Benzo- and Cyclohexanomazindol Analogues as Potential Inhibitors of the Cocaine Binding Site at the Dopamine Transporter

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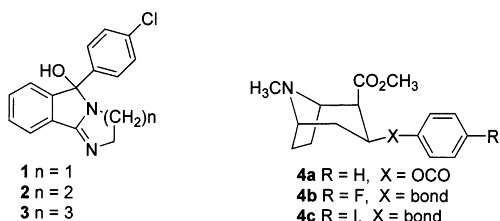
A series of mazindol (**1**), homomazindol (**2**), and bishomomazindol (**3**) derivatives with a benzo or cyclohexano ring fused at various sites were prepared as part of an SAR study to determine the effect of increased aliphatic and aromatic lipophilicity on selected in vitro assays used to identify potential cocaine-like and cocaine antagonism activity. Very good (IC_{50} = 2−3 nM) inhibition of [^{3}H] WIN 35,428 and [^{125}I] RTI-55 binding on rat or guinea pig striatal membranes and HEK cells expressing cDNA for the human dopamine transporter (HEK-hDAT) was shown by the 8,9-benzomazindol **25** and 9,10-benzohomomazindol **28**. All new compounds were weaker inhibitors of [^{3}H] DA uptake in HEK-hDAT cells than **1** and **2**. No improvement in the binding selectivity ratio (SERT/DAT and NET/DAT) was found when compared to **2**. Compounds **25** and **28** showed a considerable increase versus **1** in uptake/binding discrimination ratios at the DAT (311.0 and 182.1 vs 0.9), SERT (33.6 and 127.3 vs 1.9), and NET (7.3 and 10.0 vs 0.3).

Introduction

The search for safe and effective medications in the treatment of cocaine abuse has focused mainly on substances that can displace its binding at the dopamine transporter (DAT) in the striatum.[1] The interaction of cocaine with the DAT results in the blockage of the uptake of DA into the presynaptic axon terminal and leads to an increase of dopamine in the synapse. This results in increased interaction of DA with its receptors on postsynaptic neurons that gives rise to reinforcing and stimulatory effects.[2]

The preceding[3] and earlier papers[4,5] from our laboratories have reported on structure−activity relationships (SAR) studies on mazindol (**1**) and its ring A homologues , homomazindol (**2**), and bishomomazindol (**3**) as potential inhibitors of cocaine (**4a**) binding at the DAT. That work focused on the effect of substituents on the pendant aryl group, rings C and C-5 or C-6, enlargement of ring A, and modification of the carbonyl group in the keto tautomer of mazindol (**M-keto** in Figure 1). In addition, an interaction model of how **1** to **3** could interact at the DAT was proposed.[3]



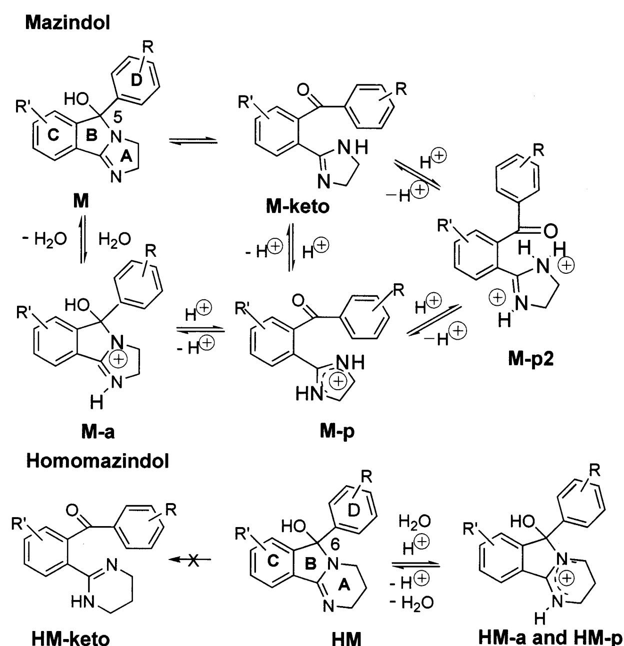


Figure 1. Tautomeric forms of aryl substituted mazindol (**M**) and homomazindol (**HM**) analogues in neutral and acidic media.

In this paper, we investigate the effects of enhancing the lipophilicity of **1** to **3** by adding one (**6**, **7**, **11**, **15**, **16**, **19**, **22**, **24**, and **28**), two (**25** and **32**), or three (**33**) benzene rings, one cyclohexane ring (**8**, **12**, and **13**), and one benzene and cyclohexane ring (**30**) on rings A, C, and D (Figure 1).

Chemistry

The synthesis of the compounds used in this study is given in Schemes 1−5.

Treatment of the known[6] *N,o*-dilithio derivative of 2-phenylimidazoline (**5**) with the methyl esters of 1- and 2-naphthoic and 5,6,7,8-tetrahydronaphthoic acids gave **6**, **7**, and **8** (Scheme 1). Compound **6** was determined to

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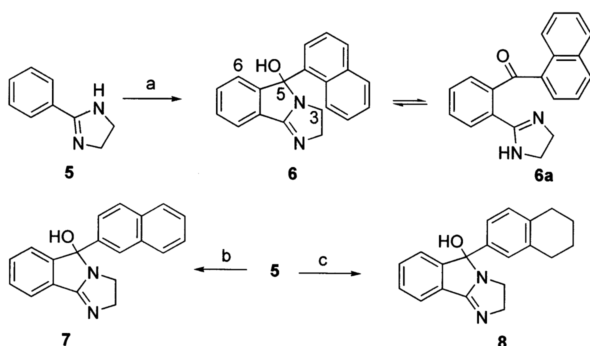
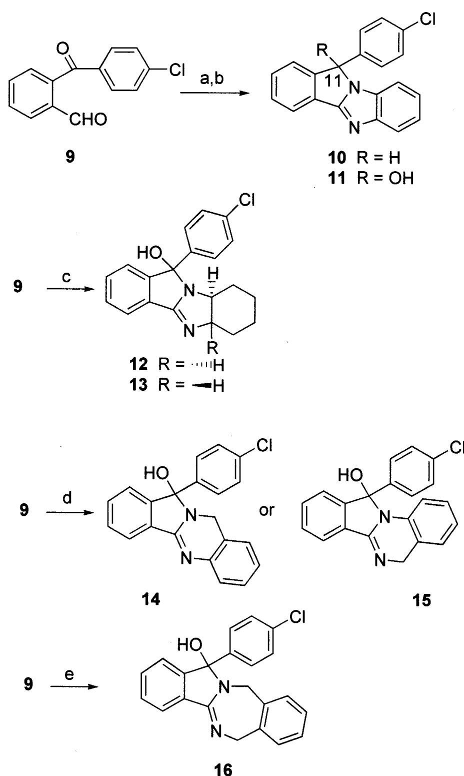
Scheme 1[a]



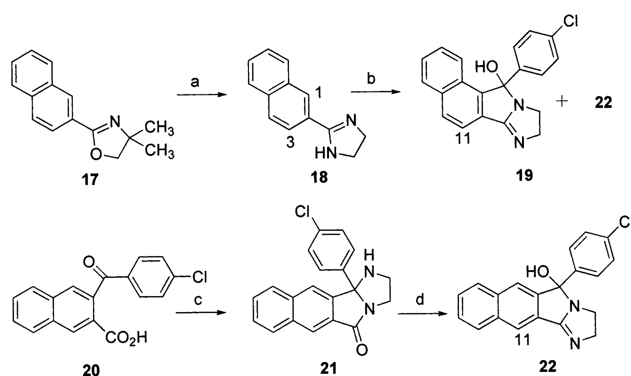
[a] Reagents/conditions. (a) (i) *n*-BuLi, TMEDA, THF, N_2, rt; (ii) methyl-1-naphthoate, THF, rt, overnight. (b) (i) *n*-BuLi, THF, N_2, rt; (ii) methyl-2-naphthoate, THF, rt, overnite. (c) (i) Same as those in panel b; (ii) methyl-5,6,7,8-tetrahydronaphthoate, THF, rt, overnight.

