use of a Beckman gamma automatic counter (Model 5500). The percent dose per organ was calculated by a comparison of the tissue counts to suitably diluted aliquots of the injected material. Total activities of blood and muscle were calculated assuming that they are 7% and 40% of total body weight, respectively.[39]

Regional brain distribution in rats was obtained after an iv injection of *R*-(+)- or *S*-(−)-[^{125}I]-**8**. By dissecting, weighing, and counting samples from different brain regions (cortex, striatum, and cerebellum), % dose/gram of samples was calculated by comparing the sample counts with the counts of the diluted initial dose. The uptake ratio of each region was obtained by dividing % dose/gram of each region with that of the cerebellum.

In Vitro Binding. Rat tissue homogenates were prepared as described previously.[31] The binding assays were performed by incubating 50 μL of tissue preparations containing 40–60 μg of protein with appropriate amounts of *R*-(+)-[^{125}I]-**8** ligand (for saturation analysis) or appropriate amounts of ligand and competitors (for competition study) in a total volume of 0.2 mL of the assay buffer. After an incubation period of 20 min at 37 °C (with stirring), the samples were rapidly filtered in the cell harvester (Brandel M-24R), under vacuum, through Whatman GF/B glass fiber filters pretreated with 0.2% protamine sulfate and washed with 3 × 5 mL of cold (4 °C) 50 mM Tris-HCl buffer, pH 7.4. The nonspecific binding was obtained in the presence of 10 μM SCH-23390. The filters were counted in a gamma counter (Beckman 5500) at an efficiency of 70%. Both Scatchard and competition experiments were analyzed by using the iterative nonlinear least-squares curve-fitting program LIGAND.[40]

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Registry No. **2**, 55-81-2; **3**, 131567-06-1; (±)-**4**, 131567-07-2; (±)-**4**·maleate, 131567-08-3; *R*-(+)-**4**, 131615-49-1; *S*-(−)-**4**, 131615-55-9; (*R*,*S*)-**5**, 131681-39-5; (*S*,*R*)-**5**, 131681-40-8; (±)-**6**·maleate, 131567-10-7; (±)-**6**, 131567-09-4; *R*-(+)-**6**, 131615-50-4; *S*-(−)-**6**, 131615-56-0; (±)-**7**, 131567-13-0; *R*-(+)-**7**, 131615-51-5; *S*-(−)-**7**, 131681-51-1; (±)-**8**, 131567-14-1; *R*-(+)-**8**, 131615-52-6; *S*-(−)-**8**, 131615-57-1; (±)-**9**, 131567-12-9; *R*-(+)-**9**, 131615-54-8; 3-chloro-4-methoxyphenethylamine, 7569-87-1; 3-bromostyrene oxide, 131567-05-0; (±)-7-chloro-8-hydroxy-3-methyl-1-(3′-bromophenyl)-2,3,4,5-tetrahydro-1*H*-3-benzazepine, 131567-11-8; (*R*)-7-chloro-8-methoxy-3-methyl-1-(3′-iodophenyl)-2,3,4,5-tetrahydro-1*H*-3-benzazepine, 131567-15-2; (*R*)-7-chloro-8-hydroxy-3-methyl-1-(3′-bromophenyl)-2,3,4,5-tetrahydro-1*H*-3-benzazepine, 131615-53-7; (*R*)-(+)-7-chloro-8-methoxy-3-methyl-1-phenyl-2,3,4,5-tetrahydro-1*H*-3-benzazepine, 129666-34-8; (*S*)-(−)-7-chloro-8-methoxy-3-methyl-1-phenyl-2,3,4,5-tetrahydro-1*H*-3-benzazepine, 73445-62-2; (±)-7-chloro-8-methoxy-3-methyl-1-(3′-iodophenyl)-2,3,4,5-tetrahydro-1*H*-benzazepine, 131615-58-2; (*S*)-(−)-7-chloro-8-methoxy-3-methyl-1-(3′-iodophenyl)-2,3,4,5-tetrahydro-1*H*-benzazepine, 131567-16-3.

