Further Studies on Oxygenated Tryptamines with LSD-like Activity Incorporating a Chiral Pyrrolidine Moiety into the Side Chain

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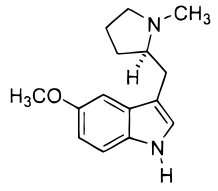
The enantiomers of 3-(*N*-methylpyrrolidin-2-ylmethyl)-5-methoxyindole, **1,** and 3-(*N*-methylpyrrolidin-2-ylmethyl)-4-hydoxyindole, **3**, were prepared using an asymmetric synthesis that employed (+)- or (−)-proline. A new approach was developed that had certain advantages over the synthesis originally reported for the isomers of **1**. (±)-3-(*N*-Methylpyrrolidin-3-yl)-4-hydroxyindole, **5**, was also prepared as a rigid analogue of psilocin and compared with its 5-methoxy counterpart **4**. Radioligand competition assays were used to assess the affinity of compounds for the 5-HT_{2A} receptor labeled with the agonist ligand [^{125}I]DOI and the antagonist ligand [^{3}H]MDL100907. Two-lever drug discrimination assays in rats trained to discriminate either LSD or DOI from saline were employed to assess the hallucinogen-like behavioral properties of these rigid tryptamine analogues. The receptor binding assay results clearly demonstrated a stereochemical preference for the *R* enantiomers that did not discriminate the position of the oxygen function. The receptor is 10−20-fold more selective for the *R* isomers. The affinities of the *R* enantiomers were virtually identical for both **1** and **3** at the agonist-labeled receptor, while racemic **4** and **5** had about one-tenth the affinity. The drug discrimination data in both LSD- and DOI-trained rats paralleled the binding data using [^{125}I]DOI displacement. Both *(R)*-**1** and *(R)*-**3** are about equipotent, comparable to DOI in activity but about 10-fold less potent than LSD. Compound **4** produced only partial substitution, even at a dose nearly 5-fold higher than for (*R*)-**1**. Based on conformational energies, it seems doubtful that these compounds bind to the 5-HT_{2A} receptor in an ergoline-like conformation. The results also suggest that both **1** and **3** would possess LSD-like psychopharmacology in humans.

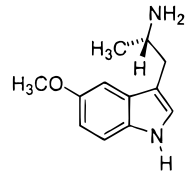
Introduction

A number of studies have examined conformational restriction of the 3-(2-aminoethyl) side chain of serotonin as an approach to 5-HT analogues with receptor subtype selectivity.[1−5] Introduction of a stereocenter α to the amine nitrogen of serotonin, while also conformationally restricting the C−N bond of the aminoethyl side chain by incorporating it into a pyrrolidine ring, affords compounds that serve as useful chiral probes of 5-HT receptors. Macor et al.[1] first reported that (*R*)-(+)-3-(*N*-methylpyrrolidin-2-ylmethyl)-5-methoxyindole, **1**, had approximately 30-fold higher affinity than the *S* enantiomer at 5-HT_{2A} receptors and was as efficacious as serotonin in stimulating phosphatidylinositol (PI) turnover in rat cortical slices.

For many years we have been exploring the structure−activity relationships of hallucinogens and other centrally active substances that interact with serotonin receptors. Because the 5-HT_{2A} receptor is believed to be the primary target for hallucinogenic agents, the report by Macor et al.[1] prompted us to study the analogous conformationally constrained 4-hydroxy compounds related to the naturally occurring hallucinogen psilocin. We were also curious as to whether transposing the oxygen substituent from the 5 to the 4 position in the tryptamines would have an effect on stereoselectivity at the 5-HT_{2A} receptor. We had previously found that the *S* isomer of 5-methoxy-α-methyltryptamine, **2**, had higher affinity than the *R* isomer at both the serotonin 5-HT_{1B} and 5-HT_2 sites.[6] This selectivity was reversed, however, for 4-oxygenated α-methyltryptamines at the 5-HT_{1B} site.



1



2

The present series of compounds was designed to elucidate further how constraining the side chain stereochemistry in 4-hydroxytryptamines affects activity at the serotonin 5-HT_{2A} receptor, compared with 5-methoxy-substituted analogues. Although the synthesis of the enantiomers of **1** is reported in the literature,[1] we describe here a new procedure for the preparation of this compound and its 4-hydroxy analogue **3.** The synthesis of **5** employed a Michael reaction between the appropriate indole and *N*-methylmaleimide, as described previously for **4**.[6] These compounds were evaluated for affinity at the 5-HT_{2A} receptor in rat prefrontal

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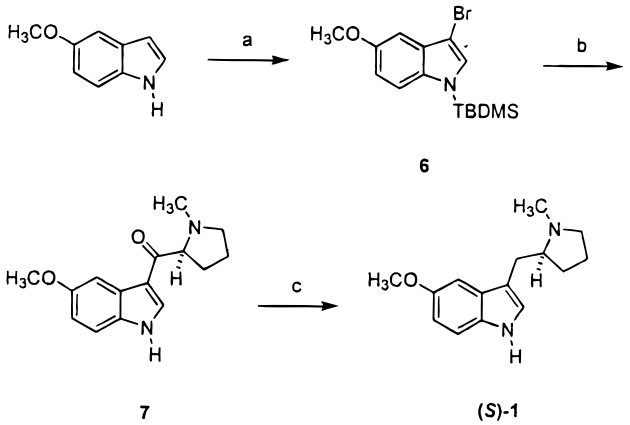
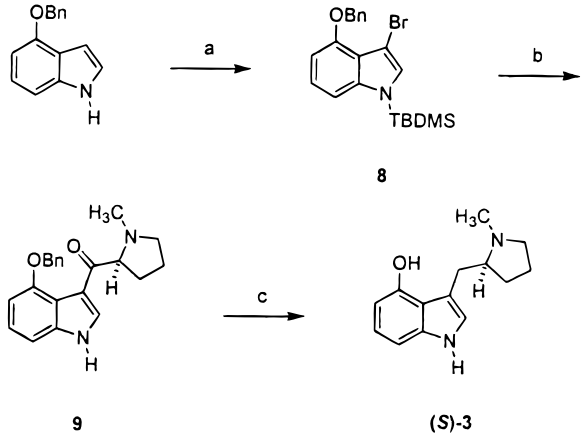
Scheme 1[a]