Scheme 2[a]



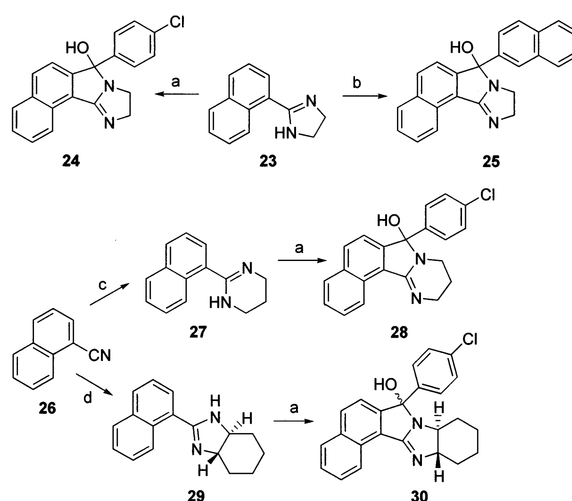
[a] Reagents/conditions. (a) 1,2-Diaminobenzene, HOAc, concd. HCl, 80°, 0.5 h. (b) (i) **10**, NaH, DMF, N_2; (ii) air, 6 h, rt. (c) *cis*- or *trans*-1,2-diaminocyclohexane, xylenes, reflux; (ii) MeOH, air, 96 h, rt. (d) (i) 2-Aminobenzylamine, xylenes, reflux; (ii) i-PrOH, air, 56 h, rt. (e) (i) 1,2-Benzenedimethanamine, xylenes, reflux; (ii) i-PrOH, air, 56 h, rt.

Scheme 3[a]



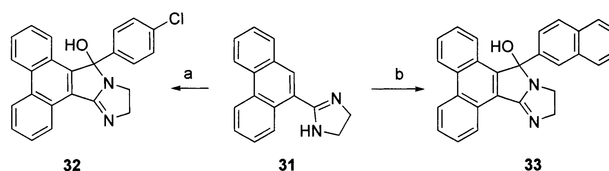
[a] Reagents/conditions. (a) (i) MeI, N_2, 6 days, rt; (ii) 17-MeI, EDA, reflux, 10 h. (b) (i) *n*-BuLi, THF, N_2, rt; (ii) methyl 4-chlorobenzoate, THF, overnight, rt. (c) EDA, xylene reflux, 7 h. (d) $LiAlH_4$, THF, N_2, 18°, 0.25 h.

Scheme 4[a]



[a] Reagents/conditions. (a) (i) *n*-BuLi, THF, N_2; (ii) methyl-4-chlorobenzoate, THF, overnight, rt. (b) (i) Same as those in panel a; (ii) methyl-2-naphthoate, THF, overnight, rt. (c) 1,3-Diaminopropane, p-tolSO_3H, 220°, 2.5 h. (d) *trans*-1,2-Diaminocyclohexane, *p*-tolSO_3H, 225°, 3 h.

Scheme 5[a]



[a] Reagents/conditions. (a) (i) *n*-BuLi, THF, argon, rt; (ii) methyl 4-chlorobenzoate, THF, rt, overnight. (b) (i) Same as thsoe in panel a; (ii) methyl 2-naphthoate, THF, overnight.

exist as a mixture of ol and keto tautomers (ca. 5:95) by the presence of characteristic ^{13}C NMR signals at 88.3 and 197.44 ppm due to C-5 and C=O, respectively, while **7** and **8** are exclusively in the ol form. Inspection of Dreiding models suggests that the keto form predominates because of steric interaction of the naphthyl group with positions 3 and 6 when **6** is in the ol form.

Condensation of **9**[7] with 1, 2-diaminobenzene in a mixture of warm HOAc/HCl gave 11H-isoindolo-[2,1-a]-benzimidazole **10**. Air oxidation[8] of the C-11 sodium salt of **10** in DMF gave the known[9] 11-ol derivative **11**. Treatment of **9** with racemic *cis*- or *trans*-1, 2-diaminocyclohexane in refluxing xylenes followed by exposure to air of the resulting 11-H compounds gave the 11-ols **12** and **13**. The ^{13}C NMR of **12** gave two C-11 signals (87.85 and 88.34) and two C=N signals (165.81 and 167.86), and **13** showed C-11 (87.82 and 89.04) and C=N signals (166.83 and 168.02), indicating that both existed as diastereomeric mixtures of at least two and possibly four compounds each. ^{1}H NMR analysis of the hydrogens at 5a and 9a in **12** disclosed that each integrated for 0.5 H, indicating a 50:50 diastereomeric mixture. A similar analysis of **13** showed that several atoms integrated for 0.4 or 0.6 H corresponding to a 40:60 diastereoismeric mixture. No attempt was made to

separate the mixtures because of the known[3] ability of a C−OH in mazindol analogues containing an imidazo ring A to form the corresponding C=O tautomer under mild conditions and then revert to the C−OH form (Figure 1).

Reaction of **9** with 2-aminobenzylamine or 1,2-benzenedimethanamine in refluxing xylenes, followed by exposure to air, gave compounds with ^{1}H and ^{13}C NMR consistent with a single entity in the ol form that could be either **14** or **15** and the expected **16**, respectively. The large separation (4.13 and 4.62 ppm) of the methylene hydrogen signals in the ^{1}H NMR of **14**/**15** suggests that one of the hydrogen atoms is in the vicinity of a polar environment. This is possible in **14**, where one hydrogen is shielded by the *p*-chlorophenyl group. Additional evidence favoring **14** is found by comparing its UV spectrum with **16**. Compound **16** has a UV spectrum similar to that of bishomomazindol (**3**), with maxima at 201 and 272 nm, whereas the UV of **14**/**15** gave multiple maxima between 203 and 356 nm that are in agreement with extended conjugation as found in **14**, but not **15**.

The known[10] oxazole **17** was converted to the imidazoline **18** by treatment with iodomethane and then reacting the resultant methiodide salt with refluxing 1,2-diaminoethane. Lithiation of **18** with *n*-butyllithium followed by treatment with methyl 4-chlorobenzoate gave a mixture of two compounds with very close R_f values (0.24 and 0.29) and NMR spectra, suggesting that lithiation had occurred at both C-1 and C-3 to give **19** and the known[11] **22**. Separation by plate chromatography resulted in the isolation of the R_f = 0.24 substance as a single entity. Comparison of the ^{1}H and ^{13}C NMR spectrum of **22**, prepared by condensation of keto-acid **20**[12] with 1,2-diaminoethane to **21** followed by lithium aluminum hydride reduction, with the R_f = 0.24 material showed the two were different compounds. Additional evidence for assigning the R_f = 0.24 substance as **19** comes from the ^{1}H NMR spectrum which has a doublet at 8.10 ppm that can be assigned to the H on C-11. The H on C-11 in **22** gave the expected singlet at 8.33 ppm. It is interesting to note that no lithiation is reported to occur at the C-3 position of *N*,*N*-dimethyl-2-naphthamide.[9]

Lithiation of naphthylimidazoline[13] (**23**), followed by treatment with methyl esters of 4-chlorobenzoic and 2-naphthoic acid, gave the expected benzo-[g]-imidazo-[2,1-a]-isoindol-5-ols **24** and **25**, respectively (Scheme 4).

Condensation of 1-cyanonaphthalene (**26**) with *p*-toluenesulfonic acid salts of 1,3-diaminopropane and racemic *trans*-1,2-diaminocyclohexane at ca. 220 °C gave **27** and **29** respectively. Treatment of **27** with *n*-butyllithium at room temperature and then methyl-4-chlorobenzoate gave **28** as the ol tautomer. Lithiation of **29** in an icebath, followed by reaction with methyl-4-chlorobenzoate, gave **30** as a mixture of up to four compounds (Scheme 4). The ^{13}C NMR spectrum of **30** gave two C-13 (88.9 and 90.1 ppm) and C=N signals (168.5 and 170.4 ppm), while the ^{1}H NMR spectrum gave several hydrogen atoms integrating for 0.5 H, indicating a 50:50 diastereoisomer mixture at C-13. No attempt was made to separate these because, like **12** and **13**, the presence of an imidazo ring A makes the C−OH at C-13 subject to ol−keto tautomerism (Figure 1).

Table 1. Inhibition of [^{3}H] WIN 35,428 Binding at the Dopamine Transporter on Guinea Pig Striatal Membranes[a]

compd no.	IC_{50}, nM[b] (% inhib. at 10^{-6} M)	IC_{50} ratio mazindol/analog
1	42.6 ± 2.6	1.0
2	4.2 ± 0.6	10.1
6	1841 ± 147	0.02
7	4.1 ± 0.3	10.4
8	(35)	<0.02
11	(30)	<0.02
12	8.4 ± 0.4	5.1
13	2.7 ± 0.2	15.8
14	(45)	<0.02
16	(12)	<0.02
19	218 ± 13	0.2
22	36.1 ± 2.7	1.3
24	10.6 ± 1.1	4.0
25	3.3 ± 0.2	13.0
28	2.1 ± 0.2	20.3
30	(5)	<0.02
32	(5)	<0.02
33	(20)	<0.02

[a] See Experimental Section for details. [b] Data represent the mean (± SEM) of three assays at five concentrations for each compound.

Lithiation of 2-(9-phenanthryl)-imidazoline (**31**) at room temperature and then reaction with methyl 4-chlorobenzoate or naphthoate gave **32** and **33** as the ol tautomers (Scheme 5).