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Synthesis and Ligand Binding of Cocaine Isomers at the Cocaine Receptor

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The cocaine binding site at the dopamine transporter has been found to be stereoselective. Thus, the seven possible stereoisomers of (−)-cocaine have been synthesized and found to inhibit [^{3}H]-2β-carbomethoxy-3β-(4-fluorophenyl)tropane ([^{3}H]WIN 35,428) with potencies ranging from $^1/_{60}$ to $^1/_{600}$ of that of (−)-cocaine. The synthesis and characterization of all new compounds is presented.

Specific binding sites for natural (−)-cocaine have been identified in the brain tissue of both rodents and humans.[1-5] Affinities of (−) natural cocaine and several cocaine-like compounds at one of these binding sites parallel their potencies for inhibiting dopamine uptake as well as their potencies for producing drug selfadministration.[6-8] Thus this binding site has several properties characteristic of biologically relevant receptors. However, since biologically relevant receptors usually show stereoselectivity, it is important to establish the stereoselectivity of (−)-cocaine for its binding site. Since cocaine has seven additional stereoisomeric forms, it is necessary to determine whether any of these cocaine isomers show high affinity for the cocaine binding sites. In this report we describe the synthesis of the seven cocaine stereoisomers and present their binding properties at the cocaine binding site(s).

Results

The chemical entity methyl 3-(benzoyloxy)-8-methyl-8-azabicyclo[3.2.1]octane-2-carboxylate can exist in eight stereoisomeric forms (Chart I); one of these, [1*R*-(*exo*,*exo*)],[9] refers to natural (−)-cocaine (**1**). Since most of the biological studies reported for cocaine use the traditional names for these compounds, the more familiar traditional names are used in this report. However, all structures derivable from the natural and unnatural cocaine ring system are designated by either an *R* or an *S* prefix, as appropriate. Thus natural cocaine and its three *R* diastereomers are designated (*R*)-cocaine (**1**), (*R*)-pseudococaine (**2**), (*R*)-allococaine (**3**), and (*R*)-allopseudococaine (**4**) while the isomers deriving from the enantiomer of natural cocaine are (*S*)-cocaine (**5**), (*S*)-pseudococaine (**6**),

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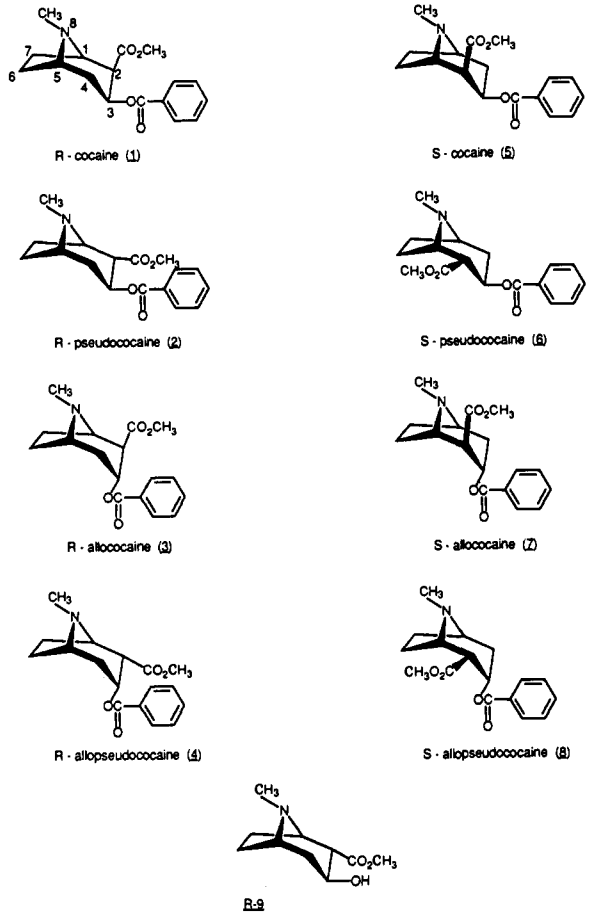
Chart I



(*S*)-allococaine (**7**), and (*S*)-allopseudococaine (**8**).

