Bioorganic & Medicinal Chemistry 16 (2008) 6675–6681

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry

journal homepage: www.elsevier.com/locate/bmc

Synthesis and pharmacological investigation of novel 2-aminothiazole-privileged aporphines ☆

Zhili Liu [a,†], Xuetao Chen [b,†], Leiping Yu [b], Xuechu Zhen [b,*], Ao Zhang [a,*]

[a] Synthetic Organic & Medicinal Chemistry Laboratory (SOMCL), Shanghai Institute of Materia Medica (SIMM), Chinese Academy of Sciences, Building 3, Room 426, 555 Zuchongzhi Road, Pudong, Shanghai 201203, China
[b] Neuropharmacological Laboratory, Shanghai Institute of Materia Medica (SIMM), Chinese Academy of Sciences, Shanghai 201203, China

ARTICLE INFO

Article history:
Received 23 April 2008
Revised 29 May 2008
Accepted 30 May 2008
Available online 5 June 2008

Keywords:
Aminothiazole
Apomorphine
Dopamine receptor
Hybridize
Serotonin receptor

ABSTRACT

A series of apomorphine ((−)-**1**, APO)-derived analogues ((±)-**3**, (−)-**4**-(−)-**6**) were designed and synthesized by hybridizing APO with a privileged 2-aminothiazole functionality which was lent from the orally available anti-parkinsonian drug, pramipexole (**2**). Among these hybridized compounds, catecholic aporphine (−)-**6** shows good affinity at the D_2 receptor with K_i of 328 nM, slightly less potent (3-fold), but more selective against the D_1 receptor than that of the parent compound, APO. Although possessing reduced affinity at the D_2 receptor, aporphines **15** and **18** show significant potency at both the D_1 and 5-HT_{1A} receptors. The former compound is equipotent at both receptors (K_i: 116 and 151 nM, respectively), while the latter is 8-fold more potent at the D_1 (K_i: 78 nM) than at the 5-HT_{1A} receptors (K_i: 640 nM). These results indicate that the catechol fragment is critical for the D_2 receptor binding of the anti-parkinsonian drug, APO ((−)-**1**), but not necessary for binding at the D_1 and 5-HT_{1A} receptors.

1. Introduction

Naturally occurring aporphine alkaloids and their synthetic derivatives have served as leads for the development of therapeutic agents for decades.[1–3] *R*-(−)-Apomorphine (APO, (−)-**1**), the semisynthetic[4,5] or total synthetic[6] prototypical aporphine, is a well-documented dopamine (DA) receptor agonist, and has been marketed for the treatment of Parkinson's disease in Europe since the 1990s and in US since 2004.[7,8] However, despite its high intrinsic agonism activity and fast onset of anti-parkinsonian effects, APO suffers from poor bioavailability, short duration of action and potential central emetic side-effects.[7–9] As part of our drug discovery program, we recently initiated an approach to developing novel aporphinoids by modification of the catechol moiety in this molecule. The rationale for this approach is on the basis of the generally accepted hypothesis that the catechol moiety influences the pharmacological profiles of APO in several ways.[1–3,10–12] Firstly, the catecholic function contributes to the high dopaminergic activity by H-bonding with DA receptors; on the other hand, the catechol fragment is not physico-chemically stable. Its high water solubility and oxidative potential contribute to APO's poor bioavailability and short duration of action. In this regard, it would be possible to improve APO's pharmacological profile by bioisosterically replacing the catecholic component with a more stable function group.[2,3] Our first effort was directed to the integration of a 2-aminothiazole function into the structure of APO (Fig. 1). The 2-aminothiazole fragment is a key pharmacophoric fragment of the widely prescribed orally stable anti-parkinsonian drug, pramipexole (**2**).[13–15] It is a widely accepted privileged structure with unique pharmacological property and has been extensively applied as a heterocyclic bioisostere of the phenol moiety.[16–18] We envisioned that replacement of the catechol component of APO with a 2-aminothiazole moiety would substantially retain the H-bonding ability and the aromaticity of the catechol moiety. Therefore, a series of aminothiazole-privileged structures ((±)-**3**, (−)-**4**-(−)-**6**) were designed and synthesized as shown in Figure 1.

Compound (±)-**3** contains a 2-aminothiazole that completely replaces the catechol moiety of APO ((−)-**1**). Compounds (−)-**4** and (−)-**5** represent the replacement of the catecholic hydroxy functions of APO by a thiazole moiety. In addition, a C2-methoxy, or a C2,C3-fused aminothiazole moiety is included to explore the hypothesized accessory binding sites around C2 or C3.[2,10,11] Compound (−)-**6** is prepared as a control to explore the effect of a C2,C3-fused aminothiazole moiety without changing the catechol group of APO. All compounds in this series possess the pharmacophoric fragments of both APO ((−)-**1**) and pramipexole (**2**), and thus can be viewed as hybrids of APO and pramipexole. In this

☆ Part of the results in this manuscript has been presented at the Gordon Research Conferences, Salve Regina University, USA, July 2, 2006, Poster 280.