Biology

The compounds listed in Table 1 were tested for their ability to displace [^{3}H] WIN 35,428 (WIN) binding from guinea pig striatal membranes. Those more active (**7**, **13**, **24**, **25**, and **28**) than mazindol were evaluated in displacing bound WIN from rat striatal membranes, displacing [^{125}I] RTI-55 (RTI) and blocking biogenic amines (DA, 5-HT, NE) uptake in HEK cells expressing cDNA for the human dopamine (hDAT), serotonin (hSERT), and norepinephrine (hNET) transporters.

Results and Discussion

The inhibition of WIN binding at the DAT of guinea pig striatal membranes for all compounds evaluated in this work is given in Table 1.

Placement of a benzene ring at the 2′,3′-positions (**6**) of the pendant aryl group in mazindol resulted in a 43-fold loss of activity, while placement at the 3′,4′-position (**7**) gave a 10-fold increase. The finding with **6** is in contrast to the work of Davies,[14] who reported that replacement of a 3β-phenyl by a 1- or 2-naphthyl group in the 2β-acyl-3β-aryl-8-azabicyclo-[3.2.1]-octane series resulted in increased activity at inhibiting the binding of RTI on rat striatal membranes. The results with **7** is consistent with the increase in inhibition of binding at the DAT when a phenyl was replaced by a 2-naphthyl group in (−)-3β-substituted ecgonine methyl esters,[15] piperidine-based analogues of cocaine,[16] and the work of Davies cited above.[14] The marked difference in activity between **6** and **7** in this series is most likely due to **6** existing almost exclusively in the keto tautomer, which has been shown[3] to reduce activity in substituted-aryl mazindol analogues. When the 2-naphthyl group in **7** was modified to the tetrahydronaphthyl **8**, a greater than 250-fold loss of activity occurred. This indicates that the Ring D binding of mazindol (Figure 1) favors only aromatic groups.

Table 2. Comparison of the Inhibition of Binding at the DAT on Guinea Pig and Rat Striatal Membranes and HEK HDAT Cells

	[3H] WIN 35,428		[125I] RTI-55	ratios		
	IC_{50}, nM		K_i, nM	rat	HEK	HEK
compd	g. pig[a]	rat	HEK hDAT[b]	g. pig	g. pig	rat
1	42.6 ± 2.6	12.94 ± 1.15	45 ± 1	0.3	1.1	3.5
7	4.1 ± 0.3	23.26 ± 3.26	45 ± 20	5.7	11.0	1.9
13	2.7 ± 0.2	34.20 ± 1.42	152 ± 40	12.7	56.3	4.4
24	10.6 ± 1.1	15.97 ± 2.19	27.2 ± 6.5	1.5	2.6	1.7
25	3.3 ± 0.2	38.11 ± 6.29	2.83 ± 0.60	11.6	0.9	0.07
28	2.1 ± 0.2	37.27 ± 1.53	2.01 ± 0.85	17.8	1.0	0.05

[a] Data from Table 1. [b] Data from Table 3.

Addition of a benzene ring at the 2,3-position (**11** and **14**) of mazindol and homomazindol and the 3,4-position (**16**) of bishomomazindol all resulted in a large loss (100−500-fold) in binding activity, whereas addition of a cyclohexane ring as a cis (**12**) or trans (**13**) isomer gave a 5- and 16-fold increase. The loss of activity in **11** and **14** is most likely due to the conversion of the strongly basic imine nitrogen in mazindol and homomazindol to the weakly basic anilino nitrogen. Although **16** contains a basic imine group, the benzene group at the 3,4-position likely extends into a binding site pocket that appears to tolerate only aliphatic interactions. Evidence for this is demonstrated by the enhanced activity shown by the cyclohexano analogues **12** and **13**.

The 5-fold loss of activity with 6,7-benzo analogue **19** of mazindol is possibly due to some interference with the ability of the OH group to interact at its binding site. Compound **22**, the 7,8-benzo analogue of mazindol showed a slight (1.3-fold) increase in activity, while **24**, the 8,9-benzo analogue, gave a 4-fold increase. Replacement of the *p*-chlorophenyl in **24** by a 2-naphthyl group (**25**) enhanced the activity of mazindol by 13-fold. However, when compared with **7**, the 2-naphthyl analogue of mazindol, the addition of the 8,9-benzo ring increased activity by 1.3-fold. When the benzo ring was added to the 9,10-position of homomazindol, there was a 20-fold increase in binding relative to mazindol, but only a 2-fold increase compared to homomazindol. In summary, adding a benzo ring at the 6,7-, 7,8-, or 8,9-positions in mazindol, or the 9,10-position in homomazindol, gave a modest increase (1.3−4-fold) or decrease (5-fold) in binding.

Addition of a trans-cyclohexano ring to the 2,3-position of **24** to form **30** resulted in a 500-fold decrease in activity, while it gave a 15-fold increase (**13**) when added to mazindol. The likely cause of this loss in activity is that the addition of a second ring to the heterocyclic nucleus of mazindol or homomazindol results in a molecule too large to fit in the same binding pocket. This is further demonstrated in compounds **32** and **33**, where the addition of a second benzo ring in the 6,7-position of **24** and **25** resulted in a >500-fold loss of activity.

Previous studies[4] from our laboratories have shown that mazindol, homomazindol, and bishomomazindol displace WIN binding on guinea pig and rat membranes in the same order (**3** > **2** > **1**). To determine if there is any correlation between the activity found on the guinea pig versus rat striatal membranes, we evaluated five of the most active compounds (**7**, **13**, **24**, **25**, and **28**) for their ability to displace WIN binding on rat striatal membranes (Table 2).

In contrast to the guinea pig, these compounds were less potent than mazindol in displacing WIN binding on the rat membranes. In addition, the order of activity and the difference between the most and least active compound was smaller. On the guinea pig membranes, the activity is **28** > **13** > **25** > **7** > **24**, and the rat membrane pattern of activity is **24** > **7** > **13** > **28** = **25**. Only compound **24** gave similar IC_{50} values, differing by 1.5-fold. The greater variation of the present compounds to fit in the guinea pig and rat DAT binding pockets in the same sequence may be due to the larger change in the molecular shape, size, or lipophilicity than that of **1**−**3**, where only the size of ring A was changed.

The selectivity of the five compounds to bind to the DAT, SERT, and NET and their discrimination for uptake inhibition at these transporters were measured by displacement of [125I] RTI-55 binding and blocking the uptake of [3H] DA, [3H] 5-HT, and [3H] NE on HEK cells expressing cDNA for the human transporters (Tables 3 and 4).

All of the new compounds, except **13**, were more effective in inhibition of RTI binding than blocking DA uptake at the DAT. Compounds **25** and **28** were potent inhibitors of RTI binding with K_i values in the same range as those of **2** and ca. 20-fold greater than those of mazindol (**1**). In addition, **25** and **28** gave the best discrimination ratios of 311 and 182, respectively, a considerable improvement over the 0.9 and 2.2 ratios for mazindol (**1**) and homomazindol (**2**).

Displacement of RTI binding on the HEK-hDAT cells also showed a pattern of activity different from that of WIN on guinea pig and rat striatal membranes with **28** > **25** > **24** > **7** > **13**. Good correlation between HEK and guinea pig activity was found with **1**, **24**, **25**, and **28**, whereas only **7** and **24** correlated with HEK and rat activity.

At both the SERT and NET, **7**, **25**, and **28** were more effective than mazindol (**1**) or homomazindol (**2**) in inhibiting the binding of RTI. However, none of the compounds showed a better selectivity ratio than **2**. Compounds **25** and **28** did show better discrimination ratios of 33.6 and 127.3 compared to 1.9 and 1.4 for **1** and **2** at the SERT and 7.3 and 10.0 compared to 0.3 for both **1** and **2** at the NET. It is interesting to note that **7**, where the *p*-chlorophenyl group in mazindol is replaced by a 2-naphthyl group, is the most potent inhibitor of RTI binding and 5-HT uptake at the SERT. Similar findings have also been found in tropane[14−16] and piperidine[17] series, where the replacement of an aryl group by a 2-naphthyl has resulted in an increase of selectivity for the cocaine binding site on the SERT.