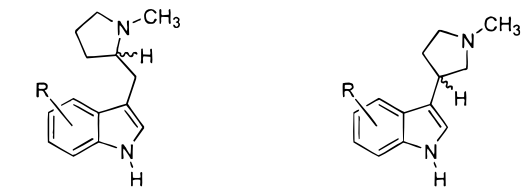
[a] (a) i. *n*-BuLi, THF, −78 °C, ii. *t*-BuMe$_2$SiCl, THF, 0 °C, iii. NBS, −78 °C; (b) i. *n*-BuLi, THF, −78 °C, ii. *N*-methylproline methyl ester,[10,11] THF, −78 °C; (c) LiAlH$_4$, THF, reflux.

Scheme 2[a]



[a] (a) i. NaH, THF, ii. *t*-BuMe$_2$SiCl, THF, 0 °C, iii. NBS, THF, −78 °C; (b) i. *n*-BuLi, THF, −78 °C, ii. *N*-methylproline methyl ester,[10,11] THF, −78 °C; (c) LiAlH$_4$, dioxane, reflux.

cortex homogenate. Compounds (*R*)-**1**, (*R*)-**3**, and (±)-**4** were evaluated in the drug discrimination assay in rats trained to discriminate saline from either DOI or LSD.



(*R*)/(*S*)-1; R = 5-OCH$_3$
(*R*)/(*S*)-3; R = 4-OH

(*R,S*)-4; R = 5-OCH$_3$
(*R,S*)-5; R = 4-OH

Chemistry

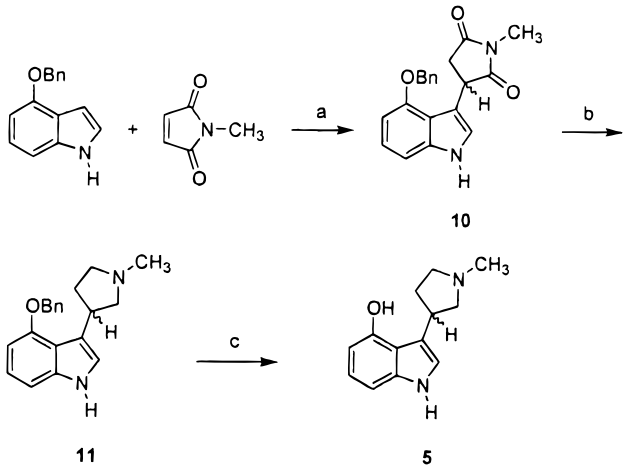
In the synthesis of (*R*)-**1** Macor et al.[1] reportedly utilized the condensation of *N*-carbobenzoxy-D-proline acid chloride and 2 equiv of the magnesium salt of 5-methoxyindole to append the conformationally restricted C-3 substituent with retention of stereochemical integrity. In our hands, this approach provided mixtures of products and unacceptably low yields. Unsuccessful attempts were made to affect the outcome of the condensation by replacing magnesium with a less electropositive metal (e.g. transmetalation with $ZnCl_2$), in order to lower the degree of dissociation of the nitrogen−metal bond, or by use of the Lewis acid catalyst $SnCl_4$. We next turned our attention to the chemistry of *N*-protected 3-lithioindoles, anticipating an enhanced reactivity of the 3 position in 5-methoxy- and 4-benzyloxyindoles. Amat et al.[7] reported preparation of 1-(*tert*-butyldimethylsilyl)-3-lithioindole by metalation of the corresponding 3-bromo derivative. This literature procedure was successfully employed with several modifications to prepare 3-bromo-1-(*tert*-butyldimethylsilyl)-5-methoxyindole, **6**, in 85% yield, in a one-pot reaction, from 5-methoxyindole, by silylation, followed by regioselective bromination with *N*-bromosuccinimide at −78 °C (Scheme 1).

In our search for a suitable electrophilic acylating reagent, we then considered simple esters. To avoid working with a labile acyl chloride, the simple methyl ester of *N*-methylproline seemed to be an excellent choice. The ester was prepared in good yield starting from proline following literature precedents[10,11] and was obtained as anhydrous material after purification by vacuum distillation. The total route leading to 3-(*N*-methylpyrrolidin-2-ylcarbonyl)-5-methoxyindole, **7** (Scheme 1), does have several advantages over the literature procedure.[1] Only 1 equiv of the indole starting material is required. The use of the bulky *tert*-butyldimethylsilyl group as the indole *N*-protecting group, because of the steric demands of this substituent, provided lateral protection of the 2 position and prevented rearrangement to the undesired 2-lithio isomer. Moreover, this protecting group was lost during the reaction/workup, which further simplified the synthetic scheme. To be certain that no racemization was occurring in the acylation step, deuterium quench experiments were performed. That is, the acylation reaction was worked up using D_2O and the resulting product was characterized by ^{1}H NMR; no deuterium incorporation was detected. Complete reduction of the ketone functionality in **7** was accomplished smoothly and quantitatively with lithium aluminum hydride in THF at reflux to afford the desired enantiomers.