Synthesis

Since the axial 2-carbomethoxy group in (*R*)-cocaine is known to epimerize to the equatorial position, saponification of (*R*)-cocaine (**1**) using sodium methoxide in anhydrous methanol was used to obtain (*R*)-pseudoecgonine methyl ester (*R*-**9**).[10] Benzoylation of *R*-**9** with benzoyl chloride in pyridine gave (*R*)-pseudococaine (**2**).[11]

(*R*)-Allococaine (**3**) and (*R*)-allopseudococaine (**4**) were prepared from (*RS*)-2-carbomethoxy-3-tropinone (*RS*-**10**) as shown in Scheme I. Resolution of *RS*-**10** using (+)- and (−)-tartaric acid provided (*R*)-2-carbomethoxy-3-tropinone (*R*-**10**) and (*S*)-2-carbomethoxy-3-tropinone (*S*-**10**).[11,12-14] Sodium borohydride reduction of *R*-**10** in methanol at −20 °C, as described for *RS*-**10** by Carroll et al.,[15] provided (*R*)-allopseudoecgonine methyl ester *R*-**11**, which on benzoylation gave (*R*)-allopseudococaine (**4**). When *R*-**11** was heated in water, isomerization at the 2-position occurred along with hydrolysis to give (*R*)-alloecgonine *R*-**12**.[16] Fisher esterification of *R*-**12** using methanol and hydrogen chloride gave (*R*)-alloecgonine methyl ester (*R*-**13**), which

Scheme I

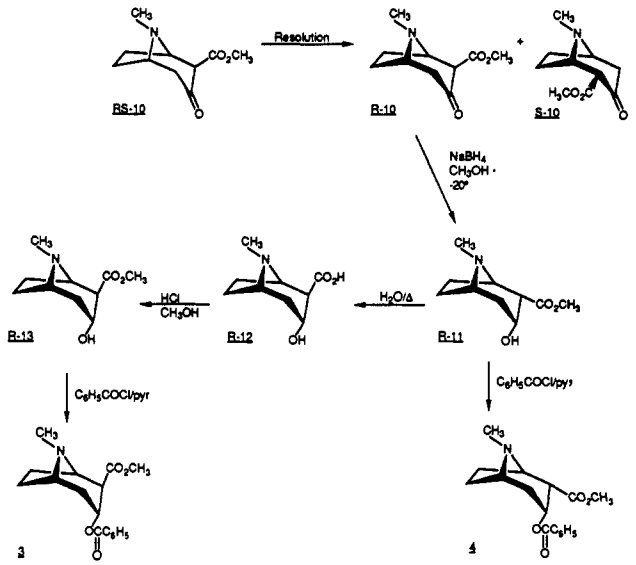


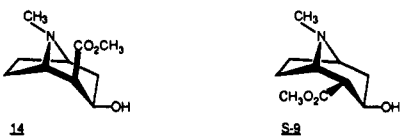
Table I. Potencies of Cocaine and Isomers in Inhibition of [^{3}H]WIN 35,428 Binding to Rat Striatal Membranes

compd	IC_{50},[a] μm	Hill coeff	% potency obsd	% potency calcd
1	0.102	0.91	100	100
2	15.80	0.96	0.65	
3	6.16	2.81	1.66	
4	28.50	0.92	0.36	
5	15.8	1.19	0.65	
6	22.5	1.30	0.45	0.42
7	9.82	2.60	1.04	1.08
8	67.7	0.95	0.15	0.23

[a] All values are the mean of 4 or 5 experiments performed in triplicate. The standard error of mean is less than 5% in all cases.

was benzoylated to give (*R*)-allococaine (**3**).