Table 3. Inhibition of [^{125}I] RTI-55 Binding at the DA, 5-HT, and NE Transporter Sites on HEK-hDAT, HEK-hSERT, and HEK-hNET Cells[a]

compd	binding,[b] K_i, nM			selectivity ratio	
	hDAT	hSERT	hNET	SERT/DAT	NET/DAT
1	45 ± 1	50 ± 15	18 ± 12	1.1	0.4
2	1.7 ± 0.8	39 ± 11	18.2 ± 2	22.9	10.7
7	45 ± 20	2.3 ± 1	11 ± 1	0.05	0.24
13	152 ± 40	600 ± 230	690 ± 170	3.9	4.1
24	27.2 ± 6.5	224 ± 84	33.4 ± 72	8.2	1.2
25	2.83 ± 0.60	11.0 ± 4.1	5.2 ± 2.1	3.9	1.9
28	2.01 ± 0.85	5.5 ± 2.0	5.3 ± 2.1	2.8	2.6
cocaine[c]	960 ± 40	384 ± 196	1530 ± 300	0.4	1.6
cocaine[d]	419 ± 36	297 ± 32	645 ± 87	0.7	1.5
cocaine[e]	640 ± 30	270 ± 59	1525 ± 280	0.4	2.4

[a] See Experimental Section for details. [b] Values are mean ± standard error of two or three independent experiments, each conducted with triplicate determinations. [c] Cocaine as reference for **1** and **7**. [d]Cocaine as the reference for **2**, **13**, and **24**. [e] Cocaine as the reference for **25** and **28**.

Table 4. Inhibition of DA, 5-HT, and NE Uptake to HEK-hDAT, HEK-hSERT, andHEK-hNET Cells[a]

compd	uptake,[b] IC_{50}, nM			discrimination ratio uptake/binding[c]		
	hDAT	hSERT	hNET	DAT	SERT	NET
1	43 ± 20	94 ± 32	4.9 ± 0.5	0.9	1.9	0.3
2	3.7 ± 0.4	53 ± 7	4.9 ± 0.5	2.2	1.4	0.3
7	66 ± 10	1.8 ± 1.3	4.5 ± 1.5	1.5	0.8	0.4
13	63 ± 25	>10 μM	323 ± 91	0.4	>17	0.5
24	85 ± 34	3,210 ± 870	13.0 ± 1.7	3.2	14.3	0.4
25	880 ± 210	370 ± 40	38 ± 12	311	34	7
28	366 ± 82	700 ± 160	53 ± 14	182	127	10
cocaine[d]	168 ± 32	300 ± 101	583 ± 178	0.2	0.8	0.4
cocaine[e]	315 ± 78	420 ± 130	220 ± 76	0.8	1.4	0.3
cocaine[f]	330 ± 58	325 ± 171	192 ± 61	0.5	1.2	0.1

[a] See Experimental Section for details. [b] Values are mean ± standard error of two or three independent experiments, each conducted with triplicate determinations. [c] See Table 3 for binding values. [d] Cocaine as the reference for **1** and **7**. [e] Cocaine as the reference for **2**, **13**, and **24**. [f] Cocaine as the reference for **25** and **28**.

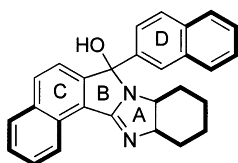


Figure 2. Sites (bold lines) where addition of a benzo or cyclohexano ring enhanced binding at the DAT or DA uptake inhibition of mazindol (**1**) or homomazindol (**2**).

Conclusions

The addition of a benzo ring at certain sites on rings C and D and a cyclohexano ring on ring A of mazindol (**1**) or homomazindol (**2**) produced several compounds (**7**, **13**, **24**, **25**, and **28**) with potent DAT binding and DA uptake inhibition (Figure 2). Some of these, in particular **25** and **28**, showed a considerable increase in uptake/binding discrimination ratios over **1** and **2** at the DAT, SERT, and NET. In vivo studies used to define potential cocaine treatment agents are in progress or completed on **7**, **13**, **24**, **25** and **28** and will be published elsewhere.

Experimental Section

Melting points were determined in a Thomas-Hoover capillary melting point apparatus and are not corrected. Analyses were performed by the Robertson Microlit Laboratories, Inc., Madison, NJ, and are within ±0.4% of theory, unless otherwise noted. Proton (^{1}H) and carbon (^{13}C) nuclear magnetic resonance (NMR) spectra were recorded at 300 or 500 and 75.5 MHz respectively, on a Bruker DPX-300 or DRX-500 spectrometer. Chemical shifts (δ) are reported in parts per million (ppm) downfield from tetramethylsilane (TMS) as the internal standard. The ultraviolet spectra (UV) were obtained in 95% EtOH on a Shimadzu Model UV-2101 PC ultraviolet spectrometer. Mass spectra (MS) were determined on a Micromass Quattro II mass spectrometer using APCI or ES ionization modes (positive or negative) and 0.2% ammonium formate in 50% aqueous acetonitrile as solvent. Thin-layer chromatography (TLC) was carried out on all compounds using glass plates coated with silica gel HF-254 (E. Merck, AG). If not otherwise specified, chemicals were obtained from the Aldrich Chemical Co.

5-(1-Naphthyl)-2,3-dihydro-5H-imidazo-[2, 1-a]-isoindol-5-ol (6) and 1-Naphthyl-[2-(4,5-dihydro-1H-imidazol-2yl)-phenyl]-methanone (6a). To a stirred solution of 2.5M n-BuLi in hexane (4.11 mL, 0.01 mol), *N,N,N',N'*-tetramethylethylenediamine (1.19 g, 1.54 mL, 0.01 mol) and THF (10 mL) under a N_2 atmosphere was added dropwise at room temperature a solution of 2-phenylimidazoline (0.68 g, 0.005 mol) in THF (10 mL). After stirring at room temperature for 2 h, the solid was treated dropwise with a solution of methyl 1-naphthoate (1.30 g, 0.007 mol) in THF (10 mL) and then stirred overnight. The mixture was cooled in an icebath and treated dropwise with saturated NH_4Cl solution (2.0 mL), followed by anhydrous $MgSO_4$ (ca. 10 g). The resultant solid was filtered and washed with THF (10 mL), and the combined filtrates were concentrated in vacuo to give 0.27 g (20%) of **6** and **6a**, mp 209−210 °C dec (CH_3OH/DMF); ^{1}H NMR (DMSO-d_6) δ 3.35 (s, 4H), 4.09 (m, 0.05H), 4.23 (m, 0.05H), 6.83 (brs, 1H), 7.13 (d, 1H), 7.39 (t, 1H), 7.62 (m, 2H), 7.68 (m, 5H), 7.98 (d, 1H), 8.06 (d, 1H), 8.72 (d, 1H); ^{13}C NMR (DMSO-d_6) δ 60.00, 122.37, 124.13, 124.95, 125.54, 126.16, 126.63, 126.98, 127.35, 127.39, 127.85, 128.74, 129.36, 129.76, 130.19, 130.25, 131.02, 131.31, 131.61, 133.20, 136.24, 141.11, 163.07 (C=N), 197.44 (C=O); MS *m/z* 301 (MH$^+$). Anal. ($C_{20}H_{16}N_2O$) C, H, N.

5-(2-Naphthyl)-2,3-dihydro-5H-imidazo-[2,1-a]-isoindol-5-ol (7). Following the procedure given for **6** and **6a**, but using methyl-2-naphthoate, gave **7** (29%), mp 185−186 °C dec (CH_3OH/DMF); ^{1}H NMR (DMSO-d_6) δ 2.93 (m, 2H), 4.18 (m, 2H), 6.95 (s, 1H), 7.29 (d, 2H), 7.46−8.01 (m, 7H), 8.11 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 42.3, 60.9, 88.9 (C-5), 122.9, 124.8, 125.1, 125.2, 125.7, 127.1, 128.0, 128.3, 128.8, 129.1, 129.7, 130.9, 133.3, 133.6, 139.3, 155.3, 167.9 (C=N); MS *m/z* 301 (MH$^+$). Anal. ($C_{20}H_{16}N_2O$) C, H, N.

5-(5,6,7,8-Tetrahydro-2-naphthyl)-2,3-dihydro-5H-imidazo-[2,1-a]-isoindol-5-ol (8). Following the procedure for **6** and **6a**, but using methyl 5,6,7,8-tetrahydro-2-naphththoate, gave **8**, (33%), mp 183−184 °C, dec (CH_3OH/DMF); ^{1}H (DMSO-d_6) δ 1.73 (m, 4H), 2.74 (m, 4H), 2.90 (q, 1H), 3.30 (q, 1H), 4.13 (m, 2H), 6.90 (t, 1H), 7.08 (s, 1H), 7.23 (t, 2H), 7.44 (m, 2H), 7.57 (m, 1H), 7.67 (m, 1H); ^{13}C NMR (DMSO-d_6) δ 22.45, 22.58, 28.72, 28.81, 41.36, 59.88, 87.99 (C-5), 125.80, 127.07,

127.47, 129.08, 129.52, 130.20, 131.23, 136.12, 136.40, 136.47, 137.93, 154.79, 167.08 (C=N); MS *m/z* 30.5 (MH^+). Anal. ($C_{20}H_{20}N_2O$) C, H, N.