Synthesis of **8** was accomplished using the same sequence as for compound **7**, but starting from 4-benzyloxyindole (Scheme 2). The starting indole was prepared from *o*-benzyloxybenzaldehyde using the Hemetsberger azide pyrrolysis method.[8,9] Initial attempts to apply the *N*-protection protocol used for 5-methoxyindole resulted in partial *O*-debenzylation, effected by *n*-butyllithium due to the relatively high temperature used in the reaction. The literature procedure for the *N*-protection was modified, and the required deprotonation was carried out using the nonnucleophilic base sodium hydride. *O*-Debenzylation was not expected to be a problem in the bromine-exchange reaction because the reaction temperature was maintained at −78 °C. Indeed lithium−bromine exchange was favored over *O*-debenzylation, and the desired acylated compound was obtained in identical yield to the 5-methoxy compound.

The reduction of **9** was initially problematic. The ketone functionality was readily reduced to an alcohol, but even after 7 days at reflux with LiAlH$_4$ in THF, only a trace of the fully reduced compound was detected. Steric hindrance by the bulky *O*-benzyl protecting group was hypothesized to be the problem. Use of a THF/

Scheme 3[a]



[a] (a) AcOH, reflux; (b) $LiAlH_4$, THF, reflux; (c) H_2 (1 atm), Pd/C, EtOH.

dioxane mixture for the reduction improved the yield only slightly. Key to the success of this reduction was the choice of a single higher-boiling solvent, dioxane, and the manner of addition of reagents (see Experimental Section for details). Surprisingly, these conditions also effected an unprecedented *O*-debenzylation.

It seems possible that coordination of the aluminum with the 3-benzylic oxygen atom of the intermediate reduction product creates a transition state that is ideally disposed to deliver hydride to the 4-benzylic carbon. Cleavage of the carbon−oxygen bond is thereby effected, with phenoxide serving as the leaving group in a nucleophilic substitution reaction. Molecular modeling using semiempirical methods and the AM1 Hamiltonian, as employed in the Spartan software (v 5.0, Wavefunction, Inc.), confirmed that ideal geometries could be obtained for such a reaction sequence.

Compound **5** was prepared using a modification of the method of Macor et al.[6] via Michael addition between 4-benzyloxyindole and 3 equiv of *N*-methylmaleimide in acetic acid at reflux. Reduction of the resulting succinimide with $LiAlH_4$ followed by removal of the *O*-benzyl protecting group by catalytic hydrogenation afforded target compound **5** (Scheme 3).

Pharmacology

The target compounds synthesized in this work were evaluated for affinity at the rat brain 5-HT_{2A} receptor, labeled with the highly selective 5-HT_{2A} receptor antagonist [^{3}H]MDL100907.[13] In addition, competition studies were also carried out using [^{125}I]-2,5-dimethoxy-4-iodoamphetamine (DOI) as an agonist to label this site. The enantiomers with highest affinity for the 5-HT_{2A} receptor were then assessed in the drug discrimination paradigm (DD) in rats trained to discriminate either LSD tartrate from saline or DOI hydrochloride from saline.

Results and Discussion

The receptor binding assay results (Table 1) clearly demonstrate a stereochemical requirement at the 5-HT_{2A} receptor, but one that does not discriminate the position of the oxygen function. As reported by Macor et al.[1] for the enantiomers of **1**, the receptor is 20−30-fold more selective for the *R* isomer. The affinities of the individual enantiomers are very similar for both **1** and **3**. As would be expected for agonists, the affinities of all the compounds are correspondingly lower for the antagonist-labeled receptor, although the relative affinity ratios for enantiomeric pairs remain approximately the same. Compounds **4** and **5** have lower affinities than the more active enantiomers (*R*)-**1** and (*R*)-**3**, respectively, but still have 2−3-fold higher affinities than the less active *S* enantiomers of **1** and **3**.

Table 1. Radioreceptor Data for New Compounds in Competing for [^{3}H]MDL100907 ($K_{0.5} \pm$ SEM, nM) and [^{125}I]-2,5-Dimethoxy-4-iodoamphetamine (DOI) ($K_I \pm$ SEM, nM) at Serotonin 5-HT_{2A} Sites in Rat Cortical Homogenate

compd	[^{125}I]DOI	[^{3}H]MDL100907
5-MeO-DMT	21 ± 3	1600 ± 250
4-OH-DMT (psilocin)	6 ± 0.5[a]	390 ± 38[b]
(*S*)-**1**	64 ± 14	9400 ± 2300
	(680 ± 70)[c]	(>8400)[d]
(*R*)-**1**	9.5 ± 1.6	400 ± 22
	(17 ± 2)[c]	(730 ± 110)[d]
(*S*)-**3**	290 ± 33	4700 ± 600
(*R*)-**3**	13 ± 2	210 ± 22
4[e]	150 ± 20	1300 ± 150
5	82 ± 33	440 ± 54

[a] Value from McKenna et al.[15] for [^{125}I]DOI displacement. [b] Value for [^{3}H]ketanserin displacement; this laboratory. [c] Values from Macor et al.[1] for [^{125}I]DOI displacement. [d] Values from Macor et al.[1] for [^{3}H]ketanserin displacement. [e] Prepared according to Macor et al.[6]

Nonbonded interactions between the pyrrolidine ring and the substituent at the 4 position (−H or −OH) hinder the "ethylamine side chain" fragment in both **1** and **3** from adopting an ergoline-like conformation. This interaction is more severe in (*R*)-**3**, however, due to the larger size of the OH, compared with H. "LSD-like" conformations for (*R*)-**1** and (*R*)-**3** are about 3 and 7 kcal/mol (semiempirical, AM1) above their global minima, respectively. Lowest-energy conformations for both compounds exist where the ethylamine fragment of the molecule is disposed in a plane nearly perpendicular to the plane of the indole ring. Even though it is tempting to envision these rigid tryptamines binding in an ergoline-like conformation, these results suggest that this is probably an incorrect view. One explanation for decreased affinity of compounds **4** and **5** could be the added bulk at the 1 position of the ethylamine side chain.