(*S*)-Cocaine (**5**) and (*S*)-pseudococaine (**6**) were prepared as previously reported.[11] Thus the reduction of *S*-**10** with sodium amalgam provided (*S*)-ecgonine methyl ester (**14**)



and (*S*)-pseudoecgonine methyl ester (*S*-**9**), which were separated by chromatography and benzoylated with benzoyl chloride in pyridine to give (*S*)-cocaine (**5**) and (*S*)-pseudococaine (**6**), respectively.[11] (*S*)-Allococaine (**7**) and (*S*)-allopseudococaine (**8**) were prepared from *S*-**10** by procedures exactly analogous to those shown for the *R* isomers in Scheme I.

The structures of compounds **1**–**8** were confirmed by their characteristic ^{1}H NMR spectra.[15] Since all four diastereomers of each cocaine enantiomer are separable by TLC, isomeric purity was determined by TLC analysis. The optical rotations of (*S*)-cocaine (**5**) and (*S*)-pseudococaine (**6**) in the positive direction were of equal magnitude to those of natural (*R*)-cocaine (**1**) and of (*R*)-pseudococaine (**2**), and are in agreement with the literature values. This observation serves to establish their optical purity, as well as the optical purity of (*S*)-2-carbomethoxy-3-tropinone (*S*-**10**). Since the new enantiomers, **3**, **4**, **7**, and **8**, were prepared from pure *R*-**10** and *S*-**10**, respectively, they should also be optically pure. Their observed rotations, of equal magnitude but opposite direction, confirm their optical purities.

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Receptor Binding

The effects of (*R*)-cocaine (1) and its seven isomeric forms on radioligand binding of [^{3}H]-2β-carbomethoxy-3β-(4-fluorophenyl)tropane (**15**)[17] at the rat striatal dopamine transporter are listed in Table I. The potency of (*R*)-cocaine is 60–670 times greater than that of its isomers. The most potent isomers of (*R*)-cocaine are the two allo isomers, **3** and **7**, with IC_{50} values of 6.16 and 9.82 μm, respectively. Moreover, in contrast to the other six, which have Hill coefficients close to unity, the isomers **3** and **7** showed Hill coefficients of 2.81 and 2.60, respectively.

Discussion

The observation that (*R*)- and (*S*)-pseudococaine (**2** and **6**, respectively) and (*S*)-cocaine (**5**) possess low affinity for the [^{3}H]mazindol binding site of the dopamine transporter[18] prompted investigation of the effects of stereochemistry on binding affinity. The ligand of choice was **15**, in view of the recent report that **15** binding correlates with that of [^{3}H]mazindol while showing high affinity, greater specificity, and better specific-to-nonspecific binding ratio than [^{3}H]mazindol.[17] The potencies of (*R*)-cocaine (1) and its seven isomers, **2–8** (Table I), to inhibit **15** binding demonstrate that the cocaine binding site is indeed stereoselective. Thus inversion of the overall configuration (compare **1** and **5**) reduces the binding affinity to 0.65%, as does epimerization at C-2 (compare **1** and **2**). Smaller effects are associated with epimerization at C-3. Specifically, the C-3 epimer of (*R*)-cocaine (**1**), (*R*)-allococaine (**3**), is 1.66% as potent as **1**, and the (*R*)-allopseudo epimer **4** has a potency 0.36% that of **1**. The potency of the isomers in the *S* series mirrors this trend. In fact, the potencies of **6–8** are predicted reasonably well by multiplying the normalized potencies of each *R* isomer by the normalized potency of (*S*)-cocaine (**5**) (Table I). Although the small number of compounds does not warrant wide ranging conclusions to be drawn, this approach to structure/activity correlation, while apparently undocumented, appears to be valid for stereochemical changes, as in this series. The correlation does not apply for prediction of the potencies of the allopseudo isomers **4** and **8** from the potencies of the pseudo isomers **2** and **6** and the allo isomers **3** and **7**. This is probably related to the observation that the allo isomers possess aberrant Hill coefficients. Thus the allo isomers may inhibit the binding of **15** by a different mechanism, perhaps involving noncompetitive binding. Since the allopseudo isomers have Hill coefficients close to unity, their potency is unlikely to be influenced by factors determining the potency of allo isomers. Consequently, the potency of the allo isomers cannot be used to predict the potency of the allopseudo isomers.