11-(4-Chlorophenyl)-11H-isoindolo-[2,1-a]-benzoimidazole (10). To a stirred solution of 1,2-diaminobenzene (2.12 g, 0.02 mol) in concd. HCl (5 mL) was added slowly a solution of 2-(4-chlorobenzoyl)-benzaldehyde (4.89 g, 0.02 mol) in acetic acid (100 mL). The mixture was heated to 80 °C for 0.5h and then concentrated in vacuo. The residue was treated with 2N NaOH until basic to litmus paper, and the resultant solid was filtered off and crystallized from 2-propanol to give 2.15 g (34%) of **10**, mp 199−202 °C; ^{1}H NMR (DMSO-d_6) δ 6.00 (s, 1H: H-11), 7.00−7.62 (m, 10H), 7.8−8.2 (d, 2H); MS *m/z* 317 (MH^+). Anal. ($C_{20}H_{13}ClN_2$) C, H, N.

11-(4-Chlorophenyl)-11H-isoindolo-[2,1-a]-benzimidazol-11-ol (11). To a stirred solution of **10** (1.00 g, 0.032 mol) in dry DMF (15 mL) under a N_2 atmosphere was added portionwise sodium hydride (0.17 g, 0.07mol as 60% in mineral oil). After ca. 0.5h stirring at room temperature, the N_2 was stopped and a slow stream of dry air was bubbled into the solution for ca. 6h. The mixture was treated with CH_3OH (0.5 mL) and then H_2O (75 mL). The resultant solid was filtered to give 0.64 g (61%) of **11**, mp 220−221 °C (lit,[8] mp 222−223 °C).

(±)-*cis*-11-(4-Chlorophenyl)-5a,6,7,8,9,9a-hexahydro-11H-isoindolo-[2,1-a]-benzimidazol-11-ol (12). A mixture of 2-(4-chlorobenzoyl) benzaldehyde (1.44 g, 0.006 mol), *cis*-1, 2-diaminocyclohexane (1.0 g, 0.009 mol), and xylene (50 mL) was stirred and refluxed in a flask equipped with a Dean−Stark water separator until the H_2O layer in the sidearm remained constant (ca. 5 h). The mixture was concentrated in vacuo and the residue dissolved in MeOH (20 mL) and stirred at room temperature in the presence of air for 96h. The solid was filtered off to give 0.57 g (28%) of **12**, mp 183−184 °C (CH_3-OH/DMF); ^{1}H NMR (DMSO-d_6) δ 1.01−1.65 (m, 6H), 1.88 (m, 2H), 2.98 (m, 0.5H), 3.78 (m, 0.5H), 3.95 (quint., 0.5H), 4.08 (quint., 0.5H), 6.78 (s, 0.5H; OH), 7.00 (s, 0.5H; OH), 7.05 (t, 0.5H), 7.09 (d, 1H), 7.29 (d, 0.5H), 7.42 (m, 3H), 7.52 (m, 1H), 7.63 (d, 1H), 7.67 (t, 0.5H), 7.73 (m, 0.5H); ^{13}C NMR (DMSO-d_6) δ 19.88, 20.35, 21.83, 22.01, 24.46,25.18, 30.47, 54.60, 56.02, 68.70, 68.87, 87.58 and 88.34 (C-11), 121.72, 121.95, 123.59, 123.79, 127.47, 127.75, 127.82, 127.87, 127.92, 128.27, 128.38, 128.85, 129.00, 129.94, 131.29, 131.60, 132.22, 136.88, 138.26, 141.96, 154.33, 154.36, 165.81 and 167.86 (C=N); MS *m/z* 339 (MH^+). Anal. ($C_{20}H_{19}ClN_2O$) C, H, N.

(±)-*trans*-11-(4-Chlorophenyl)-5a,6,7,8,9,9a-hexahydro-11H-isoindolo-[2,1-a]-benzimidazol-11-ol (13). Prepared by the procedure of Aeberli,[9] et al. ^{1}H NMR (DMSO-d_6) δ 1.13−1.67 (m, 5H), 1.78 (m, 2H), 2.04 (t, 0.4H), 2.27 (t, 0.6H), 3.07 (t, 0.6H), 3.43 (t, 0.4H), 3.58 (m, 1H), 6.82 (s, 0.5H; OH), 7.02 (d, 0.5H), 7.09 (s, 0.5H; OH), 7.18 (t, 0.5H), 7.32 (d, 0.5H), 7.44 (m, 3H), 7.55 (quint, 1H), 7.68 (m, 2.5H); ^{13}C NMR (DMSO-d_6) δ 23.86, 23.95, 24.76, 25.08, 29.38, 29.68, 30.97, 31.18, 63.55, 65.74, 87.82 and 89.04 (C-11), 121.69, 122.07, 123.77, 124.07, 127.25, 127.73, 127.93, 127.95, 128.18, 128.96, 129.19, 129.84, 131.41, 131.92, 132.29, 132.38, 137.79, 141.93, 153.44, 153.82, 166.85 and 168.62 (C=N); MS *m/z* 339 (MH^+).

12-(4-Chlorophenyl)-10,12-dihydro-isoindolo-[1, 2-b]-quinazolin-12-ol (14). A mixture of 2-(4-chlorobenzoyl)-benzaldehyde (4.89 g, 0.02 mol), 2-aminobenzylamine (3.67 g, 0.03 mol) and xylene (150 mL) was stirred and refluxed in a flask equipped with a Dean−Stark water separator until the H_2O layer in the sidearm remained constant. The solution was concentrated in vacuo and the semisolid residue treated with *i*-PrOH (75 mL) and stirred at room temperature in the presence of air for ca. 56h. The resultant light yellow solid was filtered off to give 2.68 g (39%) of **14**, mp > 225 °C (i-PrOH/DMF); ^{1}H NMR (CD_3OD) δ 4.13 (d, 1H), 4.62 (d, 1H), 7.02 (m, 2H), 7.12 (s, 1H; OH), 7.17 (d, 1H), 7.28 (t, 1H), 7.45 (m, 4H), 7.53 (t, 1H), 7.87 (t, 1H); ^{13}C NMR (DMSO-d_6) δ 92.86 (C-12), 121.57, 122.80, 123.72, 125.55, 127.75, 128.86, 128.93, 129.12, 129.51, 130.05, 132.50, 132.73, 133.74, 138.83, 143.74, 148.49, 155.53 (C=N); UV λ max 203 nm (° 29,500) 224 (22,463), 275 (4,060), 292 (5,954), 305 (7,037), 318 (6,225) 332 (5,572), 356 (2,479); MS *m/z* 347 (MH^+). Anal. ($C_{21}H_{15}ClN_2O$) C, H, N.

13-(4-Chlorophenyl)-6,13-dihydro-11H-isoindolo-[2,1-b]-[2,4]-benzodiazepin-13-ol (16). Following the procedure used to prepare **14**, but using 1, 2-benzenedimethanamine, gave **16** (46%), mp > 225 °C (i-PrOH/DMF); ^{1}H NMR (CD_3-OD) δ 4.38 (d, 1H), 4.49 (d, 1H), 4.70 (d, 1H), 4.85 (d, 1H), 6.93 (t, 2H), 7.12 (t, 1H), 7.19 (t, 1H), 7.27 (m, 2H), 7.35 (m, 5H), 7.58 (t, 1H); ^{13}C NMR (DMSO-d_6) δ 45.23, 50.51, 93.81 (C-13), 122.06, 123.23, 128.02, 128.54, 128.61, 128.98, 129.19, 129.46, 131.26, 133.37, 135.19, 137.78, 141.32, 141.76, 146.70, 153.93 (C=N); UV λ max 201 nm (ϵ 21,500), 272 (1,820); MS *m/z* 361 (MH^+). Anal. ($C_{22}H_{17}ClN_2O$) C, H, N.

2-(2-Naphthyl)-4,5-dihydro-1H-imidazole (18). A solution of iodomethane (31.7 mL, 72.4 g, 0.51 mol), and 4,4-dimethyl-2-(2-naphthyl)-4,5-dihydro-oxazole[10] (**17**: 11.5 g, 0.051 mol) under a N_2 atmosphere was stirred at room temperature for 6 days. The resultant solid was filtered off to give 10.3 g (55%) of **17**·MeI; MS *m/z* 241 (MH^+).

A solution of **17**·MeI (10.0 g, 0.027 mol) and 1,2-diaminoethane (2.0 mL, 1.80 g, 0.03 mol) was stirred and refluxed for 10h, cooled in an icebath, and treated dropwise with 2N KOH (50 mL) and toluene (50 mL). The organic layer was separated, dried ($MgSO_4$), and filtered, and the filtrate was concentrated in vacuo to give 1.73 g (33%) of **18** mp 105−106 °C (acetone); ^{1}H NMR ($CDCl_3$) δ 3.91 (bs, 4H), 7.47 (m, 2H), 9.30 (m, 4H), 8.20 (s, 1H); MS *m/z* 197 (MH^+). Anal. ($C_{13}H_{12}N_2$) C, H, N.

