromatic ring as a possible rational for the fluorine-induced specificity of both the fluoroisoproterenols and the fluoronorepinephrines.

We recently reported the syntheses and biological properties of three isomeric ring-fluorinated norepinephrines (FNE; **1a–c**).[1] The most striking feature of




these analogues is the adrenergic agonist specificity they exhibit in a number of central and peripheral systems.[2] While the specificity and affinity of 5-FNE for both $\alpha$- and $\beta$-adrenergic receptors is nearly identical with that of NE, it does, however, consistently exhibit slightly greater adrenergic agonist activity in most systems. 2-FNE is an essentially pure $\beta$-adrenergic agonist, while 6-FNE is a relatively pure $\alpha$-adrenergic agonist.

Their adrenergic agonist specificities and their structural similarities to NE have made the FNE's quite useful pharmacological tools;[3] the basis of the striking agonist specificities, however, remains to be elucidated. In the past several years, much research and speculation has focused on the structure and function of adrenergic receptors.[4] Nonetheless, the specificities exhibited by the FNE's are not readily explained by existing models of the substrate–receptor complex, models which have been formulated on the basis of structure–activity data from a host of adrenergic agonists and antagonists.[5] In fact, relatively little appears to be known concerning the precise nature of the interaction of the aromatic ring of a catecholamine and the adrenergic receptor.[6]

As a first step toward gaining more information concerning the effects of ring fluorination on adrenergic agonist properties of biogenic amines, we have prepared the three isomeric ring-fluorinated derivatives of isoproterenol (ISO; **2a–c**). By measuring the adrenergic agonist specificities of FISO's, we hoped to determine (1) if the effect of fluorine in the 2 position will be additive to the effect of the *N*-isopropyl group, thus increasing $\beta$-adrenergic potency, and (2) if fluorine in the 6 position can overcome the strong influence of the isopropyl group and, if so, whether or not $\alpha$-adrenergic activity will increase with decreasing $\beta$-adrenergic activity.

**Chemistry.** Reductive alkylation of the ring-fluorinated dimethoxyphenethanolamines seemed an attractive ap-

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Table I. Yields and Physical Data of Ring-Fluorinated Isoproterenols

| compd | yield, % | mp, °C | purification[a] | formula | anal. |
|---|---|---|---|---|---|
| **4** | 59 | 93-94 | EtOAc/cyclohexane | $C_{13}H_{20}FNO_3$ | C, H, N |
| **2a**·HCl | 66 | not cryst | | $C_{11}H_{16}FNO_3 \cdot HCl$[b] | |
| **6a** | 64 | 164-166 | sublime | $C_7H_5FO_3$ | C, H |
| **7a** | 44[a] | 89-90 | $Et_2O$/PE | $C_{21}H_{17}FO_3$ | C, H |
| **3a** | 76 | 114-115 | EtOAc/cyclohexane | $C_{22}H_{22}FNO_3$ | C, H, N |
| **8a** | 97 | 93.5-94.0 | EtOAc/cyclohexane | $C_{25}H_{28}FNO_3$ | C, H, N |
| **2a** | 32 | 168-170 | $CH_3CN$/water | $C_{11}H_{16}FNO_3 \cdot 0.5H_2C_2O_4$ | C, H, N |
| **1a** oxalate | 88 | 199-201 | water/acetone | $C_8H_{10}FNO_3 \cdot 0.5H_2C_2O_4$ | C, H, N |
| **6b** | 61 | 190-191 | sublime | $C_7H_5FO_3$ | C, H |
| **7b** | 48 | 81-82 | $Et_2O$/PE | $C_{21}H_{17}FO_3$ | C, H |
| **3b** | 89 | 82-83 | EtOAc/cyclohexane | $C_{22}H_{22}FNO_3$ | C, H, N |
| **8b** | 68 | 101-102 | cyclohexane | $C_{25}H_{28}FNO_3$ | C, H, N |
| **2b** oxalate | | | $CH_3CN$/water | $C_{11}H_{16}FNO_3 \cdot 0.5H_2C_2O_4 \cdot 0.75H_2O$ | C, H, N |
| **8c** | 54 | 115-116 | $Et_2O$/pentane | $C_{25}H_{28}FNO_3$ | C, H, N |
| **2c** oxalate | 23 | 176-177 | ethanol | $C_{11}H_{16}FNO_3 \cdot 0.5H_2C_2C_2O_4$ | C, H, N |