The behavioral data from the drug discrimination assay in both LSD (Table 2) and DOI (Table 3) trained rats parallels the binding data using [^{125}I]DOI displacement. We have only obtained full substitution in this assay when compounds were agonists. Macor[1] has previously shown that (*R*)-**1** is an agonist, and these data suggest, not surprisingly, that (*R*)-**3** is also an agonist. Both *(R)*-**1** and *(R)*-**3** are about 10-fold less potent than LSD but have activity comparable to that of DOI. Although it is possible that (*R*)-**3** is slightly more potent than (*R*)-**1** in the LSD-trained rats, the overlap of the ED_{50} confidence intervals prevents a firm conclusion on this point.

Compound **4** produced only partial substitution, even at a dose nearly 5-fold higher than for (*R*)-**1**. Because of the similar loss in receptor affinity, and in view of these results, compound **5** was not tested in the behavioral assay.

Table 2. Data from Substitution Tests in LSD-Trained Rats[a]

drug	dose mg/kg	dose μmol/kg	*N*	%D	%SDL	ED_{50} (95% CI) μmol/kg	ED_{50} (95% CI) mg/kg
LSD	0.01	0.023	13	0	46		
	0.02	0.046	14	0	78	0.026	0.012
	0.04	0.093	16	0	81	(0.014−0.045)	(0.006−0.02)
	0.08	0.186	15	0	100		
(*R*)-**1**	0.08	0.25	6	17	20		
	0.16	0.50	9	11	63	0.44	0.14
	0.33	1.00	12	8	82	(0.25−0.74)	(0.08−0.24)
	0.65	2.00	8	17	100		
(*R*)-**3**	0.03	0.125	10	0	30		
	0.06	0.25	10	0	40	0.26	0.06
	0.12	0.50	12	8	64	(0.15−0.45)	(0.04−0.1)
	0.23	1.00	10	10	100		
4	0.68	2.0	12	0	33		
	1.36	4.0	10	0	50	PS	PS
	2.72	8.0	12	0	75		

[a] Slopes of dose−response curves for (*R*)-**1** and (*R*)-**3** are not significantly different and are not different from the slope of the LSD dose−response curve ($p > 0.05$).

Table 3. Data from Substitution Tests in DOI-Trained Rats[a]

drug	dose mg/kg	dose μmol/kg	*N*	%D	%SDL	ED_{50} (95% CI) μmol/kg	ED_{50} (95% CI) mg/kg
DOI	0.1	0.28	9	0	56		
	0.2	0.56	10	10	89	0.28	0.12
	0.4	1.12	10	0	100	(0.19−0.43)	(0.08−0.16)
(*R*)-**1**	0.08	0.25	8	12.5	43		
	0.16	0.50	9	11	63	0.32	0.11
	0.33	1.00	9	11	88	(0.14−0.7)	(0.04−0.23)
(*R*)-**3**	0.06	0.25	7	0	14		
	0.12	0.50	7	0	43	0.47	0.11
	0.23	1.00	8	12.5	100	(0.26−0.84)	(0.06−0.2)
4	0.68	2.0	7	0	14		
	1.36	4.0	6	0	50	PS	PS
	2.72	8.0	8	0	62.5		

[a] None of the slopes of the dose−response curves deviated significantly from parallelism.

In conclusion, oxygenation at either the 4- or 5-indole position has no consequences for receptor stereoselectivity and affords rigid tryptamine analogues with identical pharmacological properties, at least in the assays employed here. Further, based on conformational energy scans, it seems doubtful that these compounds bind to the 5-HT_{2A} receptor in an ergoline-like conformation. These results also suggest that both **1** and **3** would possess LSD-like psychopharmacology in humans.

Experimental Section

Chemistry. Melting points were determined using a Thomas-Hoover apparatus and are uncorrected. ^{1}H NMR spectra were recorded using either a 500 MHz Varian VXR-500S or 300 MHz Bruker ARX-300 NMR spectrometer. Chemical shifts are reported in δ values ppm relative to an internal reference (0.03%, v/v) of tetramethylsilane (TMS) in $CDCl_3$, except where noted. Chemical ionization mass spectra (CIMS) using methane as a carrier gas were obtained with a Finnigan 4000 spectrometer. Elemental analyses were performed by the Purdue University Microanalysis Laboratory and are within ±0.4% of the calculated values unless otherwise noted. All reactions were carried out under an inert atmosphere of argon.

3-Bromo-1-*(tert*-butyldimethylsilyl)-5-methoxyindole, 6. To a solution of 5-methoxyindole (705 mg, 4.80 mmol) in 30 mL of anhydrous THF at −78 °C was added dropwise a solution of *n*-butyllithium in hexane (3.5 mL of a 1.6 M solution, 5.76 mmol) and the temperature was raised to −10 °C over 10 min. After 15 min of stirring at this temperature, the reaction mixture was cooled back to −50 °C, and a solution of *tert*-butyldimethylsilyl chloride (844 mg, 5.76 mmol) in 5 mL of anhydrous THF was added. After stirring at 0 °C for 3 h the temperature was lowered to −78 °C and freshly crystallized *N*-bromosuccinimide (900 mg, 5.28 mmol) was added to the reaction mixture. After stirring for 3 h the temperature was raised to 25 °C and hexane (25 mL) followed by pyridine (0.25 mL) was added. The resulting suspension was filtered through a Celite pad, and the filtrate was concentrated under reduced pressure. The crude product was purified by column chromatography over silica gel with 5% ethyl acetate/hexane as the eluent to afford the title compound as a yellow solid (1.38 g, 85% yield). An analytical sample was recrystallized from hexane to give white crystals: mp 75 °C; ^{1}H NMR δ 7.36 (d, 1H, ArH, $J = 6.0$ Hz), 7.12 (s, 1H, ArH), 6.97 (d, 1H, ArH, $J = 2.2$ Hz), 6.83 (dd, 1H, ArH, $J = 6.0$ and 2.2 Hz), 3.88 (s, 3H, OCH_3), 0.92 (s, 9H, *tert*-butyl), 0.58 (s, 6H, dimethylsilyl); CIMS 341 (MH^+). Anal. ($C_{15}H_{22}BrNOSi$) C, H, N.