5-(4-Chlorophenyl)-2,3-dihydro-5H-benz-[e]-imidazo-[2,1-a]-isoindol-5-ol (19). To a stirred solution of 1.6M n-BuLi in hexanes (8 mL, 0.013 mol) under a N_2 atmosphere was added dropwise a solution of **18** (1.00 g, 0.005 mol) in THF (15 mL). After an additional 15 min, a solution of methyl 4-chlorobenzoate (1.75 g, 0.01 mol) in THF (15 mL) was added dropwise and allowed to stir overnight. The mixture was treated dropwise with saturated NH_4Cl solution (5 mL), and H_2O (10 mL), and after 15 min, the resultant solid was filtered off to give 0.41 g (25%) of a mixture of **19** and **22**, mp 193−196 °C; Rf 0.24 and 0.29 (5% MeOH/CH_2Cl_2); ^{1}H NMR (DMSO-d_6) δ 2.63 (q, 0.5H), 3.01 (q, 0.5H), 3.38 (m, 1.0H), 4.14 (q, 0.4H), 4.23 (m, 0.6H), 6.91 (s), 7.02 (s), 7.18 (s), 7.23 (d), 7.38 (d), 7.42−7.48 (m), 7.58 (m), 7.68 (m), 7.75 (s), 7.81 (d), 7.87 (d), 7.97 (m), 8.02 (s), 8.05 (d), 8.10 (m), 8.32 (s), 8.33 (s); ^{13}C NMR (DMSO-d_6) δ 41.1, 42.0, 60.8, 60.9, 88.9, 89.3, 119.5, 122.8, 123.6, 125.1, 126.1, 127.6, 128.0, 128.3, 128.5, 128.7, 128.9, 129.2, 129.3, 129.8, 129.9, 130.9, 131.2, 133.7, 135.2, 135.2, 135.9, 140.1, 141.5, 150.8, 151.3, 167.1, 167.7; MS *m/z* 335 (MH^+).

A 30 mg sample of **19** and **22** was dissolved in CH_2Cl_2 (15 mL) and spotted on 25 plates of 20 × 20 cm Fisherbland plates 500 μM thickness and developed with 7% MeOH/CH_2Cl_2. The upper spot was isolated to give 17 mg of **19**, mp 194−196 °C dec, R_f = 0.24 (5% MeOH/CH_2Cl_2); ^{1}H NMR (DMSO-d_6) δ 2.68 (d, 1H), 3.38 (bs, 1H), 4.15 (bs, 2H), 7.01 (bs, 1H: OH), 7.28 (bs, 2H), 7.38 (d, 2H), 7.48 (t, 1H), 7.57 (t, 1H), 7.82 (d, 1H), 7.88 (bs, 1H), 8.05 (d, 1H), 8.10 (d, 1H); ^{13}C NMR (DMSO-d_6) δ 42.00, 60.52, 88.94 (C-5), 119.08, 124.76, 126.02, 127.26, 127.48, 128.34, 128.81, 129.27, 130.63, 133.20, 135.51, 139.92, 150.52, 167.75 (C=N); MS *m/z* 335 (MH^+). Anal. ($C_{20}H_{15}ClN_2O$) C, H, N.

11b-(4-Chlorophenyl)-1,2,3,11b-tetrahydro-5H-benz-[f]-imidazo-[2,1-a]-isoindol-5-one (21). A mixture of 3-(4-chlorobenzoyl)-2-naphthoic acid (4.00 g, 0.013 mol), 1,2-diaminoethane (1.55 g, 1.72 mL, 0.026 mol), and xylene (100 mL) was stirred and refluxed in a flask equipped with a Dean−Stark water separator until the H_2O layer in the sidearm remained constant (ca. 7h). The mixture was stirred overnight at room temperature, and the resultant solid was filtered off to give 3.85 g (90%) of **21**, mp 213−215 °C (*i*-PrOH) (lit.[11] mp 217−219 °C); ^{1}H NMR (DMSO-d_6) δ 2.18 (brs, 1H; NH), 3.21 (m, 2H), 3.68 (m, 1H), 3.91 (m, 1H), 7.35 (d, 2H), 7.53 (m, 2H); 7.68 (m, 3H), 7.79 (t, 1H), 7.95 (t, 1H), 8.35 (s, 1H); MS *m/z* 335 (MH^+).

5-(4-Chlorophenyl)-2,3-dihydro-5H-benz-[f]-imidazo-[2,1-a]-isoindol-5-ol (22). To a stirred mixture of $LiAlH_4$

(0.144 g, 0.004 mol) and THF (25 mL) under a N_2 atmosphere and cooled in an icebath was added dropwise a solution of **21** (1.005 g, 0.003 mol) in THF (15 mL) at such a rate that the internal temperature did not excede 18 °C. After an additional 15 min, it was treated with 2N KOH (0.45 mL), H_2O (0.6 mL), and anhydr. Na_2SO_4 (4 g) and stirred overnight at room temperature. The solid was filtered off to give 0.257 g (25%) of **22**, R_f = 0.29 (5% MeOH/CH_2Cl_2) mp 194−195 °C (lit.[11] mp 224−226 °C); ^{1}H NMR (DMSO-d_6) δ 3.01 (q, 1H), 3.38 (m, 1H), 4.21 (m, 2H), 6.79 (s, 1H; OH), 7.43 (A_2B_2, 4H), 7.58 (m, 2H), 7.75 (s, 1H), 7.93 (m, 1H), 8.11 (m, 1H), 8.33 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 41.26, 60.08, 87.58 (C-5), 122.02, 122.85, 125.26, 126.86, 127.58, 128.18, 128.45, 128.58, 129.16, 132.57, 132.95, 134.47, 140.71, 150.53, 166.42 (C=N); MS *m*/*z* 335 (MH^+). Anal. ($C_{20}H_{15}ClN_2O$) C, H, N.

5-(4-Chlorophenyl)-2,3-dihydro-5H-benz-[g]-imidazo-[2,1-a]-isoindol-5-ol (24). To a stirred solution of 2-(1-naphthyl)-4, 5-dihydro-1H-imidazole[13] (**23**: 1.26 g, 0.0064 mol) in THF under a N_2 atmosphere was added at room temperature a solution of 1.6 M n-BuLi in hexanes (10 mL, 0.016 mol). After an additional 2h stirring, the solution was treated dropwise with a solution of methyl-4-chlorobenzoate (2.18 g, 0.013 mol). The mixture was cooled in an icebath and treated dropwise with a saturated NH_4Cl solution (3 mL), H_2O (4 mL) and stirred overnight at room temperature. The resultant solid was filtered off to give 0.462 g (22%) of **24**, mp 200−201 °C dec (i-PrOH/DMF); ^{1}H NMR (DMSO-d_6) δ 2.93 (q, 1H), 3.36 (q, 1H), 4.28 (m, 2H), 6.98 (s, 1H; OH), 7.38 (d, 1H), 7.44 (A_2B_2, 4H), 7.63 (t, 1H), 7.72 (t, 1H), 8.05 (t, 2H), 8.27 (d, 1H); ^{13}C NMR (DMSO-d_6) δ 41.93, 61.48, 88.68 (C-5), 121.88, 123.04, 125.43, 127.74, 128.24, 128.87, 129.02, 129.24, 129.31, 132.88, 133.42, 133.78, 140.46, 154.15, 168.56 (C=N); MS *m*/*z* 335 (MH^+). Anal. ($C_{20}H_{15}ClN_2O$) C, H, N.

5-(2-Naphthyl)-2,3-dihydro-5H-benzo-[g]-imidazo-[2,1-a]-isoindol-5-ol (25). Following the procedure for **24**, but using methyl-2-naphthoate, gave **25** (33%), mp 218−219 °C dec (CH_2Cl_2/MeOH); ^{1}H NMR (DMSO-d_6) δ 2.93 (q, 1H), 4.32 (m, 2H), 7.01 (s, 1H; OH), 7.32 (d, 1H), 7.42 (d, 1H), 7.54 (m, 2H), 7.67 (t, 1H), 7.77 (t, 1H), 7.90 (m, 1H), 8.01 (t, 1H), 8.08 (d, 2H), 8.17 (s, 1H), 8.82 (d, 1H); ^{13}C NMR (DMSO-d_6) δ 41.21, 60.57, 88.21 (C-5), 121.18, 122.24, 124.22, 124.55, 125.01, 126.23, 126.78, 127.37, 127.89, 127.94, 128.21, 128.41, 131.91, 132.46, 132.74, 132.85, 137.87, 153.62, 167.87 (C=N); MS *m*/*z* 351 (MH^+). Anal. ($C_{24}H_{18}N_2O$) C, H, N.