* Corresponding authors. Tel./fax: +86 21 50806750 (X.Z.); tel.: +86 21 50806035; fax: +86 21 50806600 (A.Z.).
E-mail addresses: xczhen@mail.shcnc.ac.cn (X. Zhen), aozhang@mail.shcnc.ac.cn (A. Zhang).
† These authors contributed equally to this work.

0968-0896/$ - see front matter © 2008 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmc.2008.05.077

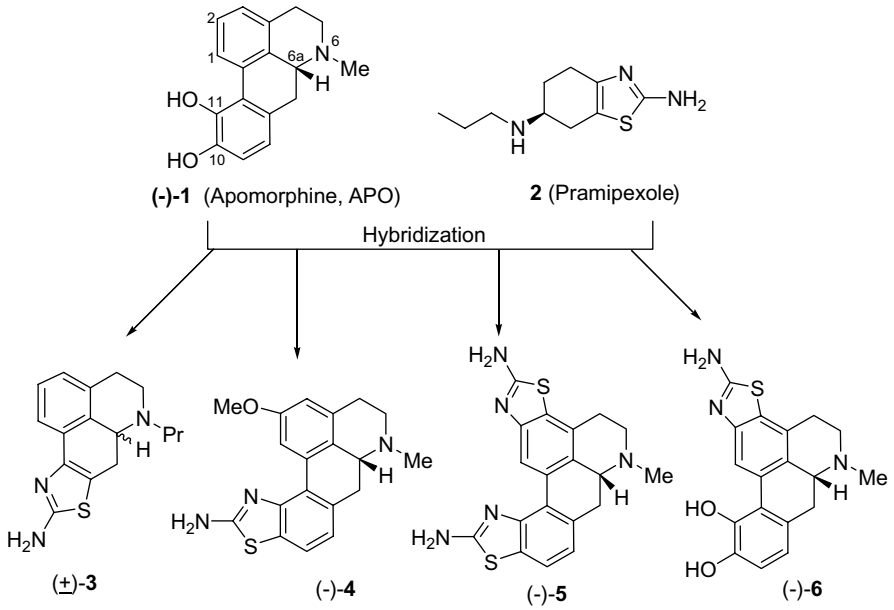


Figure 1. Apomorphine, pramipexole, and proposed 2-aminothiazole-privileged aporphines.

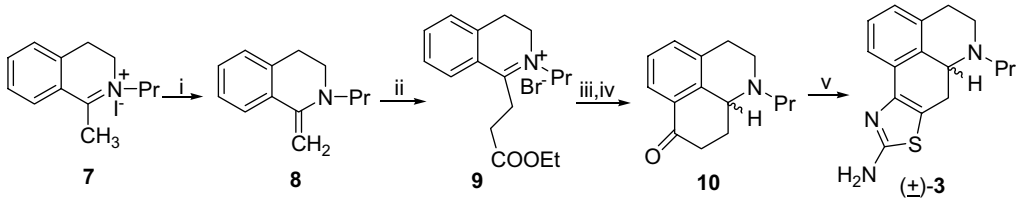
report, we present our synthetic efforts to these aminothiazole-privileged compounds and the results from our preliminary pharmacological investigation.