The psychostimulant drug (*R*)-cocaine (**1**) has been shown to inhibit the transport of dopamine. In previous studies we had presented evidence strongly suggesting that the [^{3}H]mazindol binding site of the dopamine transporter was related to the cocaine binding site associated with the reinforcing properties of (*R*)-cocaine and related drugs.[6,7] Structural requirements for (*R*)-cocaine binding to this site had been found to include the presence of an aryl group connected either directly or indirectly to C-3 and of an ester group at C-2, both in β orientation.[18,19] The present data extend these observations. Since all seven stereoisomers of (*R*)-cocaine show a 60-fold or greater reduction in potency for inhibiting **15** binding, it is now possible to claim true stereoselectivity for the binding site of (*R*)-cocaine at the dopamine transporter.

Experimental Section

Melting points were determined on a Thomas-Hoover capillary tube apparatus. All optical rotations were determined at the sodium D line with a Rudolph Research Autopol III polarimeter (1-dm cell). NMR spectra were recorded on a Bruker WM-250 spectrometer using tetramethylsilane as an internal standard. Thin-layer chromatography was carried out on Whatman silica gel 60 TLC plates using $CHCl_3$–MeOH–concentrated NH_4OH (40:9:1) unless otherwise noted. Visualization was accomplished under UV or in an iodine chamber. Microanalyses were carried out by Atlantic Microlab, Inc.

Resolution of (*RS*)-2-Carbomethoxy-3-tropinone (*RS*-10). (*RS*)-2-Carbomethoxy-3-tropinone[11] (*RS*-**10**) was resolved with a modification of a reported procedure.[11,14] Treatment of *RS*-**10** (31 g, 0.14 mol) with 10% excess (23.9 g, 0.16 mol), in EtOH (300 mL), of (–)-tartaric acid gave a hydrogen tartrate salt (35 g) which was recrystallized once from 10:1 Me_2CO–water (1200 mL) mixture and once from MeOH to give (*S*)-2-carbomethoxy-3-tropinone (*S*-**10**) (–)-hydrogen tartrate as a pale yellow crystalline solid (9.5 g, 37% yield) with mp 158–159 °C (lit.[12] mp 159.5 °C) and $[\alpha]^{20}_D$ –16.8° (*c* 2, H_2O) [lit.[12] $[\alpha]^{24}_D$ –16.9° (*c* 2, H_2O)]. The salt was dissolved in aqueous Na_2CO_3 (pH 8), and the free base generated was extracted with CH_2Cl_2. The dried (Na_2SO_4) extracts were concentrated to give *S*-**10** as a white solid: mp 104–105 °C (lit.[12] mp 108.6–109.6 °C); $[\alpha]^{25}_D$ –25.7° (*c* 1, MeOH) [lit.[12] $[\alpha]^{20}_D$ –18.3° (*c* 1, MeOH)].

The mother liquors from above were evaporated to dryness, dissolved in water, adjusted to pH 8 with Na_2CO_3, and extracted with CH_2Cl_2. The dried (Na_2SO_4) extracts were evaporated to give a tan solid. Treatment of the solid with excess (+)-tartaric acid gave a salt which was recrystallized and converted to the free base as described for the *S* isomer. The salt had mp 158–159 °C and $[\alpha]^{20}_D$ +16.9° (*c* 2, H_2O). The (*R*)-2-carbomethoxy-3-tropinone obtained had mp 108–109 °C (lit.[12] mp 108.5–109.5 °C) and $[\alpha]^{25}_D$ +25.4° (*c* 1, MeOH); [lit.[12] $[\alpha]^{20}_D$ +18.3° (*c* 1, MeOH)].