2-(1-Naphthyl)-1,4,5,6-tetrahydropyrimidine (27). A stirred mixture of 1, 3-diaminopropane (14.80 g, 16.8 mL, 0.20 mol), 1-cyanonaphthalene (14.05 g, 0.09 mol), and *p*-toluenesulfonic acid monohydrate (19.1 g, 0.10 mol) was heated in an oil bath at 210−220 °C for 2.5 h. The viscous mixture was cooled to ca. 95 °C, treated with H_2O (100 mL), allowed to cool to room temperature, and then treated with 5N NaOH (75 mL). After stirring overnight, the solid was filtered off and crystallized from toluene to give 10.80 g (57%) of **27**, mp 152−154 °C; ^{1}H NMR ($CDCl_3$) δ 1.95 (quin, 2H), 3.55 (t, 4H), 7.40−7.58 (m, 4H), 7.84 (d, 2H), 8.26 (dd, 1H); MS *m*/*z* 211 (MH^+). Anal. ($C_{14}H_{14}N_2$) C, H, N.

6-(4-Chlorophenyl)-2,3,4,6-tetrahydro-benz-[g]-pyrimido-[2,1-a]-isoindol-6-ol (28). To a stirred solution of 1.6 M n-BuLi in hexanes (15.6 mL, 0.025 mol) under N_2 at room temperature, there was added dropwise a solution of **27** (2.10 g, 0.01 mol) in THF (40 mL). After an additional 2h, the mixture was treated dropwise with a solution of methyl 4-chlorobenzoate (4.26 g, 0.025 mol) in THF (25 mL), stirred overnight, and then treated with saturated NH_4Cl solution (7 mL). After stirring 2h, the resultant solid was filtered off to give 0.70 g (21%) of **28**, mp 220−221 °C; ^{1}H NMR (DMSO-d_6) δ 1.82 (m, 2H), 2.91 (m, 1H), 3.58 (m, 2H), 6.78 (s, 1H; OH), 7.28 (d, 1H), 7.42 (s, 4H), 7.57 (t, 1H), 7.62 (t, 1H), 7.94 (t, 2H), 9.40 (d, 1H); ^{13}C NMR (DMSO-d_6) δ 20.50, 36.55, 44.30, 90.73 (C-6), 119.99, 124.90, 126.16, 126.33, 127.36, 128.04, 128.25, 128.45, 131.03, 132.45, 133.22, 139.85, 146.57, 155.17, 162.30 (C=N), MS *m*/*z* 349 (MH^+). Anal. ($C_{21}H_{17}ClN_2O$) C, H, N.

***trans*-2-(1-Naphthyl)-3a,4,5,6,7,7a-hexahydro-1H-benzimidazole (29).** A solution of *trans*-1, 2-diaminocyclohexane (28.55 g, 30.70 mL, 0.25 mol), *p*-toluenesulfonic acid monohydrate (47.60 g, 0.25 mol) in H_2O (50 mL) was stirred at room temperature for ca. 23h, concentrated in vacuo to give 49.2 g (69%) of *trans*-1, 2-diaminocyclohexane·p-tol SO_3H acid salt, mp > 225 °C (i-PrOH); ^{1}H NMR (DMSO-d_6) δ 1.10 (d, 4H), 1.35 (d, 2H), 1.75 (d, 2H), 2.21 (s, 3H), 2.40 (t, 2H), 7.05 (d, 2H), 7.43d (2H). Anal. ($C_{13}H_{22}N_2O_3S$) C, H, N.

A stirred mixture of 1-cyanonaphthalene (7.66 g, 0.05 mol), and the above salt (15.75 g, 0.055 mol) was heated in an oilbath to 225 °C for 3h, allowed to cool to room temperature, dissolved in DMF (35 mL), poured into H_2O (100 mL), and stirred for ca. 12h. The solid was filtered off to give 7.75 g (37%) of **29**·p-TolSO_3H, mp 180−181 °C (DMF/i-PrOH); MS *m*/*z* 251 (MH^+). Anal. ($C_{24}H_{26}N_2O_3S$) C, H, N.

A mixture of **29**·p-TolSO_3H (6.32 g, 0.015 mol), 5N NaOH (20 mL) and CH_2Cl_2 (50 mL) was stirred at room temperature for ca. 2h. The CH_2Cl_2 layer was separated, dried ($MgSO_4$), filtered, and concentrated in vacuo to give 3.52 g (94%) of **29**, mp 184−185 °C (i-PrOH); ^{1}H NMR (DMSO-d_6) δ 1.35 (q, 2H), 1.52 (d, 2H), 1.82 (d, 2H), 2.21 (d, 2H), 3.05 (d, 2H), 7.00 (s, 1H: NH), 7.53 (m, 3H), 7.73 (d, 1H), 7.96 (m, 2H), 8.83 (q, 1H); MS *m*/*z* 251 (MH^+). Anal. ($C_{17}H_{18}N_2$) C, H, N.

(±)-*trans*-13-(4-Chlorophenyl)-7a,8,9,10,11,11a-hexahydro-13H-benzo-[e]-isoindolo-[2,1-a]-benzoimidazol-13-ol (30). To a stirred solution of 1.6M n-BuLi in hexanes (4.8 mL, 0.008 mol) under N_2 and cooled in an icebath was added dropwise a solution of **29** (1.00 g, 0.0035 mol) in THF (25 mL). After stirring an additional 15 min, the mixture was treated dropwise with a solution of methyl *p*-chlorobenzoate (1.19 g, 0.007 mol) in THF (10 mL) and then stirred overnight at room temperature. The mixture was treated dropwise with saturated NH_4Cl solution (5 mL), H_2O (5 mL), and $MgSO_4$ (5 g). The solids were filtered off and the filtrate concentrated in vacuo to give 0.44 g (32%) of **30**, mp 216−217 °C, dec (CH_3OH); ^{1}H NMR (DMSO-d_6) δ 1.28−1.51 (m, 2.5H), 1.64−1.75 (m, 3H), 1.88 (t, 2H), 2.42 (d, 1.0H), 3.18 (t, 0.5H), 3.65 (t, 0.5H), 3.76 (t, 0.5H), 6.96 (s, 0.5H: OH), 7.12 (d, 0.5H), 7.22 (s, 0.5H), 7.36 (d, 0.5H), 7.48 (m, 3H), 7.62−7.81 (m, 4H), 8.08 (d, 1H), 8.15 (d, 0.5H), 8.18 (d, 0.5H); ^{13}C NMR (DMSO-d_6) δ 24.8, 24.9, 25.7, 26.0, 30.3, 30.6, 31.9, 32.1, 64.6, 66.4, 78.2, 78.3, 88.9 and 90.1 (C-13), 121.8, 122.1, 123.2, 123.7, 125.5, 127.8, 127.9, 128.3, 128.9, 129.0, 129.1, 129.3, 129.4, 132.9, 133.4, 133.8, 138.1, 142.5, 153.5, 153.8, 168.5 and 170.4 (C=N); MS *m*/*z* 389 (MH^+). Anal. ($C_{24}H_{21}ClN_2O$) C, H, N.

2-(9-Phenanthryl)-4,5-dihydro-1H-imidazole (31). A solution of 9-cyanophenanthrene (4.07 g, 0.02 mol), 1,2-diaminoethane (1.04 g, 1.6 mL, 0.024 mol), *p*-toluene sulfonic acid·H_2O (2.28 g, 0.012 mol), and ethylene glycol (15 mL) under N_2 was stirred and refluxed for ca. 48 h. The mixture was treated at room temperature with 3% NaOH (100 mL) and stirred for 3 h, and the resultant solid filtered off to give 2.50 g (50%) of **31**, mp 190−191 °C; ^{1}H NMR (DMSO-d_6) δ 3.76 (bs, 4H), 6.98 (s, NH), 7.70 (m, 4H), 8.05 (d, 1H), 8.14 (s, H-10), 8.88 (m, 3H); ^{13}C NMR (DMSO-d_6) δ 122.76, 122.92, 126.69, 126.83, 127.04, 127.69, 127.82, 127.85, 128.93, 129.09, 129.87, 130.10, 130.21, 164.24 (C=N); MS *m*/*z* 247 (MH^+). Anal. ($C_{17}H_{14}N_2$), C, H, N.