2. Chemistry

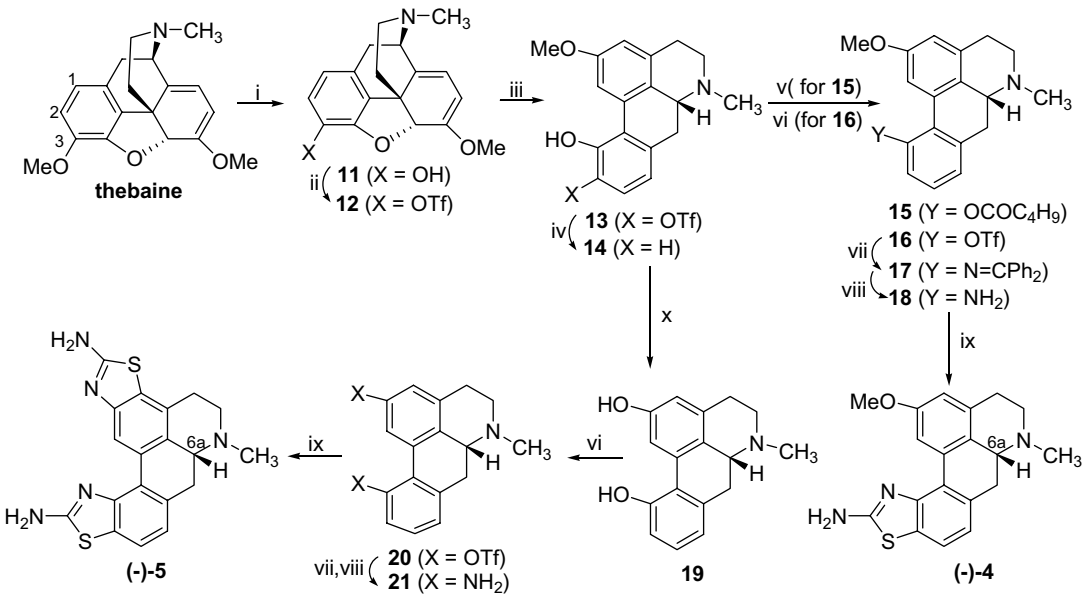
Synthesis of (±)-**3** was started from *N*-propyl-1-methyl-3,4-dihydro-isoquinoline iodide **7**, which was prepared according to a literature procedure[19–22] (Scheme 1). Treating salt **7** with 10% NaOH, followed by alkylation with ethyl bromoacetate yielded **9** in quantitative yield. $NaBH_4$ reduction followed by cyclization with polyphosphoric acid (PPA) gave the key intermediate ketone **10**.[19–22] Aminothiazole (±)-**3** was prepared by bromination of ketone **10** and then treating the resulting bromo-intermediate with thiourea in AcOH.[17,23] A similar preparative strategy had been reported in 1982 by Berney[20] and Schneider[21,22] who reported the preparation of a series of 5-membered heterocycle analogues of APO ((−)-**1**), including (±)-**3** and its *N*-methyl analogue. In their report, they stated that dopaminergic properties were observed from some of their compounds, but the detail was not disclosed, especially the property at each of the DA receptor subtypes. In our current report, we synthesized the *N*-propyl aminothiazole (±)-**3** as a racemate and evaluated its activity at both DA and serotonin receptors.

Using thebaine as starting material, aminothiazoles (−)-**4** and (−)-**5** were prepared in multiple steps (Scheme 2). The *R*-(−)-absolute configuration of C6a in these compounds is derived from natural alkaloid thebaine. Thus, treating thebaine with L-selectride (1 M)[24,25] selectively gave the 3-*O*-demethylated compound **11** (oripavine), which was triflated with Tf_2O and pyridine to yield compound **12**.[26,27] Treating triflate **12** with $MeSO_3H$ gave the rearranged aporphinic skeleton **13**.[26,27] Reduction of triflate **13** using Pd/C and Mg metal gave 11-hydroxy-aporphine **14**[10] in 41% yield following a literature procedure reported recently.[27,28] Aporphine **14** was reacted with Tf_2O and pyridine to afford triflate **16**, which was subsequently converted to imine **17** and then amine **18** by using a similar C–N coupling reaction as described before in 28% yield (2 steps).[16,17] Mono-hydroxyaporphine **14** was esterified[29] with valeric acid to yield valeroyl ester **15**. Treating amine **18** with Br_2 and KSCN yielded a dark complex, and the expected aminothiazole (−)-**4** was isolated in 20% yield.

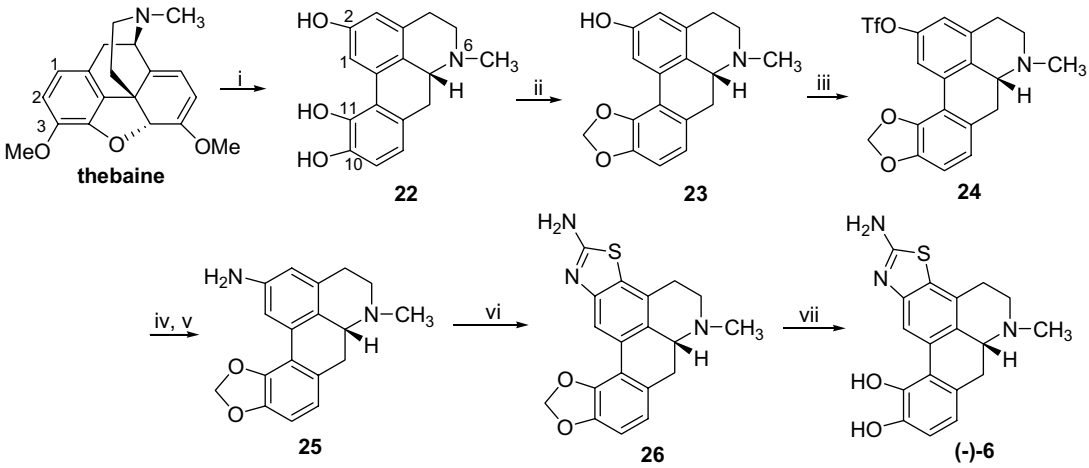
In addition, compound **14** was reacted with BBr_3 (1 M) to give *O*-demethylated 2,11-dihydroxyaporphine **19** in 89% yield. Compound **19** was then converted to diamine **21** in 38% yield (3 steps) by triflating both hydroxy groups to yield bistriflate **20** followed by a C–N coupling reaction using a similar procedure described above.[16–18,30,31] Treating 2,11-diaminoaporphine **21** with Br_2 and KSCN gave the much polar bisaminothiazole **(−)-5** as the sole product in 28% yield, and no other regioisomer was observed. The chemical shift of H-1 in this compound is 7.83 ppm as a singlet (300 M ^{1}H NMR). The down-field of the chemical shift of H-1 could be explained by the effect of the N atom in the C10,C11-fused thiazole moiety. Such an effect was also observed in compounds **13**–**21** where the chemical shift of H-1 is in much down-field than that of H-3 (~1 ppm). The regioselective formation of the aminothiazole component at C2,C3 instead of C2,C1 in this reaction can be rationalized by the relatively less steric effect at C3 than that at C1 in amine **21**, similar to our observation in preparation of aminothiazole-derived opioids.[16–18]