(*S*)-(−)-3-(*N*-Methylpyrrolidin-2-ylcarbonyl)-5-methoxyindole, (*S*)-7. A solution of *n*-butyllithium (0.56 mL of a 2.5 M solution in hexane, 1.53 mmol) was added dropwise at −78 °C to a solution of 3-bromo-1-(*tert*-butyldimethylsilyl)-5-methoxyindole (435 mg, 1.28 mmol) in 10 mL of anhydrous THF. The mixture was stirred at this temperature for 30 min and transferred via cannula into a flask containing a −78 °C solution of *N*-methyl-L-proline methyl ester (276 mg, 192 mmol) in anhydrous THF (3 mL). After stirring at −78 °C for 9 h the reaction mixture was warmed to −10 °C over a period of 1 h and quenched by addition of ice−water. The resulting reaction mixture was concentrated under reduced pressure. Trituration of the resulting residue with ether produced an off-white solid, which was collected by filtration, washed on the filter with ether, and dried under vacuum to provide the title compound (115 mg, 35% yield). An analytical sample was crystallized from acetonitrile to afford shiny white needles: mp 180 °C dec; ^{1}H NMR (DMSO-d_6) δ 11.80 (br s, 1H, NH), 8.48 (s, 1H, ArH), 7.75 (d, 1H, ArH, $J = 2.1$ Hz), 7.33 (d, 1H, ArH, $J = 8.5$ Hz), 6.82 (dd, 1H, ArH, $J = 8.5$ and 2.1 Hz), 3.78 (s, 3, OCH_3), 3.51 (m, 1H, −CH−N), 3.12 (m, 1H, NCH_2CH_2), 2.30 (m, 1H, NCH_2CH_2), 2.25 (s, 3H, NCH_3), 2.13 (m, 1H, NCH_2CH_2), 1.80 (m, 3H, $NCH_2CH_2CH_2$ and $NCH_2CH_2CH_2$); $[\alpha]_D^{25} = -125°$ [$c = 0.01$, DMF]; CIMS 259 (MH^+). Anal. ($C_{15}H_{18}N_2O_2$) C, H, N.

(*R*)-(+)-3-(*N*-Methylpyrrolidin-2-ylcarbonyl)-5-methoxyindole, (*R*)-7. The above-described procedure was used, but employing *N*-methyl-D-proline methyl ester: mp 185 °C. The spectral properties of this compound were identical to those described above for (*S*)-**7**, except: $[\alpha]_D^{25} = +114°$ [$c = 0.01$, DMF]. Anal. ($C_{15}H_{18}N_2O_2$) C, H, N.

(*S*)-(−)-3-(*N*-Methylpyrrolidin-2-ylmethyl)-5-methoxyindole, (*S*)-1.[1] To a stirred mixture of lithium aluminum hydride (570 mg, 15 mmol) in anhydrous THF (100 mL) at 0 °C was added (*S*)-**7** (780 mg, 3 mmol) portionwise. The resulting mixture was heated at reflux for 18 h. After cooling, 1 g of sodium sulfate decahydrate was added carefully, followed by ethyl acetate (10 mL) and then water (0.2 mL). The resulting slurry was stirred at room temperature for 6 h. The mixture was filtered through Celite with copious washing with ethyl acetate, and the combined filtrates were evaporated under reduced pressure. The residual yellow oil was purified by column chromatography over silica gel with methylene chloride/methanol/ammonium hydroxide (9:1:0.1) as the eluent to afford the title compound (700 mg, 95%) as a slightly tan thick oil, which solidified upon standing in the freezer: mp for the free base 52−54 °C (lit.[1] mp 54−57 °C); ^{1}H NMR (fumarate salt; DMSO-d_6) δ 10.7 (br s, 1H, NH), 7.22 (d, 1H, ArH, $J = 8.8$ Hz), 7.15 (s, 1H, ArH), 7.02 (d, 1H, ArH, $J = 2.4$ Hz), 6.71 (dd, 1H, ArH, $J = 8.8$, 2.4 Hz), 6.53 (s, 2H, fumarate vinyl), 3.73 (s, 3H, OCH_3), 3.34 (m, 1H, −CH−N), 3.16 (dd, 1H, $ArCH_2$, $J = 15$, 6 Hz), 3.02 (m, 1H, NCH_2CH), 2.75−2.62 (m, 2H, $ArCH_2$ and NCH_2CH), 2.6 (s, 3H, NCH_3), 1.9−1.55 (m, 4H, $NCH_2CH_2CH_2$, $NCH_2CH_2CH_2$); $[\alpha]_D^{25} = -103°$ [$c = 1$, $CHCl_3$] (lit. $[\alpha]_D^{25} = -98°$ [$c = 0.01$, $CHCl_3$]).

(*R*)-(+)-3-(*N*-Methylpyrrolidin-2-ylmethyl)-5-methoxyindole, (*R*)-1.[1] The above-described procedure was used, but employing (*R*)-3-(*N*-methylpyrrolidin-2-ylcarbonyl)-5-methoxyindole. The physical and spectral properties of this compound were identical to the physical and spectral properties of (*S*)-**1**, except: $[\alpha]_D^{25} = +94°$ [$c = 0.01$, $CHCl_3$] (lit. $[\alpha]_D^{25} = +100°$ [$c = 0.01$, $CHCl_3$]).