[a] EtOAc = ethyl acetate; $Et_2O$ = ethyl ether; PE = petroleum ether. [b] Not analyzed; mass spectrum (M + 1) was in agreement with calculated molecular weight.

Table II. NMR Parameters[a]

| no. | aromatic protons $\delta$ | aromatic protons $J$ | nonaromatic protons $\delta$ | nonaromatic protons $J$ |
|---|---|---|---|---|
| **2a** | 6.79 (dq, $H_5$, $H_6$) | $HH^o$ = 8.6[b] | 1.38 (d, $CHCH_3$) | 6.5 |
| | | $HF^m$ = 7.1[c] | 3.32 (d, $CH_2N$) | 6.0 |
| | | $HF^p$ = −2.0[c] | 3.54 [q, $CH(CH_3)_2$] | 6.5 |
| | | | 5.16 (t, Ar *CH*) | 6.0 |
| **2b** | 6.8 (m, unresolved AB portion of ABX multiplet) | | 1.34 (d, $CHCH_3$) | 6.6 |
| | | | 3.24 (d, $CH_2N$) | 5.9 |
| | | | 3.45 [q, $CH(CH_3)_2$] | 6.6 |
| | | | 4.85 (t, Ar *CH*) | 5.9 |
| **2c** | 6.72 (d, H5) | $HF^o$ = 11.2 | 1.45 (d, $CHCH_3$) | 6.6 |
| | 6.94 (d, H2) | $HF^m$ = 7.2 | 3.31 (d, $CH_2N$) | 5.9 |
| | | | 3.48 [q, $CH(CH_3)_2$] | 6.6 |
| | | | 5.15 (t, Ar *CH*) | 5.9 |

[a] 100-MHz spectra were measured in $D_2O$ on a JEOL FX100 spectrometer. [b] Superscript abbreviations used are: o, ortho; m, meta; p, para. [c] The two HF coupling constants are of opposite sign. The assignment of the negative value to the para coupling constant is arbitrary.

proach to the FISO's, since these same intermediates were used in our FNE syntheses.[1] Indeed, catalytic (Pt) or sodium borohydride reduction of the Schiff base formed from acetone and 2-fluoro-3,4-dimethoxyphenethanolamine smoothly produced the desired *N*-isopropylphenethanolamine **4**. Boron tribromide demethylation gave 2-FISO (**2a**) as the noncrystalline hydrochloride, following Dowex ion-exchange chromatography (Tables I and II).