Scheme 1. Synthesis of (±)-**3**. Reagents and conditions: (i) NaOH (10% NaOH); (ii) ethyl bromoacetate; (iii) $NaBH_4$; (iv) PPA, 100 °C; (v) Br_2, AcOH, then $(NH_2)_2C{=}S$, reflux.



Scheme 2. Synthesis of aminothiazoles (−)-**4** and (−)-**5**. Reagents and conditions: (i) L-selectride (1 M), rt; (ii) Tf_2O, Py, THF; (iii) $MeSO_3H$, 90 °C; (iv) Pd/C, Mg, MeOH; (v) C_4H_9COOH, DCC, DMAP; (vi) Tf_2O, Py, DCM; (vii) $Ph_2C{=}NH$, $Pd(PPh_3)_4$, BINAP, DMF; (viii) NH_2OH, NaOAc; (ix) Br_2, AcOH, then KSCN; (x) BBr_3 (1 M), CH_2Cl_2.



Scheme 3. Synthesis of aminothiazoles (−)-**6**. Reagents and conditions: (i) $MeSO_3H$, 90 °C, then HBr (48%), reflux; (ii) $BrCH_2Br$, THF, reflux; (iii) Tf_2O, Py; (iv) $Ph_2C{=}NH$, $Pd(PPh_3)_4$, BINAP, DMF; (v) NH_2OH, NaOAc; (vi) Br_2, HOAc, then KSCN, rt; (vii) BBr_3, CH_2Cl_2, −78 °C.

Similarly, starting from thebaine, aminothiazole (−)-**6** was prepared as described in Scheme 3. $MeSO_3H$-catalyzed rearrangement of thebaine followed by *O*-demethylation yielded 2,10,11-trihydroxy-aporphine **22**.[32,33] After protection of the catecholic hydroxyls, the resulting 2-hydroxy-10,11-methylene dioxoaporphine **23**[32,33] was subjected to triflation yielding triflate **24**, followed by a C–N coupling reaction to give amine **25**. The subsequent aminothiazole formation reaction was conducted using a similar procedure described above, to yield aminothiazole **26**, which was *O*-deprotected by reacting with BBr_3 at −78 °C to give the expected highly polar aminothiazoloaporphine (−)-**6** in 7% overall yield (6 steps from **22**). Again, the regioisomer containing a C2,C1-fused aminothiazole was not identified during aminothiazole formation step. This was confirmed by the significant down-field of chemical shift of H-1 which is at 8.71 ppm (300 M ^{1}H NMR) indicating a remarkable effect of the C11–OH on this proton in compound (−)-**6**. Same as that of thiazole (−)-**5**, the regiochemical selectivity of the aminothiazole formation can be rationalized by the relatively less steric effect at C3 compared to that at C1 in amine **25**.

3. Results and discussion

The synthesized aminothiazole-privileged aporphines ((±)-**3**, (−)-**4**-(−)-**6**), valeroyl ester **15**, and related intermediates (**18**, **21**) were subjected to the competitive binding assays for DA receptors (D_1, D_2) and serotonin receptor (5-HT_{1A}), respectively, using membrane preparation obtained from stable transfected HEK293 or CHO cells with individual receptor. First, the ability at 10 μM concentration to inhibit the binding of a tritiated radioligand to the corresponding receptor was tested. Compounds with binding inhibited by more than 80% were further assayed at six or more concentrations, ranging above and below IC_{50}. The K_i ± SE was then derived from the equation $K_i = IC_{50}/1 + [C/Kd]$. These procedures are similar to those reported previously[10–12,26,27,29] by us or others.