3-Bromo-1-*(tert*-butyldimethylsilyl)-4-benzyloxyindole, 8. To a solution of 4-benzyloxyindole (2.50 g, 11.21 mmol) in anhydrous THF (100 mL) at 0 °C was added portionwise sodium hydride (60% suspension in mineral oil, 538 mg, 13.5 mmol). The resulting suspension was allowed to warm to room temperature over a period of 1 h and then was cooled to 0 °C, and a solution of *tert*-butyldimethylsilyl chloride (1.85 g, 12.31 mmol) in anhydrous THF (25 mL) was added. After stirring at 0 °C for 2.5 h the resulting mixture was cooled to −78 °C and freshly crystallized *N*-bromosuccinimide (2.19 g, 12.31 mmol) was added. After stirring at this temperature for 4 h the resulting mixture was allowed to warm to 25 °C, and hexane (100 mL) and pyridine (2 mL) were added. The resulting suspension was filtered through Celite and the filtrate was concentrated under reduced pressure. The crude product was purified by column chromatography over silica gel with 50% CH_2Cl_2/hexane as eluent to afford the title compound (3.95 g, 85% yield) as an off-white solid. An analytical sample was crystallized from hexane: mp 95 °C; ^{1}H NMR δ 7.61 (d, 2H, ArH, $J = 7$ Hz), 7.42 (t, 2H, ArH, $J = 7$ Hz), 7.32 (m, 2H, ArH, $J = 7$ Hz), 7.15−7.04 (m, 2H, ArH), 6.62 (dd, 1H, $J = 7.5$ and 0.89 Hz), 5.23 (s, 2H, CH_2), 0.93 (s, 9, *tert*-butyl), 0.58 (s, 6, dimethyl); CIMS 417 (MH^+). Anal. ($C_{21}H_{26}BrNOSi$) C, H, N.

(*S*)-(−)-3-(*N*-Methylpyrrolidin-2-ylcarbonyl)-4-benzyloxyindole, (*S*)-9. A three-neck flask containing a solution of 3-bromo-1-*(tert*-butyldimethylsilyl)-4-benzyloxyindole (1.5 g, 3.6 mmol) in anhydrous THF (50 mL) was cooled to −78 °C. A solution of *n*-butyllithium (1.65 mL of a 2.5 M solution in hexane, 4.32 mmol) was then slowly added, allowing it to run in along the inside walls of the flask in order to cool to −78 °C, and the mixture was stirred at this temperature for 1 h. The resulting 3-lithioindole was transferred via cannula into a precooled solution of *N*-methyl-L-proline methyl ester (776 mg, 5.4 mmol) in anhydrous THF (10 mL). After stirring at −78 °C for 20 h, the reaction mixture was allowed to warm to −30 °C over a period of 2 h and was then poured into ice. The resulting mixture was concentrated under reduced pressure. Upon addition of ether to the resulting residue, an off-white precipitate was formed, which was collected by filtration, washed on the filter with ether ,and dried under high vacuum to afford the desired product (450 mg, 35% yield). An analytical sample was crystallized from acetonitrile: mp 175 °C dec; ^{1}H NMR (DMSO-d_6) δ 8.08 (s, 1H, ArH), 7.6 (d, 2H, ArH, $J = 7.0$ Hz), 7.3−7.4 (m, 3H, ArH), 7.14−7.03 (m, 2H, ArH), 6.75 (dd, 1H, ArH, $J = 8.1$ and 1.1 Hz), 5.19 (s, 2H, $ArCH_2$), 3.76 (m, 1H, N−CH), 2.94 (m, 1H, N−CH_2), 2.14 (s, 3, NCH_3), 2.14−2.05 (m, 1H, N−CH_2), 2.05−1.89 (m, 1H, CH), 1.78−1.56 (m, 3H, CH); $[\alpha]_D^{25} = -102°$ [$c = 0.01$, DMF]; CIMS 335 (MH^+). Anal. ($C_{21}H_{22}N_2O_2$) C, N, H.

(*R*)-(+)-3-(*N*-Methylpyrrolidin-2-ylcarbonyl)-4-benzyloxyindole, (*R*)-9. The above-described procedure was used, but employing *N*-methyl-D-proline methyl ester. The physical and spectral properties of this compound were identical to the physical and spectral properties of (*S*)-**9**, except: $[\alpha]_D^{25} = +91°$ [$c = 0.01$, DMF]. Anal. ($C_{21}H_{22}N_2O_2$) C, N, H.

(*S*)-(−)-3-(*N*-Methylpyrrolidin-2-ylmethyl)-4-hydroxyindole, (*S*)-3. A suspension of $LiAlH_4$ (142 mg, 3.72 mmol) in anhydrous dioxane (6 mL) was brought to reflux, and a heated solution of (*S*)-3-(2-*N*-methylpyrrolidin-2-ylcarbonyl)-4-benzyloxyindole (250 mg, 0.75 mmol) in anhydrous dioxane (6 mL) was added via syringe. The resulting mixture was held at reflux until TLC indicated that the reaction was complete (72−96 h). The reaction mixture was cooled to 25 °C, water (0.5 mL) was added carefully, followed by ether (12 mL), and the resulting slurry was stirred at room temperature for 15 min. The mixture was filtered through Celite, the filter cake was washed copiously with ether, and the combined filtrates were evaporated under reduced pressure. The residual oil was dissolved in ether, washed with brine, dried with $MgSO_4$, and placed under high vacuum to provide the title compound (160 mg, 92% yield) as a white solid. An analytical sample was crystallized from ethyl acetate/hexane: mp 135 °C; ^{1}H NMR δ 13.8 (br s, 1H, OH), 7.92 (s, 1H, NH), 7.04 (t, 1H, ArH, $J = 8$ Hz), 6.84 (m, 2, ArH), 6.54 (d, 1, ArH, $J = 8$ Hz), 3.12 (m, 2H, CH_2), 2.89 (m, 2H, CH_2), 2.43 (s, 3H, NCH_3), 2.36 (dt, 1H, N−CH), 1.94 (m, 1H, N−CH_2), 1.68 (m, 2H, CH_2), 1.47 (m, 1H, CH_2); $[\alpha]_D^{25} = -18°$ [$c = 0.01$, DMF]. Measured exact mass (FAB, M + H^+): 231.1495. Calcd: 231.1497.