While the above procedure gave homogeneous material, as judged by mass spectrometry, HPLC, TLC, and NMR spectroscopy, difficulties in freeing the final product from the inorganic residue during the final Dowex ion-exchange chromatography and the inability to crystallize 2-FISO·HCl led us to consider a modification in the synthetic approach. We had encountered similar difficulties in our FNE synthesis in that of the three FNE·HCl salts prepared by boron tribromide demethylation and purification by ion exchange chromatography, only 5-FNE was obtained in crystalline form. We circumvented the cumbersome final step in the case of 6-FNE by replacing the methyl group with the benzyl group for protection of the catechol and thus obtained crystalline 6-FNE oxalate following hydrogenolysis of the dibenzyl derivative **3c**. Efforts to use a similar approach to prepare 2-FNE oxalate were thwarted by our inability to benzylate 2-fluoro-3,4-dihydroxybenzaldehyde (**6a**).[1] This difficulty appeared to stem from loss of fluorine under the basic conditions of benzylation, presumably because of activation by the *o*-formyl group. We now have succeeded in carrying out this sequence by blocking the aldehyde as the dimethyl acetal prior to benzylation. In situ acid hydrolysis of the bis(benzyloxy)benzaldehyde dimethyl acetal afforded **7a**, which was converted to **3a** through the trimethylsilyl cyanide addition/lithium aluminium hydride reduction sequence.[1] Hydrogenolysis of **3a** in the presence of oxalic acid afforded 2-FNE (**1a**) as the crystalline neutral oxalate, thus ameliorating a persistent problem in providing adequate supplies of the three FNE's. Reductive alkylation (acetone/sodium borohydride)[7] followed by hydrogenolysis, likewise produced 2-FISO (**2a**) oxalate. 5-FISO oxalate was prepared by use of the same sequence from 5-fluoro-3,4-dimethoxybenzaldehyde (**5b**). Reductive alkylation of the previously prepared **3c**, followed by hydrogenolysis, afforded 6-FISO oxalate.

## Results and Discussion

The $\beta$-adrenergic agonist properties of **2a–c** were assayed in isolated guinea pig atria as described previously.[1] In this system, 2-FISO, 5-FISO, and (±)-ISO were essentially equipotent, yielding $EC_{50}$ values of 0.01, 0.02, and 0.03 $\mu$M, respectively. 6-FISO, on the other hand, was approximately 1000 times less active ($EC_{50} \simeq 30$ $\mu$M) than ISO (Figure 1). The weak agonist activity of 6-FISO was not due to partial agonist activity, however, since a maximal rate increase could be approached at concentrations of 100 to 200 $\mu$M. The atrial response to ISO and to the fluoro isomers was completely abolished by propranolol (1 $\mu$M) and was unaffected by the presence of phentolamine (10

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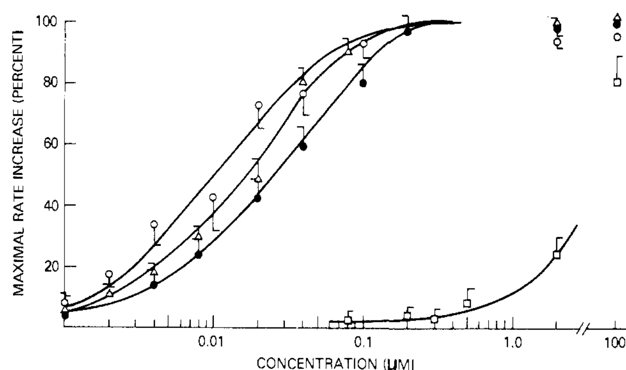


**Figure 1.** Dose–response curves for (±)-ISO (●), 5-FISO (○), 2-FISO (△), and 6-FISO (□) in the isolated guinea pig atria. Preparations removed from 250-g male Hartley guinea pigs were individually bathed in 10-mL chambers containing guinea pig Tyrode's solution at 37 °C and were aerated with 95% $O_2$/5% $CO_2$. Tension and rate were monitored with a Grass Model 79D polygraph and Grass FTO3C force-displacement transducers. Results are expressed as the percentage of maximal rate increase. Preparations were allowed to equilibrate for 30 min before application of the initial dose. Rate measurements were taken 60 s after addition of the drug to the bath. Results are the means ± SD of nine separate atrial preparations.