Table 1
Competitive binding assay of novel aporphines[a]

Compound	D_1 [^{3}H]SCH23390		D_2 [^{3}H]spiperone		5-HT_{1A} [^{3}H]8-OH-DPAT	
	%	K_i (nM)	%	K_i (nM)	%	K_i (nM)
(±)-**3**	−1.7	—	26.6	—	ND	ND
(−)-**4**	27.8	—	14.0	—	48.1	—
(−)-**5**	34.3	—	6.3	—	0.0	—
(−)-**6**	82.0	2520 ± 313	82.0	328 ± 110	64.4	—
14		46.0 ± 2.8[b]	—	235 ± 32[b]		
15	94.9	116 ± 13	24.0	—	100	151 ± 6
18	95.0	78 ± 20	32.7	—	94.0	640 ± 210
21	53.4	2920 ± 583	24.0	—	48.1	2300
Apomorphine	89	290 ± 60	83.5	98 ± 20	ND	ND
SCH-23390	100	0.8 ± 0.1	ND	ND	ND	ND
Spiperone	ND	ND	100	0.44 ± 0.1	ND	ND
5-HT	ND	ND	ND	ND	100	3.0 ± 1.1

[a] Dashed lines indicate that the compound has K_is of higher than 20 μM.
[b] From Ref. [10]. ND denotes that the activity was not determined.

[^{3}H]SCH23390, [^{3}H]spiperone, and [^{3}H]8-OH-DPAT were used as the standard radioligands for DA D_1, D_2, and serotonin 5-HT_{1A} receptors, respectively. APO ((−)-**1**) was also tested for comparison. Data for compound **14** were taken from Ref. 10.

The binding inhibition of the radioligands is described in Table 1. To our surprise, compound (±)-**3**, in which the catechol fragment of APO is replaced directly by the aminothiazole moiety, shows only 27% binding inhibition (K_i > 20 μM) of [^{3}H]spiperone (D_2 receptor) in the competitive binding assays, whereas an inhibition of 84% is observed from the parent compound, APO ((−)-**1**). Compound (±)-**3** also has poor inhibition ability on [^{3}H]SCH23390 binding to the D_1 receptor indicating that (±)-**3** is inactive at this receptor. Although dopaminergic property of this compound was described early in Berney and Schneider's report,[20–22] our results suggest that this compound is inactive at both D_1 and D_2 DA receptor subtypes.

Aminobenzothiazoles (−)-**4** and (−)-**5** also show poor binding affinity at both D_1 and D_2 DA receptor sites. Since a C2-methoxy and C6a *R*-configuration in aporphine derivatives are supposed to contribute to DA receptor binding of these compounds,[1–3] the poor binding of (−)-**4** and (−)-**5** at the D_2 receptor clearly demonstrates that replacement of the catechol moiety of APO by an aminothiazole functionality results in a complete loss of DA receptor binding. These two compounds also show poor binding at the serotonin 5-HT_{1A} receptor. To rule out the possible effect that the aminothiazole moiety at C2,C3 has on the loss of DA receptor binding activity of(−)-**5** (both sterically and electronically), catecholic aporphine (−)-**6** was examined in the same assays. To our surprise, aminothiazole (−)-**6** shows a good K_i binding value of 328 nM at the D_2 receptor but the affinity at the D_1 receptor is poor (2.5 μM). Therefore, (−)-**6** is slightly (3-fold) less potent than APO ((−)-**1**) at the D_2 receptor, but has improved binding selectivity (7.6-fold) than APO (3.4-fold) for the D_2 over D_1 receptors. This result indicates the importance of the catecholic function for the D_2 receptor binding in APO and its derivatives, and that a relative large C2/C3 substituent, for example, aminothiazole, is tolerated. This is an important appendage to the early observations by Neumeyer and others[2,3,32,33] that a relative by small C2-substituent in APO, such as MeO-, OH-, NH_2-, and F-, is beneficial to the DA receptor binding.

It is of note that 11-*O*-valeroyl-(**15**), and 11-amino-(**18**) aporphines show remarkable inhibition of [^{3}H]SCH23390 binding at the D_1 receptor, but not of [^{3}H]spiperone binding at the D_2 receptor. Both compounds produce a same level of binding inhibition of 95% with K_i values of 116 and 78 nM, respectively, at the D_1 receptor. The good affinity at the D_1 and poor affinity at the D_2 receptors of valeroyl ester **15** is intriguing since a similar 11-valeroyl ester without the C2-MeO substituent has been reported possessing good D_2 receptor affinity in our previous report.[29] However, a similar result was reported by Neumeyer et al.[10] recently that 2-methoxy-11-hydroxy-aporphine **14**, the precusor of compound **15**, displays good binding affinity at the D_1 receptor but moderate affinity at the D_2 receptor. Interestingly, compounds **15** and **18** also show good inhibition of [^{3}H]8-OH-DAPT binding at the serotonin 5-HT_{1A} receptor, with K_i values of 151 and 640 nM, respectively. Thus, compound **15** is equally potent at both D_1 and 5-HT_{1A} receptors, whereas compound **18** is 8-fold more potent for the D_1 receptor against the 5-HT_{1A} receptor. 2,11-Bisamino-aporphine **21** does not show appreciable affinity at either D_1 or 5-HT_{1A} receptors. Again, the loss of D_2 receptor binding activity of these three compounds can be attributed to the absence of the catecholic function.