(*R*)- and (*S*)-Pseudoecgonine Methyl Ester (9) and (*R*)- and (*R*)-Allopseudoecgonine Methyl Ester (11). To a solution of the appropriate isomer of **10** in MeOH (35 mL/mol) at –78 °C was added $NaBH_4$ (2 mol/mol of **10**) in small portions over a 30-min period. The mixture was kept at –20 °C in the freezer for 24 h. The excess $NaBH_4$ was carefully destroyed with concentrated HCl. Water was added to form a clear solution, and the pH was adjusted to 11 with concentrated NH_4OH. The free base was extracted with CH_2Cl_2. The organic phase was washed with saturated NaCl solution and dried (Na_2SO_4). The residue obtained after removal of the solvent under reduced pressure was chromatographed on SiO_2. Elution with a solvent mixture of CH_2Cl_2–$CHCl_3$–CH_3OH–NH_4OH (100:40:9:1) gave pseudoecgonine methyl ester (**9**) in the first fraction and allopseudoecgonine methyl ester (**11**) in later fractions.

Thus 5 g (0.023 mol) of *R*-**10** gave 0.472 g (10%) of (*R*)-pseudoecgonine methyl ester (*R*-**9**) [mp 113–114 °C (lit.[10] mp 114–116 °C); $[\alpha]^{23}_D$ +23.1° (*c* 1, H_2O) [lit.[10] $[\alpha]$ +22.8° (*c* 1, H_2O)]] and 2.37 g (52%) of (*R*)-allopseudoecgonine methyl ester (*R*-**11**) [mp 78–79 °C (from hexane); $[\alpha]^{23}_D$ –36.8° (*c* 1, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 2.31 (s, 3 H, NCH_3), 3.75 (s, 3 H, OCH_3), 4.27 (bs, 1 H, H3). Anal. Calcd for $C_{10}H_{17}NO_3$: C, H, N].

S-**10** (2.15 g, 0.011 mol) gave 0.407 g (19%) of (*S*)-pseudoecgonine methyl ester (*S*-**9**) [mp 114–115 °C (from Et_2O) (lit.[11] mp 114–115 °C); $[\alpha]^{20}_D$ –23.3° (*c* 1, H_2O) [lit.[11] $[\alpha]^{20}_D$ –22.5° (*c* 1, H_2O)]; ^{1}H NMR ($CDCl_3$) δ 2.39 s, 3 H, NCH_3), 3.73 (s, 3 H, OCH_3, 4.10 (m, 1 H, H3)] and 1.23 g (56%) of (*S*)-allopseudoecgonine methyl ester (*S*-**11**) [mp 79–80 °C (from hexane); $[\alpha]^{20}_D$ +37.7° (*c* 1, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 2.32 (s, 3 H, NCH_3), 3.75 (s, 3 H, OCH_3), 4.28 (t, 1 H, H3). Anal. Calcd for $C_{10}H_{17}NO_3$: C, H, N].

(*R*- and *S*)-Alloecgonine (12). A solution of the appropriate allopseudoecgonine methyl ester (**11**) in water (2.7 mL/mmol) was refluxed for 9 h. The reaction mixture was evaporated to dryness and dried under reduced pressure. The resulting waxy residue was slurried with $CHCl_3$ (15 mL mmol) for 72 h. The

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insoluble material was collected by filtration, washed with $CHCl_3$, and dried under reduced pressure to give alloecgonine.

Thus 2.03 g (0.01 mol) of (*R*)-allopseudoecgonine methyl ester (*R*-11) gave 1.39 g (74%) of (*R*)-alloecgonine (*R*-12): mp 208–209 °C dec (from EtOH); $[\alpha]^{20}_D$ −47.6° (*c* 1, H_2O); ^1H NMR ($CDCl_3$–CD_3OH) δ 2.75 (s, 3 H, NCH_3), 4.08 (d, 1 H, H3). Anal. Calcd for $C_9H_{15}NO_3$: C, H, N.

A 0.75 g (3.8 mmol) sample of (*S*)-allopseudoecgonine methyl ester (*S*-11) gave 0.49 g (70%) of (*S*)-alloecgonine (*S*-12): mp 207–208 °C (from EtOH); $[\alpha]^{20}_D$ +47.1° (*c* 1, H_2O); ^1H NMR ($CDCl_3$–CD_3OD) δ 2.75 (s, 3 H, NCH_3) 4.08 (d, 1 H, H3). Anal. Calcd for $C_9H_{15}NO_3 \cdot 0.5H_2O$: C, H, N.