Table III. Effect of Fluorine Substitution on the Affinity of Isoproterenol for α- and β-Adrenergic Receptors in Rat Brain Cerebral Cortical Membranes

| ligand | displacement of α- and β-specific ligands:[a] $IC_{50}$, μM | |
|---|---|---|
| | α | β |
| (±)-ISO | >100 | 0.1 |
| 2-FISO[b] | 59 | 0.5 |
| 5-FISO | 55 | 0.6 |
| 6-FISO | 80 | 5.5 |
| (±)-NE | 5 | 1.0 |
| propranolol | | 0.03 |
| phentolamine | 0.09 | |

[a] $IC_{50}$ values were estimated from dose–response curves (six) for displacement of an α-adrenergic receptor ligand, 2-[[[2,6-dimethoxyphenoxy)ethyl]amino]methyl]benzodioxan ([$^3$H]WB-4101), or for displacement of a β-adrenergic receptor ligand, [$^3$H]dihydroalprenolol. Measurements were carried out as previously described; see ref 1b and 2e. [b] In all cases, the fluorine-substituted derivatives of isoproterenol were the racemates.

μM). None of the fluoro derivatives or ISO gave measurable α-adrenergic agonist activity (contraction) in the isolated guinea pig aorta at concentrations up to 200 μM. Control values in the same aortic preparations with the α-adrenergic agonists, (±)-norepinephrine and phenylephrine, gave $EC_{50}$ values of 2.6 and 6 μM, respectively.

The displacement of α- and β-adrenergic receptor specific radioligands by the fluoroisoproterenols was determined as described previously.[2e] As shown in Table III, both isoproterenol and the fluoroisoproterenols were only minimally effective in displacing the α-adrenergic receptor specific ligand [$^3$H]WB-4101 from rat brain cerebral cortical membranes. Thus, the affinities of these amines for the α-adrenergic receptor are 10 to >20 times less than that obtained for (±)-norepinephrine ($IC_{50}$ = 5 μM) and much less than the α-adrenergic antagonist, phentolamine ($IC_{50}$ = 0.09 μM). The relatively low affinity of these derivatives is in agreement with the absence of response observed in the isolated guinea pig aorta. The displacement of the β-adrenergic receptor specific ligand [$^3$H]dihydroalprenolol by 2-FISO and 5-FISO was slightly greater than (±)-NE ($IC_{50}$ = 1.0 μM) and less effective than the β-specific agonist (±)-ISO ($IC_{50}$ = 0.1 μM). The displacement by the β-adrenergic antagonist, propranolol ($IC_{50}$ = 0.03 μM), was much more effective than the agonists. 6-FISO, while it displayed some affinity for the β-adrenergic receptor ($IC_{50}$ = 5.5 μM), was approximately 10-fold less than 2- or 5-FISO and 55-fold less than (±)-ISO. Thus, the affinities of 2-FISO, 5-FISO, and ISO are in agreement with their β-agonist potencies in the guinea pig atrial preparation. The lower affinity of 6-FISO for the β-adrenergic receptor compared to 2-FISO, 5-FISO, and ISO is also partially in agreement with the low potency of 6-FISO in stimulating the β-specific guinea pig atrial preparation. 6-FISO, at high concentrations (Figure 1), can induce a maximal rate of contraction in the atria, indicating a full agonist character but possessing a reduced affinity for the β-adrenergic receptor.

At least two conclusions can be drawn from these results. First, the effect of fluorine substitution on the 2 and 5 position of ISO does not alter affinity of this catecholamine for the adrenergic receptor. Fluorine substitution of ISO in the 6 position, on the other hand, effectively reduces the agonist activity, overcoming the strong influence of the *N*-isopropyl group. The second point is that the effect of fluorine substitution is a *negative* effect. Thus, no apparent α-adrenergic activity is imparted to ISO by fluorine substituted in the 6 position; furthermore, 2-FISO is no better as a β-adrenergic agonist than ISO itself.