4. Conclusions

In summary, we have designed and synthesized a series of apomorphine derivatives ((±)-**3**, (−)-**4**-(−)-**6**) with a privileged 2-aminothiazole functionality which is lent from the orally available anti-parkinsonian drug, pramipexole (**2**). Compound (±)-**3** was obtained by total synthesis in racemic form. Aminothiazoles (−)-**4**-(−)-**6**, 11-valeroyl ester **15**, and the intermediates **18**, and **21** were prepared in *R*-(−)-configuration from alkaloid thebaine. All these compounds were screened for their binding ability to dopamine D_1, D_2, and serotonin 5-HT_{1A} receptors. Among these compounds, only catecholic aporphine (−)-**6** shows a good affinity at the D_2 receptor with K_i of 328 nM, slightly less potent (3-fold), but more selective against the D_1 receptor than that of the parent compound, APO ((−)-**1**). Although possessing reduced affinity at the D_2 receptor, aporphines **15** and **18** show significant affinity at both D_1 and 5-HT_{1A} receptors. The former compound is equipotent at both receptors (K_i: 116 and 151 nM, respectively), whereas the latter is 8-fold more potent at the D_1 (K_i, 78 nM) than at the 5-HT_{1A} receptor (K_i, 640 nM). These results indicate that the catechol fragment is critical for the D_2 receptor binding of apomorphine (APO, (−)-**1**), but not necessary for binding at the D_1 and 5-HT_{1A} receptors.

5. Experimental

Chemistry. Melting points were determined on a Thomas–Hoover capillary tube apparatus and are reported uncorrected. ^{1}H and ^{13}C NMR spectra were recorded on a Brucker AC300 spectrometer using tetramethylsilane as an internal reference. Element analyses, performed by the Analytic Lab, SIMM, were within ±0.4% of theoretical values. Analytical thin-layer chromatography (TLC) was carried out on 0.2-mm Kieselgel 60F 254 silica gel plastic

sheets (EM Science, Newark). Flash chromatography was used for the routine purification of reaction products. The column output was monitored with TLC. Yields of all the reactions were not optimized.

5.1. *N*-Propyl-2,3,9,9a-tetrahydro-1H-benzo[de]quinolin-7(8H)-one (10)

This compound was prepared from phenylethylamine in 7 steps using a slightly modified procedure reported by Berney[20,21] or Dijkstra.[19] MS (EI) 229 (M^+); ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.86 (m, 1H), 7.27 (m, 2H), 3.56 (m, 1H), 3.15 (m, 2H), 2.83 (m, 3H), 2.54 (m, 4H), 1.80 (m, 1H), 1.57 (m, 2H), 0.94 (m, 3H).

5.2. *N*-Propyl-5,6,6a,7-tetrahydro-4H-benzo[de]thiazolo[4,5-*g*]quinolin-9-amine ((±)-3)

To a solution of **10** (0.215 g, 0.9 mmol) in AcOH (2 mL), 4 drops of 40% HBr solution were added dropwise, followed by Br_2 (0.162 g, 1.01 mmol). The reaction was heated to 60 °C overnight. The mixture was cooled and then evaporated in vacuo. The residue was basified (pH 8–9) with NH_4OH, and extracted with CH_2C1_2 (3× 30 mL). The combined organic layer was washed with brine, dried over anhyd Na_2SO_4, and evaporated in vacuo. The residue was suspended in 5 mL of abs EtOH, and thiourea (30 mg, 0.39 mmol) was added in one portion. The mixture was refluxed for 12 h and cooled to rt. The solution was evaporated in vacuo, basified with NH_4OH, and extracted with CH_2Cl_2 (3× 30 mL). The combined organic phase was washed with brine, dried over anhyd Na_2SO_4, and evaporated in vacuo. The residue was purified by silica gel chromatography (petroleum/EtOAc = 2:1) to give the title compound (±)-**3** (22 mg, 22%). ^{1}H NMR (300 MHz, $CDC1_3$) δ 7.51 (d, 1H, J = 7.2 Hz), 7.18 (dd, 1H, J = 7.5, 7.5 Hz), 6.99 (d, 1H, J = 7.8 Hz), 4.96 (s, 2H), 3.81 (m, 1H), 3.15 (m, 3H), 2.74 (m, 5H), 1.62 (m, 2H), 0.95 (m, 3H). ^{13}C NMR (75 MHz, $CDC1_3$-CD_3OD) δ 167.8, 143.5, 133.2, 129.9, 129.6, 126.8, 126.5, 119.9, 116.2, 59.0, 55.4, 48.8, 27.8, 25.3, 17.2, 11.0. MS (EI-LR) 285 (M^+). HRMS calcd for $C_{16}H_{19}N_3S$ (M^+) 285.1300; found 285.1278.

5.3. 3-*O*-((Trifluoromethyl)sulfonyl)oripavine (12)[24–27]

This compound was prepared using a similar procedure reported by Neumeyer et al. from oripavine **11**[24,25] in 90% yield.