***(R)*-3-(*N*-Methylpyrrolidin-2-ylmethyl)-4-hydroxyindole, (*R*)-3.** The above-described procedure was used, but employing (*R*)-3-(2-*N*-methylpyrrolidin-2-ylcarbonyl)-4-benzyloxyindole. The physical and spectral properties of this compound were identical to the physical and spectral properties of (*S*)-**3**, except: $[\alpha]_D^{25} = +15°$ [$c = 0.01$, DMF]. Measured exact mass (FAB, M + H^+): 231.1496.

(*R,S*)-(±)-3-(4-Benzyloxyindol-3-yl)-*N*-methylsuccinimide, 10. Following a modification of the method of Macor et al.[6] a solution of 4-benzyloxyindole (1 g, 4.48 mmol) and *N*-methylmaleimide (1.49 g, 13.44 mmol) in AcOH (25 mL) was heated at reflux for 6 days. Upon consumption of the starting material as observed by TLC, the mixture was cooled, the solution was evaporated under reduced pressure, and the residue was purified by crystallization from ethyl acetate to afford the desired compound as a tan solid in 75% yield. An analytical sample was obtained following several recrystallizations from EtOAc: mp 195 °C dec; ^{1}H NMR (DMSO-d_6) δ 11.02 (s, 1H, NH), 7.41−7.25 (m, 5, ArH), 7.2 (s, 1H, ArH), 6.98−6.82 (m, 2H, ArH), 6.36 (d, 1, ArH, $J = 7.8$ Hz), 5.08 (s, 2H, $ArCH_2$), 4.28−4.16 (dd, 1H, CO*CH*CH_2, $J = 9.1$, 7.4 Hz), 3.08 (dd, 1H, COCH*CH*$_2$, $J = 9.1$ and 17.4 Hz), 2.72 (dd, 1H, COCH*CH*$_2$, $J = 7.4$ and 17.4 Hz), 2.65 (s, 3, NCH_3); CIMS 335 (MH^+). Anal. ($C_{20}H_{18}N_2O_3$) C, H, N.

(*R,S*)-(±)-3-(*N*-Methylpyrrolidin-3-yl)-4-benzyloxyindole, 11. To a stirred suspension of $LiAlH_4$ (570 mg, 15 mmol) in anhydrous THF (25 mL) at 0 °C was added **10** (1.0 g, 3.0 mmol) as a solid portionwise, and the resulting mixture was heated at reflux for 3 h. The reaction mixture was cooled and water (2 mL) was added slowly, followed by ether (70 mL). The resulting slurry was filtered through Celite and the filtrate was evaporated under reduced pressure. The residual oil was dissolved in ether, washed with brine, dried with $MgSO_4$, and concentrated under reduced pressure. The resulting off-white solid was purified by crystallization from chloroform/petroleum ether to afford the title compound (780 mg, 84% yield): mp 130 °C; ^{1}H NMR δ 8.0 (br s, 1H, NH), 7.46 (d, 2H, ArH, $J = 7.5$ Hz) 7.45−7.34 (m, 3H, ArH), 7.05 (t, 1H, ArH, $J = 8.1$ Hz), 6.95−6.86 (m, 2H, ArH), 6.52 (d, 1H, ArH, $J = 7.6$ Hz), 5.15 (s, 2H, $ArCH_2$), 4.02−3.90 (p, 1H, CH_2CHCH_2, $J = 6.8$ Hz), 2.95 (t, 1H, CH_2N, $J = 6.8$ Hz), 2.70−2.52 (m, 3H, $CHCH_2$N and CH_2CH_2N), 2.32−2.20 (m, 4H, CH_2CH_2N, NCH_3), 1.95 (m, 1H, CH_2CH_2N); CIMS 307 (MH^+). Anal. ($C_{20}H_{22}N_2O$) C, H, N.

(*R,S*)-(±)-3-(*N*-Methylpyrrolidin-3-yl)-4-hydroxyindole, 5. A solution of (*R,S*)-**11** (570 mg, 1.86 mmol) in 95% EtOH (20 mL) was stirred under 1 atm of hydrogen over 10% Pd/C (60 mg) for 7 h. The catalyst was removed by filtration, the filtrate was concentrated under reduced pressure, and the residue was dried under vacuum to afford the title compound as a white solid in 95% yield: mp 170 °C dec; ^{1}H NMR δ 13.75 (br s, 1H, OH), 7.98 (s, 1H, NH), 7.08 (t, 1H, ArH, $J = 7.6$ Hz), 6.84−6.81 (m, 2H, ArH), 6.55 (d, 1H, ArH, $J = 7.7$ Hz), 3.71−3.59 (m, 1H, CH_2CHCH_2), 3.33 (t, 1H, $CHCH_2$N, $J = 7.5$ Hz), 3.12 (d, 1H, $CHCH_2$N, $J = 7.5$ Hz), 2.58 (t, 1H, CH_2CH_2N, $J = 9$ Hz), 2.50 (s, 3H, NCH_3), 2.46−2.32 (m, 1H, CH_2CH_2N), 2.32−2.10 (m, 2H, CH_2CH_2N). Measured exact mass (FAB, M + H^+): 217.1340. Calcd: 217.1341.