(*R*)- and (*S*)-Alloecgonine Methyl Ester (13). A solution of alloecgonine (12) was stirred overnight in ~10% HCl(g) in MeOH (25 mL/mmol). After removal of the solvents under reduced pressure, the residue was dried under vacuum and recrystallized from MeOH–Et_2O to give the hydrogen chloride salt. Thus 1.29 g (7.0 mmol) of (*R*)-alloecgonine (*R*-12) gave 1.1 g (67%) of (*R*)-alloecgonine methyl ester hydrochloride (*R*-13·HCl): mp 190–191 °C; $[\alpha]^{20}_D$ −27.6° (*c* 1, MeOH); ^1H NMR ($CDCl_3$) δ 2.84 (s, 3 H, NCH_3), 3.83 (s, 3 H, OCH_3), 4.41 (d, 1 H, H3). Anal. Calcd for $C_{10}H_{18}ClNO_3$: C, H, N.

A 0.40 g (2.2 mol) sample of (*S*)-alloecgonine (*S*-12) gave 0.35 g (65%) of (*S*)-alloecgonine methyl ester hydrochloride (*S*-13·HCl): mp 189–190 °C; $[\alpha]^{20}_D$ +27.3° (*c* 1, MeOH); ^1H NMR ($CDCl_3$) δ 2.84 (s, 3 H, NCH_3), 3.83 (s, 3 H, OCH_3), 4.45 (d, 1 H, H3). Anal. Calcd for $C_{10}H_{18}ClNO_3$: C, H, N.

(*R*)- and (*S*)-Allococaine (3 and 7). (*R*)- and (*S*)-Allococaine were prepared following the procedure described by Lewin et al.[11] for the preparation of (+)-cocaine. Thus 0.712 g (3.6 mmol) of (*R*)-alloecgonine methyl ester (*R*-13) was treated with a solution of benzoyl chloride (0.52 g, 3.6 mmol) in 4 mL of pyridine at 0 °C and stirred at room temperature for 24 h. The solvent was removed under reduced pressure. The residue was treated with 50% NH_4OH solution. The aqueous mixture was extracted with CH_2Cl_2 and the organic extract dried (Na_2SO_4). The residue obtained after removal of the solvent under reduced pressure was chromatographed on SiO_2. On elution with 5% MeOH–CH_2Cl_2, 0.39 g (36%) of (*R*)-allococaine (3) was obtained: mp 81–82 °C (from petroleum ether); $[\alpha]^{23}_D$ −47.0° (*c* 1, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 2.24 (s, 3 H, NCH_3), 3.77 (s, 3 H, OCH_3), 5.66 (d, 1 H, H3), 7.44–8.04 (m, 5 H, aromatics). Anal. Calcd for $C_{17}H_{21}NO_4$: C, H, N.

A sample of 0.43 g (2.2 mmol) of (*S*)-alloecgonine methyl ester (*S*-13) and benzoyl chloride (0.31 g, 2.2 mmol) in 2 mL of pyridine gave 0.39 g (56%) of (*S*)-allococaine (7): mp 81–82 °C (from petroleum ether); $[\alpha]^{23}_D$ +47.9° (*c* 1, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 2.24 (s, 3 H, NCH_3), 3.77 (s, 3 H, OCH_3), 5.66 (d, 1 H, H3), 7.44–8.05 (m, 5 H, aromatics). Anal. Calcd for $C_{17}H_{21}NO_4$: C, H, N.