5.4. (*R*)-2-Methoxy-10[(trifluoromethyl)sulfonyl]-11-hydroxyaporphine (13)[26,27]

Compound **12** (0.160 g, 0.47 mmol) was dissolved in 1 mL of 98% $MeSO_3H$ at rt. The mixture was stirred for 25 min at 95–100 °C, and then cooled to rt, and quenched with ice. The mixture was basified with NH_4OH, and extracted with CH_2Cl_2 (4× 20 mL). The organic layer was combined and washed with brine, and dried over anhydrous Na_2SO_4. The solvent was evaporated yielding the title compound **13** in quantitative yield. ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.7 (br s, 1H), 7.42 (d, 1H, J = 1.5 Hz), 7.22 (d, 1H, J = 8.7 Hz), 6.95 (dd, 1H, J = 8.4, 3.2 Hz), 6.70 (dd, 1H, J = 14.1, 2.1 Hz), 3.74 (s, 3H), 3.10 (m, 5H), 2.43 (s, 3H), 2.29 (m, 2H). MS (EI-LR) 429 (M^+).

5.5. (*R*)-2-Methoxy-11-hydroxyaporphine (14)[26,27]

To a mixture of triflate **13** (1.33 g, crude) and 10% Pd/C (0.3 g) in anhydrous MeOH (100 mL) at rt was added Mg metal (freshly polished, 1.33 g) and NH_4OAc (1.5 g). The mixture was stirred at rt for 2 days and then filtered, and the filtrate was evaporated. The residue was diluted with NH_4OH (100 mL) and extracted with CH_2Cl_2 (5 × 50 mL). The combined organic layer was washed with brine and dried over anhydrous Na_2SO_4. The solvent was evaporated. The residue was purified with silica gel column chromatography eluting with $CHCl_3$/MeOH = 20:1 (1% Et_3N) to afford the title compound **14** as yellow oil (519 mg, 41%). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.67 (d, 1H, J = 2.4 Hz), 7.06 (dd, 1H, J = 7.5, 7.8 Hz), 6.82 (d, 1H, J = 7.5 Hz), 6.74 (d, 1H, J = 8.1 Hz), 6.59 (d, 1H, J = 2.4 Hz), 3.78 (s, 3H), 3.16 (m, 4H), 2.62 (m, 6H).

5.6. (*R*)-2-Methoxyaporphin-11-yl pentanoate (15)

Phenol **14** (70 mg, 0.25 mmol), valeric acid (36 μL, 0.33 mmol), and a catalytic amount of DMAP were dissolved in 15 mL of anhydrous CH_2Cl_2 under nitrogen. To the stirred mixture, a solution of *N*,*N*-dicyclohexylcarbodiimide (DCC, 72 mg, 0.35 mmol) in 5 mL of anhydrous CH_2Cl_2 was added at rt. After stirring for 4 h, the reaction mixture was filtered and evaporated to dryness. Purification by silica gel chromatography (petroleum/EtOAc = 2:1, 1% Et_3N) yielded the title compound **15** as oil (33 mg 36%).^{1}H NMR (300 MHz, $CDCl_3$) δ 7.39 (d, 1H, J = 2.7 Hz), 7.20 (m, 2H), 6.99 (d, 1H, J = 7.8 Hz), 6.61 (d, 1H, J = 2.7 Hz) 3.80 (s, 3H), 3.14 (m, 3H), 3.03 (m, 1H), 2.60 (m, 8H), 1.68 (m, 2H), 1.40 (m, 2H), 0.93 (t, 3H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 171.9, 157.9, 147.4, 138.5, 134.3, 131.6, 127.8, 127.6, 127.1, 126.0, 122.2, 112.3, 111.9, 61.4, 55.2, 52.9, 43.8, 35.2, 34.4, 29.4, 26.7, 22.2, 13.7. MS (EI) 337 (M^+). HRMS (EI) calcd for $C_{19}H_{19}N_3OS$ 337.1249; found 337.1251.

5.7. (*R*)-2-Methoxy-11[(trifluoromethyl)sulfonyl]-aporphine (16)

This compound was prepared from phenol **14** in 89% yield using a same procedure as preparation of triflate **12**. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.32 (d, 1H, J = 2.4 Hz), 7.26 (m, 3H), 6.79 (d, 1H, J = 2.4 Hz), 3.83 (s, 3H), 3.12 (m, 5H), 2.75 (1H, dd, J = 3.3, 16.5 Hz), 2.16 (s, 3H), 2.15 (m, 1H).

5.8. (*R*)-11-Amino-2-methoxyaporphine (18)

To a solution of triflate **16** (0.102 g, 0.24 mmol) in THF (10 mL) were added $Pd(OAc)_2$ (20 mg), *rac*-2,2-bis (diphenyl- phosphino)-1,1-binaphthyl (25 mg), benzophenone imine (0.070 mL, 0.42 mmol), Cs_2CO_3 (250 mg), and 4,7,13,16,21,24-hexaoxa-1,10-diazabicyclo[8.8.8]hexacosane (20 mg) under nitrogen. The mixture was heated to 65–70 °C with stirring overnight. The solvent was removed. The residue was diluted with CH_2Cl_2, washed with brine, dried, and concentrated. The crude product was purified by column chromatography eluting with petroleum/EtOAc = 1:2 (with 1% of Et_3N), to yield imine **17** as yellow oil (72 mg, 66%).