5-(4-Chlorophenyl)-2,3-dihydro-5H-dibenz-[e.g]-imidazo-[1, 2-a]-isoindol-5-ol (32). To a stirred solution of **31** (1.23 g, 0.005 mol) and THF (20 mL) under argon was added dropwise at room temperature 1.6M n-BuLi in hexanes (9 mL, 0.014 mol). After an additional 15 min, the mixture was treated dropwise with a solution of methyl 4-chlorobenzoate (1.70 g, 0.01 mol) in THF (5 mL), stirred overnight, and then treated with saturated NH_4Cl solution (10 mL) and H_2O (5 mL). After ca. 15 min, the resultant solid was filtered off to give 0.76 g (39%) of **32**, mp 226 °C dec (CH_3OH/CH_2Cl_2); ^{1}H NMR (DMSO-d_6) δ 2.60 (q, 1H), 3.35 (q, 1H), 4.23 (q, 2H), 7.30 (s, 1H, OH), 7.35 (s, 1H), 7.39 (m, 3H), 7.60 (t, 1H), 7.75 (t, 1H), 7.84 (m, 2H), 8.02 (d, 1H), 8.95 (d, 1H), 9.05 (t, 1H); ^{13}C NMR (DMSO-d_6) δ 42.04, 60.75, 88.76 (C-5), 122.50, 123.79, 124.22, 125.88,

126.02, 126.07, 126.57, 127.48, 128.14, 128.22, 128.53, 128.91, 130.88, 132.25, 132.81, 139.13, 149.91, 167.79 (C=N); MS *m/z* 385 (MH+). Anal. ($C_{24}H_{17}ClN_2O$) C, H, N.

5-(2-Naphthyl)-2,3-dihydro-5H-dibenzo-[e.g]-imidazo-[1, 2-a]-isoindol-5-ol (33). Following the procedure for **32**, but using methyl 2-naphthoate, gave **33** (28%), mp 212−214 °C dec (CH_2Cl_2/MeOH); ^{1}H NMR (DMSO-d_6) δ 2.59 (q, 1H), 3.38 (q, 1H), 4.23 (m, 2H), 7.07 (d, 1H), 7.34 (s, 1H: OH), 7.48 (d, 3H), 7.66 (t, 1H), 7.78 (d, 1H), 7.83 (m, 4H), 7.95 (d, 1H), 8.08 (d, 1H), 8.18 (s, 1H), 8.88 (m, 2H), 9.07 (d, 1H); ^{13}C NMR (DMSO-d_6) δ 41.77, 60.35, 88.79 (C-5), 122.21, 123.40, 123.47, 123.75, 123.82, 125.00, 125.45, 125.75, 126.31, 126.38, 127.02, 127.33, 127.48, 127.68, 127.86, 128.16, 130.51, 131.84, 132.40, 132.78, 137.24, 149.85, 167.67 (C=N); MS *m/z* 351 (MH+). Anal. ($C_{28}H_{20}N_2O$) C, H, N.

[^{3}H]-WIN 35,428 Binding Assays. Rat. Brains from male Sprague−Dawley rats weighing 200−225 g (Taconic Labs) were removed, striatum dissected, and placed on ice. Membranes were prepared by homogenizing tissues in 20 volumes (w/v) of ice cold modified Krebs-HEPES buffer (15 mM HEPES, 127 mM NaCl, 5 mM KCl, 1.2 mM $MgSO_4$, 2.5 mM $CaCl_2$, 1.3 mM NaH_2PO_4, 10 mM D-glucose, pH adjusted to 7.4) using a Brinkman Polytron (setting 6 for 20 s.) and centrifuged at 20 000×*g* for 10 min at 4 °C. The resulting pellet was suspended in buffer, recentrifuged, and resuspended in buffer to a concentration of 10 mg/mL. Ligand binding experiments were conducted in assay tubes containing 0.5 mL modified Krebs-HEPES buffer for 60 min on ice. Each tube contained 1.5 nM [^{3}H] WIN 35,428 (specific activity 84 Ci/mmol) and 2.5 mg striatal tissue (original wet weight). Nonspecific binding was determined using 0.1 mM cocaine HCl. For determination of binding affinity, triplicate samples of membranes were preincubated for 5 min in the presence or absence of the compound being tested. Incubations were terminated by rapid filtration through Whatman GF/B filters, presoaked in 0.1% BSA, using a Brandel R48 filtering manifold (Brandel Instruments Gaithersburg, Maryland). The filters were washed twice with 5 mL cold buffer and transferred to scintillation vials. Beckman Ready Safe (3.0 mL) was added, and the vials were counted the next day using a Beckman 6000 liquid scintillation counter (Beckman Coulter Instruments, Fullerton, CA). Data were analyzed by using GraphPad Prism software (San Diego, CA).

B.Guinea Pig. Compounds listed in Table 1 were tested at NOVASCREEN, a division of Oceanix Biosciences Corporation, Hanover, MD. In brief, guinea pig striatal membranes, [^{3}H] WIN 35,428, and the test compound at concentrations of 10^{-10} to 10^{-6} M in DMSO/H_2O), were incubated with 50 mM TRIS−HCl (pH 7.4) containing 100 mM NaCl at 25 °C for 2 h. The reaction assay was terminated by rapid vacuum filtration onto glass fiber filters. Radioactivity trapped onto the fibers is determined and compared to control values in order to ascertain any interaction of test compound with the uptake site.

[^{125}I] RTI-55 Binding Assay. All test compounds were prepared as 10 mM stock solution in DMSO. Subsequent dilutions were made in assay buffer, achieving a final concentration of 0.1%. Pipetting was conducted using a Biomek 2000 robotic work station. HEK293 cells expressing hDAT, hSERT, or hNET inserts were grown to 80% confluence on 150 mm diameter tissue culture dishes and serve as the tissue source. The medium was poured off the plate, the plate was washed with 10 mL of calcium- and magnesium-free phosphate-buffered saline, and lysis buffer (10 mL; 2 mM HEPES with 1 mM EDTA) was added. After 10 min, cells were scraped from the plates, poured into centrifuge tubes, and centrifuged 30 000*g* for 20 min. The supernatant fluid was removed, and the pellet was resuspended in 12−32 mL of 0.32 M sucrose using a Polytron at setting 7 for 10 s. The resuspension volume depends on the density of binding sites within a cell line and is chosen to reflect binding of 10% or less of the total radioactivity. Each assay tube contained 50 μL of membrane preparation (about 10−15 μg of protein), 25 μL of test compound or buffer (Krebs-HEPES, pH 7.4; 122 mM NaCl, 2.5 mM $CaCl_2$, 1.2 mM $MgSO_4$, 10μM pargyline, 100 μM tropolone, 0.2% glucose and 0.02% ascorbic acid, buffered with 25 mM HEPES), 25 μL of [^{125}I] RTI-55 (40−80 pM final concentration), and additional buffer sufficient to bring the final volume to 250 μL. The membranes are preincubated with unknowns for 10 min prior to the addition of [^{125}I] RTI-55. The assay tubes were incubated at 25 °C for 90 min, and the binding was terminated by filtration over GF/C filters using a Tomtec 96-well cell harvester. Filters are washed for six seconds with ice-cold saline. Scintillation fluid was added to each square and radioactivity remaining on the filter was determined using a Wallac μ- or β-plate reader. Specific binding was defined as the difference in binding observed in the presence and absence of 5 μM mazindol (HEK-hDAT and HEK-hNET) or 5 μM imipramine (HEK-hSERT). Two or three independent competition experiments were conducted with duplicate determinations. GraphPAD Prism was used to analyze the ensuing data, with IC_{50} values converted to K_i values using the Cheng-Prusoff equation.

Inhibition of [^{3}H] Neurotransmitter Uptake in HEK293 Cells Expressing Recombinant Biogenic Amine Transporters. HEK293 cells expressing hDAT, hSERT, or hNET were grown to confluence as described above. The medium was removed, and the cells were washed twice with phosphate buffered saline (PBS) at room temperature. Following the addition of Krebs Hepes buffer (3 mL), the plates were warmed in a 25 °C water bath for 5 min. The cells were gently scraped and then triturated with a pipet. Cells from multiple plates were combined. One plate provides enough cells for 48 wells, which is required to generate data on two complete curves for the test compounds. Krebs-HEPES (350 μL) and test compounds (50 μL) were added to 1-mL vials and placed in a 25 °C water bath. Specific uptake is defined as the difference in uptake observed in the presence and absence of 5 μm imipramine (HEK-hSERT). Cells (50 μL) are added and preincubated with the unknowns for 10 min. The assay is initiated by the addition of [^{3}H] dopamine, [^{3}H] serotonin, or [^{3}H] norepinephrine (50 μL, 20 nM final concentration). Filtration through Whatman GF/C filters presoaked in 0.05% polyethylenimine is used to terminate uptake after 10 min. The IC_{50}s are calculated applying the GraphPAD Prism program to triplicate curves made up of 6 drug concentrations each. Two or three independent determinations of each curve are made. Additional details about this assay have been published.

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