Since it is apparent that fluorine substitution influences adrenergic agonist specificities by inhibiting binding to the α- or β-adrenergic receptor (2-substitution and 6-substitution, respectively), it is instructive to consider possible mechanisms for this inhibition. We have suggested in previous reports that rotamer stabilization through intramolecular hydrogen bonding of the benzylic hydroxyl group to an ortho fluorine substituent conveniently explains the essentially equal and opposite effect of fluorine substituted in the 2 and 6 position of NE and the lack of adrenergic agonist specificities shown by the corresponding fluorodopamines.[2b] Concern over this formulation arises from the apparent reluctance of 2-fluorobenzyl alcohol to form an intramolecular hydrogen bond.[8] Faced with the present evidence that fluorine substitution can remove a substantial amount of energy from the substrate–receptor binding interaction, we now consider an alternate proposal. The benzene ring itself is an excellent hydrogen bond acceptor, especially when bearing electron-donating substituents. In fact, it has been estimated that, in carbon tetrachloride solutions, 65% of benzyl alcohol is present in an intramolecularly hydrogen-bonded form.[9] It is attractive, however, to involve the benzylic hydroxyl group in bonding interactions with the receptor.[5] On the other hand, intermolecular bonding interactions (e.g., dipole–dipole) from the receptor protein directed to the 2 and 6 position of the aromatic ring could, indeed, be affected by the electron-withdrawing effects of fluorine substitution. In this formulation, binding of the catecholamine to the α-adrenergic receptor would involve a bonding interaction between a positive center on the receptor and the 2 position of the ligand. The β-adrenergic receptor would have a similar interaction directed toward the 6 position. Fluorine would determine specificities by blocking this interaction through electron withdrawal from the carbon to which it is attached.

This proposal implies that the aromatic binding site is essential for agonist activity and that the receptor–aro-

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matic ring interaction differs sterically on α- and β-adrenergic receptors. An added role of the catechol hydroxyl group would be to increase the electron density of the aromatic ring, thus increasing the strength of this interaction.

The lack of adrenergic agonist specificities of 2- and 6-fluorodopamine remains to be explained. The absence of the benzylic hydroxyl group renders DA (and the FDA's) relatively weak adrenergic agonists. It is reasonable to assume that fluorine-induced specificities might be evident only when the benzylic hydroxyl group is also present to induce greater structural rigidity to the receptor–substrate complex.

We have several projects underway to investigate further the mechanism of fluorine-induced adrenergic agonist specificities. These include preparation of additional fluorinated and polyfluorinated adrenergic agonists and antagonists. Other substituent effects are being examined, and molecular orbital calculations are in progress to determine the effect of fluorine on the electron density distributions in fluorocatecholamines.

## Experimental Section

Melting points (uncorrected) were obtained with a Thomas-Hoover capillary melting point apparatus. Identities of all compounds were verified by chemical-ionization mass spectrometry (Finnigan, Model 1015D) where, in all cases, the M + 1 peak was in agreement with the calculated molecular weights. Reactions were monitored by thin-layer chromatography on silica gel GF plates (Analtech). Microanalysis, NMR spectra (JEOL-FX100, 100 MHz), and mass spectra were provided by the Section on Microanalytical Services and Instrumentation of the Laboratory of Chemistry, NIADDK, under the direction of Dr. David F. Johnson. Yields and physical data are summarized in Table I. NMR spectra for **2a–c** are given in Table II.

***N*-[2-(3,4-Dimethoxy-2-fluorophenyl)-2-hydroxyethyl]isopropylamine (4).** A 300-mg sample of 2-fluoro-3,4-dimethoxyphenethanolamine[1] dissolved in 3 mL of methanol and 5 mL of acetone was stored for 24 h at room temperature. Platinum oxide (40 mg) was hydrogenated in 3 mL of ethanol for a few minutes. To this suspension was added the amino alcohol–acetone reaction mixture, and the solution was hydrogenated (30–40 psi) for 5 h. After the catalyst was removed by filtration, the solvent was evaporated to give crystalline 4, 247 mg after recrystallization from cyclohexane.