(*R*)- and (*S*)-Allopseudococaine Hydrochloride (4·HCl and 8·HCl). Allopseudoecgonine methyl ester hydrochlorides were prepared by dissolving the free base in dry methanolic HCl. The residue obtained after removal of the solvent under reduced pressure was dried under vacuum. The salt was placed in a test tube with 1.35 equiv of benzoyl chloride and heated on an oil bath at 150 °C. After 15 min, the solids melted with evolution of HCl gas, giving a homogeneous mixture. After an additional 5 min at 150 °C, the reaction mixture turned into a tan solid. The mixture was cooled, dissolved in MeOH, treated with charcoal, and filtered. The residue obtained after removal of solvent was recrystallized from MeOH/Et_2O. Thus 0.275 g (1.2 mmol) of (*R*)-allopseudoecgonine methyl ester (*R*-11) gave 0.225 g (55%) of (*R*)-allopseudococaine (4) hydrochloride: mp 184–185 °C; $[\alpha]^{25}_D$ −1.20 (*c* 1, MeOH); ^1H NMR ($CDCl_3$–CD_3OD) δ 2.86 (s, 3 H, NCH_3), 3.63 (s, 3 H, OCH_3), 5.78 (t, 1 H, H3), 7.47–7.92 (m, 5 H, aromatics). Anal. Calcd for $C_{17}H_{22}ClNO_4 \cdot 0.25H_2O$: C, H, Cl, N.

A 0.40 g (2.0 mmol) sample of (*S*)-allopseudoecgonine methyl ester (*S*-11) gave 0.25 g (37%) of (*S*)-allopseudococaine (8) hydrochloride: mp 183–184 °C; $[\alpha]^{23}_D$ +1.27° (*c* 1, MeOH); ^1H NMR ($CDCl_3$–CD_3OD) δ 2.85 (s, 3 H, NCH_3), 3.60 (s, 3 H, OCH_3), 5.80 (t, 1 H, H3), 7.44–7.91 (m, 5 H, aromatics). Anal. Calcd for $C_{17}H_{22}ClNO_4$: C, H, Cl, N.

Radioligand Binding Assays. Tissues for all binding experiments were dissected from the brains of male Sprague–Dawley rats, age 60–120 days old. The rats were sacrificed by decapitation; their brains were removed and washed in cold saline; the striata were then dissected, frozen, and stored at −70 °C until used in the assay procedures. Tissues were homogenized in 20 volumes of the assay buffer, then centrifuged at 30000*g* for 10 min. The resulting pellet was washed, recentrifuged, and resuspended in buffer to yield the desired tissue concentration for addition to the assay.

Compound 15, 0.5 nM final concentration, was used to label dopamine uptake sites in rat striatal tissue. Nonspecific binding was defined by the addition of 30 μM cocaine. Approximately 1.0 mg original weight of homogenized tissue was incubated for 2 h at 0 °C in buffer (10 mM sodium phosphate, 0.32 M sucrose, pH 7.4) with 0.5 nM final ligand concentration and 0.5 mL final assay volume.

At the end of the incubation period, assay mixtures were filtered through Whatman GF/B filters presoaked with 0.5% polyethylimine and washed with buffer. The filters were placed in plastic vials, scintillation fluid was added, and the vials were shaken for 1 h. Radioactivity in each was measured by liquid scintillation spectrometry.

IC_{50} values were determined from analyses of competition curves using the nonlinear least-squares, curve-fitting program EBDA. Mean values and standard errors were calculated for 4 or 5 assays for each test drug.

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Registry No. 1, 50-36-2; 2, 478-73-9; 3, 668-19-9; 4, 518-97-8; 4·HCl, 109582-53-8; 5, 47195-07-3; 6, 85354-45-6; 7, 131829-33-9; 8, 131722-66-2; 8·HCl, 131829-34-0; (*R*)-9, 65913-90-8; (*S*)-9, 112574-75-1; (*RS*)-10, 112574-77-3; (*R*)-10, 53955-90-1; (*R*)-10·(+)-tartrate, 131722-67-3; (*S*)-10, 112652-64-9; (*S*)-10·(−)-tartrate, 112708-65-3; (*R*)-11, 53955-92-3; (*S*)-11, 50373-09-6; (*R*)-12, 5878-35-3; (*S*)-12, 131722-68-4; (*R*)-13·HCl, 93738-78-4; (*S*)-13·HCl, 131722-69-5.