Pharmacology Methods. Competition Assays in Rat Brain Homogenate. The procedure of Johnson et al.[12] was employed with minor modifications. Briefly, 50 male Sprague−Dawley whole rat brains (unstripped) were purchased from Harlan Bioproducts for Science, Inc. and dissected over dry

ice. The frontal cortex tissue was homogenized (Kinetica polytron, setting 4, 2 × 20 s) in 4 volumes (w/v) of ice-cold 0.32 M sucrose and centrifuged at 36000*g* for 10 min at 4 °C. The pellet was again suspended in the same volume of sucrose, homogenized (Kinematica polytron, setting 4, 20 s), and separated into aliquots of 4.5 mL and stored at −70 °C.

For each experiment one aliquot of frontal cortex tissue was thawed and diluted with 25 volumes (w/v) of 50 mM tris-(hydroxymethyl)aminomethane (Aldrich Chemicals) buffer, adjusted to pH 7.4 by HCl. The tissues were homogenized (Kinematica polytron, setting 4, 20 s) and incubated for 10 min in a 37 °C shaking water bath. The homogenate was then centrifuged twice at 36000*g* for 10 min at 4 °C, with the pellet being resuspended in 25 volumes of Tris-HCl buffer between centrifugations. The supernatant was discarded, and the pellet was resuspended with 25 volumes of Te Pac buffer (0.5 mM Na_2EDTA, 10.0 μM pargyline, 5.7 mM $CaCl_2$, 0.1% sodium ascorbate), homogenized using the Kinematica polytron as above, and incubated in a 37 °C shaking water bath for 10 min. The homogenate was then placed in an ice bath to cool. Binding was initiated by adding 800 μL of homogenate tissue (200−400 μg of protein) to assay tubes containing 100 μL of [^{3}H]MDL100907 (0.2 nM) and 100 μL of competing drug solution. Nonspecific binding was determined in the presence of cinanserin (10 μM). Binding assays were incubated for 15 min in a 37 °C shaking water bath. Incubation was stopped by rapid vacuum filtration using a Brandel cell harvester (Brandel Instruments, Gaithersburg, MD) through GF/C filters. The filters were washed twice with 5-mL aliquots of ice-cold Tris-HCl buffer, allowed to air-dry, and placed into scintillation vials containing 10 mL of Ecolite scintillation cocktail (ICN Biomedicals). Eight hours later the radioactivity was measured using liquid scintillation spectroscopy (Packard model 4430) at 37% efficiency for tritium. For experiments with [^{125}I]DOI, the dried filters were counted directly for ^{125}I at 79% efficiency in a gamma counter. EC_{50} (nM) values were calculated from at least three experiments, each done in triplicate, using GraphPad PRISM.

[^{3}H]MDL100907 and [^{125}I]DOI were purchased from Amersham Life Sciences (Arlington Heights, IL) or New England Nuclear (Boston, MA) at specific activities of 82 and 2000 Ci/mmol, respectively. (+)-LSD tartrate was obtained from the National Institute on Drug Abuse. Cinanserin was a gift from the SQUIBB Institute for Medical Research, and 5-HT was purchased from Sigma (St. Louis, MO).

Drug Discrimination Studies. The procedures for the drug discrimination assays were essentially as described in previous reports.[14,15] Male Sprague−Dawley rats (Harlan Laboratories, Indianapolis, IN), weighing 200−220 g at the beginning of the study, were trained to discriminate LSD or DOI from saline. None of the rats had previously received drugs or behavioral training. Water was freely available in the individual home cages, and a rationed amount of supplemental feed (Purina Lab Blox) was made available after experimental sessions to maintain approximately 80% of free-feeding weight. Lights were on from 0700 to 1900. The laboratory and animal facility temperature was 22−24 °C, and the relative humidity was 40−50%. Experiments were performed between 0830 and 1700 each day, Monday−Friday.

A fixed ratio (FR) 50 schedule of food reinforcement (Bioserv 45 mg dustless pellets) in a two-lever paradigm was used. To avoid positional preference, half of the rats were trained on drug-L (left), saline-R (right) and the other half on drug-R, saline-L. Training sessions lasted 15 min and were conducted at the same time each day. Response levers were cleaned with 10% ethanol solution between animals to avoid olfactory cues. Presses on the incorrect lever had no programmed consequences.

Test sessions were interspersed between training sessions, either one or two times/week. At least one drug and one saline session separated each test session. Rats were required to maintain the 85% correct responding criterion on training days in order to be tested. In addition, test data were discarded when the accuracy criterion of 85% was not achieved on the two training sessions following a test session. Test sessions were run under conditions of extinction, with rats removed from the operant chamber when 50 presses were emitted on one lever. If 50 presses on one lever were not completed within 5 min, the session was ended and scored as a disruption. Treatments were randomized at the beginning of the study.

The training drugs were (+)-lysergic acid diethylamide tartrate (LSD, 0.08 mg/kg, 0.186 μmol/kg; NIDA) and (±)-2,5-dimethoxy-4-iodoamphetamine hydrochloride (DOI, 0.4 mg/kg, 1.12 μmol/kg; NIDA). All drugs were dissolved in 0.9% saline and were injected intraperitoneally in a volume of 1 mL/kg, 30 min before the sessions. Data from the drug discrimination study were scored in a quantal fashion, with the lever on which the rat first emitted 50 presses in a test session scored as the "selected" lever. The percentage of rats selecting the drug lever (%SDL) for each dose of test compound was determined. The degree of substitution was determined by the maximum %SDL for all doses of the test drug. "No substitution" was defined as 59% SDL or less, and "partial" substitution is 60−79% SDL. If the drug was one that completely substituted for the training drug (at least one dose resulted in a %SDL = 80% or higher), the method of Litchfield and Wilcoxon[16] was used to determine the ED_{50} (log−probit analysis as the dose producing 50% drug−lever responding) and 95% confidence interval (95% CI). This method also allowed for tests of parallelism between dose−response curves of the drug and the training drug. If 50% or more of the animals tested were disrupted at a dose, even if the nondisrupted rats gave 80% SDL, no ED_{50} was calculated.

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