**2-Fluoroisoproterenol (2a) Hydrochloride.** To 100 mg of 4 dissolved in 3 mL of methylene chloride and cooled to dry ice temperature under an argon atmosphere was added, with stirring, 0.3 mL of boron tribromide. The solution was allowed to warm to room temperature and was stirred for 24 h. The methylene chloride and excess boron tribromide were removed in vacuo, and the residue was cooled in dry ice and was treated with 10 mL of water. The solution was then warmed to room temperature, stirred for 1 h, and then applied to a Dowex $H^+$ ion-exchange resin. Elution with water (50 mL), 0.5 N hydrochloric acid (50 mL), and 3 N hydrochloric acid gave 51 mg of 2-FISO HCl (free of inorganic residue) (**2a**·HCl), homogenous on the basis of NMR, mass spectrum, HPLC, and TLC. All attempts to crystallize **2a**·HCl were unsuccessful.

**2-Fluoro-3,4-dihydroxybenzaldehyde (6a).** To 320 mg (1.74 mmol) of 2-fluoroveratraldehyde dissolved in 5 mL of methylene chloride and cooled to −78 °C was added in one portion, with stirring, 0.5 mL (5.2 mmol) of boron tribromide. The solution was stirred at room temperature overnight and then was cooled in ice and treated with 10 mL of water. The reaction mixture was extracted with ether, and the ether was extracted with 25 mL of 2 N NaOH. The alkaline extract was neutralized with dilute HCl and extracted with ether. The extract was dried, the ether was evaporated, and the residue was sublimed to give 175 mg of **6a**. For the preparation of **7a**, an alternative procedure was used to work up the demethylation reaction of **5a**, during which **6a** was not isolated (see below).

**2-Fluoro-3,4-bis(benzyloxy)benzaldehyde (7a).** 2-Fluoroveratraldehyde, 1.89 g (10.2 mmol), was demethylated as above. The methylene chloride solution was stirred overnight, chilled, and treated with excess absolute methanol. The solution was then evaporated under vacuum to remove solvent and trimethyl borate. Methanol was added, and the solution was evaporated again to remove traces of trimethyl borate. The residue was dissolved in 20 mL of absolute methanol, 5 g of Dowex $H^+$ was added, and the mixture was stirred overnight to complete acetal formation. The Dowex resin was removed by filtration, and then 3.0 g of potassium carbonate, 2.6 mL of benzyl chloride, and 25 mg of potassium iodide were added. After the solution had been refluxed for 24 h, water was added (25 mL), and the methanol was removed by evaporation. The aqueous layer was extracted with ethyl acetate (3 × 25 mL), and the ethyl acetate layer was shaken with 1 N HCl (3 × 25 mL). After each acid treatment, approximately 30 min was allowed for acetal hydrolysis before separation of the layers. The ethyl acetate was dried ($Na_2SO_4$) and evaporated to give 1.53 g of crystalline **7a**.

**3,4-Bis(benzyloxy)-2-fluorophenethanolamine (3a).** The ethanolamine side chain was elaborated through zinc iodide catalyzed addition of trimethylsilyl cyanide (TMSCN) to the aldehyde, followed by lithium aluminum hydride reduction, as described for the preparation of **3c**.[1] Thus, a mixture of 960 mg (2.68 mmol) of **7a**, 0.4 mL of trimethylsilyl cyanide, and a few crystals of $ZnI_2$ was stirred overnight. After removal of excess TMSCN (vacuum), reduction with 400 mg of lithium aluminum hydride gave 746 mg (2.03 mmol) of **3a**.

***N*-[2-[3,4-Bis(benzyloxy)-2-fluorophenyl]-2-hydroxyethyl]isopropylamine (8a).** A solution of 300 mg (0.82 mmol) of **3a** in 3 mL of absolute ethanol and 5 mL of acetone was stored overnight at room temperature. The solvent was removed by rotary evaporation (room temperature), the oily residue was dissolved in 5 mL of dry methanol, and the solution was cooled in an ice bath. To this solution was added, in portions and with stirring, a solution of 200 mg of $NaBH_4$ in cold methanol. The solution was allowed to warm as reduction and decomposition of borohydride proceeded and was then heated for 0.5 h on a steam bath. The solution was then cooled in ice, an additional 50 mg of $NaBH_4$ was added, and the reaction was allowed to warm spontaneously for 0.5 h. Water was added, and the solution was concentrated by rotary evaporation. The resulting precipitate was removed by filtration, dried, and recrystallized from cyclohexane to give pure **8a**. The presence of the isopropyl group was verified by the appearance of a doublet at 1.04 ppm ($J = 6.5$ Hz) in the 100-MHz NMR spectrum of **8a** ($CDCl_3$).

**2-Fluoronorepinephrine (1) Oxalate.** A solution of 300 mg (0.82 mmol) of **3a** and 107 mg (0.84 mmol) of oxalic acid dihydrate in 30 mL of methanol was hydrogenated at 20 psi over 50 mg of 10% Pd/C for 2 h. Removal of the catalyst by filtration and evaporation of solvent gave **1a**, oxalate, 180 mg after recrystallization from water/acetone. The salt was obtained as the neutral oxalate.

**2-Fluoroisoproterenol (2a) Oxalate.** Hydrogenolysis of **8a** (200 mg, 0.49 mmol), as described for the synthesis of **1a**, oxalate, gave 44 mg (0.15 mmol) of crystalline **2a**, oxalate, obtained as the neutral salt from acetonitrile/water.

**3,4-Dihydroxy-5-fluorobenzaldehyde (6b).** 5-Fluoroveratraldehyde was demethylated as described for the preparation of **6a**. From 1 g (5.43 mmol) there was obtained, after sublimation, 520 mg (3.33 mmol) of **6b**.

**3,4-Bis(benzyloxy)-5-fluorobenzaldehyde (7b).** As above, **7b** was prepared from 5-fluoroveratraldehyde without isolation of **6b**. From 980 mg of **5b** (5.32 mmol) there was obtained 859 mg (2.55 mmol) of **7b**, after recrystallization from ether/petroleum ether.

**3,4-Bis(benzyloxy)-5-fluorophenethanolamine (3b).** Addition of trimethylsilyl cyanide to **7b**, followed by hydride reduction, was carried out as above. From 200 mg (0.59 mmol) of **7b** there was obtained, after recrystallization from cyclohexane/ether, 194 mg (0.53 mmol) of **3b**.

***N*-[2-[3,4-Bis(benzyloxy)-5-fluorophenyl]-2-hydroxyethyl]isopropylamine (8b).** Reductive alkylation was carried out as described for the preparation of **8a**. From 200 mg (0.54 mmol) of **3b** there was obtained 150 mg (0.36 mmol) of **8b** after recrystallization from cyclohexane. The presence of the isopropyl group was evident from the 100-MHz NMR spectrum ($CDCl_3$): δ 1.06 (d, $J = 6.5$ Hz).

**5-Fluoroisoproterenol (2b) Oxalate.** Hydrogenolysis of 125 mg (0.31 mmol) of **8b** in 25 mL of methanol in the presence of 23 mg (0.38 mmol) of oxalic acid dihydrate as in the preparation of **1a** oxalate gave, after recrystallization from acetonitrile/water, 30 mg (0.13 mmol) of **2b** oxalate.

**6-Fluoroisoproterenol (2c) Oxalate.** 4,5-Bis(benzyloxy)-2-fluorophenethanolamine (**3c**; 300 mg, 0.81 mmol) was reductively alkylated as above to give 178 mg (0.43 mmol) of **8c**: NMR ($CDCl_3$) δ 1.06 (d, $J$ = 6.5 Hz). Catalytic hydrogenolysis of 178 mg (0.43 mmol) of **8c** in the presence of oxalic acid dihydrate (58 mg, 0.46 mmol) gave, after recrystallization from ethanol, 27 mg (0.1 mmol) of **2c** oxalate.

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# Synthesis, X-Ray Crystallographic Determination, and Opioid Activity of *erythro*-5-Methylmethadone Enantiomers. Evidence Which Suggests That μ and δ Opioid Receptors Possess Different Stereochemical Requirements[1]

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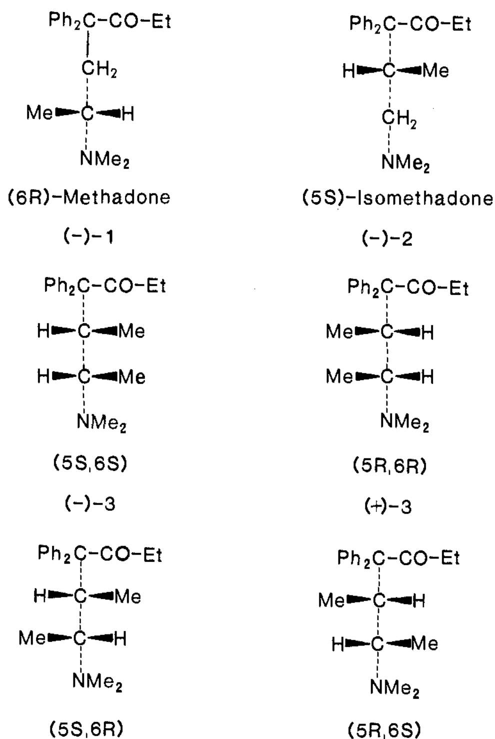
Enantiomers of *erythro*-5-methylmethadone (**3**) were synthesized from optical antipodes of *erythro*-3-(dimethylamino)-2-butanol. X-ray crystallographic analysis of (−)-**3** perchlorate revealed that it possesses the 5*S*,6*S* absolute configuration. It was found that (−)-**3** is substantially more potent than its enantiomer (+)-**3** as an opioid agonist in vivo and in vitro. In vitro tests (guinea pig ileal longitudinal muscle and mouse vas deferens preparations) suggest that (−)-**3** mediates its effect chiefly through μ opioid receptors. On the other hand, (+)-**3** and the more potent enantiomers of methadone, (−)-**1**, and isomethadone, (−)-**2**, appear to have less μ-receptor selectivity and interact with a greater fraction of δ receptors than does (−)-**3**. The fact that the solid-state conformation of (−)-**3** differs from that of (−)-**1** and (−)-**2**, which show great similarity in conformational features, suggests that μ and δ receptors have different conformational requirements. The possibility of different modes of interaction with a single opioid receptor population also is discussed.

In an effort to investigate the stereochemical relationship between the more potent enantiomers of methadone [(−)-**1**]




and isomethadone [(−)-**2**], we have previously synthesized two diastereomeric racemates of 5-methylmethadone (**3** and **4**) and evaluated them for antinociceptive activity in mice.[2] The threo racemate **4** contains an enantiomer (5*S*,6*R*) which can be considered a hybrid of (−)-**1** and (−)-**2** by virtue of possessing the same absolute configuration at common chiral centers. It was found that the threo racemate **4** is totally devoid of antinociceptive activity, while the erythro racemate **3** is approximately 5-fold more potent than methadone, (±)-**1**.

Since the active racemate (±)-**3** cannot contain the same combination of configurations found in (−)-**1**[3] and (−)-**2**[4] by virtue of its erythro stereochemistry, it was concluded[2] that each chiral center does not behave independently but rather interacts intramolecularly to afford a conformational population which facilitates the ligand–receptor recognition process.

We now report on the preparation, absolute configuration, and activity of *erythro*-5-methylmethadone enantiomers [(−)- and (+)-**3**] in an attempt to sort out the contributions of configurational and conformational factors in the ligand–receptor recognition process.

**Chemistry.** Attempts to resolve the erythro racemate (±)-**3** or its immediate precursor, methyl *erythro*-2,2-diphenyl-3-methyl-4-(dimethylamino)valerate, with a variety of resolving agents were unsuccessful. Ultimately, resolution of *erythro*-3-(dimethylamino)-2-butanol[5,6] (through

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