To a solution of the intermediate **17** (72 mg, 0.16 mmol) in 10 mL of MeOH were added $NH_2OH.HCl$ (78 mg, 1.1 mmol) and anhydrous NaOAc (120 mg, 1.46 mmol). The mixture was stirred overnight at rt. The solvent was removed. The residue was diluted with 0.1 M NaOH solution, and extracted with CH_2Cl_2 (5× 20 mL). The combined organic layer was washed with brine, dried over anhydrous Na_2SO_4, and the solvent was evaporated. The residue was purified by column chromatography eluting with $CHCl_3$/MeOH = 30:1 to give 2-methoxy-11-aminoaporphine **18** as yellow solid (32 mg, 70%). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.54 (d, 1H, J = 2.4 Hz), 7.03 (t, 1H), 6.70 (t, 2H), 6.60 (d, 1H, J = 2.4 Hz), 4.08 (br s, 2H), 3.82 (s, 3H), 3.05 (m, 4H), 2.75 (m, 1H), 2.52 (m, 5H). MS (EI-LR) 280 (M^+).

5.9. (*R*)-10,11:[5,4-*m*]-2′-Aminothiazolo-2-methoxyaporphine ((−)-4)

Amine **18** (17 mg, 0.06 mmol) and KSCN (25 mg, 1.36 mmol) were mixed in a solution of AcOH (8 mL). A solution of Br_2

(65 mg, 0.41 mmol) in AcOH (0.4 mL) was added dropwise. The reaction mixture was stirred overnight at rt and then the solvent was evaporated. The residue was diluted with 15 mL of NH_4OH (8%), and extracted with CH_2Cl_2 (4× 20 mL). The combined organic layer was washed with brine, dried over anhydrous Na_2SO_4, and then evaporated. The residue was subjected to chromatography ($CHCl_3$/MeOH) to yield the title compound (−)-**4** as yellow solid (15 mg, 91%). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.43 (d, 1H, J = 1.8 Hz), 7.36 (d, 1H, J = 8.7 Hz), 6.70 (d, 1H, J = 8.7 Hz), 6.64 (d, 1H, J = 1.2 Hz), 4.40 (br s, 2H), 3.81 (s, 3H), 3.70 (m, 1H), 3.10 (m, 3H), 2.73 (m, 1H), 2.55 (m, 4H), 2.38 (m, 1H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 157.7, 145.7, 141.3, 134.7, 134.3, 132.4, 127.4, 121.5, 116.1, 111.5, 111.3, 109.6, 109.2, 60.9, 55.0, 52.5, 43.7, 33.1, 29.4. MS (EI) 337 (M^+). HRMS: calcd for $C_{19}H_{19}N_3OS$ (M^+) 337.1249; found 337.1251.

5.10. (*R*)-2,11-Dihydroxyaporphine (19)

To a solution of 2-methoxy-11-hydroxy-aporphine **14** (127 mg) in 10 mL of CH_2Cl_2, cooled to −78 °C was added dropwise a solution of BBr_3 (1 M in CH_2Cl_2, 10 mL). The mixture was stirred at −78 °C for 2 h, then at rt overnight. After cooling to −78 °C again, a solution of MeOH (10 mL) was added dropwise. The mixture was stirred at rt for 2 h, and then evaporated. The residue was dissolved in MeOH, evaporated again. After repeating this procedure twice, the title compound **19** was obtained as a pale solid (140 mg, 89%). This compound was used for the next step without further purification.

5.11. (*R*)-2,11-Di[(trifluoromethyl)sulfonyl]aporphine (20)

To a solution of **19** (53 mg, 0.15 mmol) and pyridine (0.42 mmol) in CH_2Cl_2 (10 mL), cooled to −30 °C to 40 °C, Tf_2O (110 mL, 0.66 mmol) was added dropwise. The reaction mixture was allowed to reach rt and stirred for 2 h. The reaction was quenched with cold water. The organic layer was separated, washed with brine and dried (Na_2SO_4). After removal of the solvent, the residue was subjected to column chromatography (petroleum/EtOAc = 5:1, with 1% Et_3N) to yield bistriflate **20** as light yellow oil (80 mg, 98%).^{1}H NMR (300 MHz, $CDCl_3$) δ 7.67(d, 1H, J = 2.4 Hz), 7.35 (m, 1H), 7.09 (d, 1H, J = 2.4 Hz), 6.74 (d, 1H, J = 8.1 Hz), 6.59 (d, 1H, J = 2.4 Hz), 3.78 (s, 3H), 3.16 (m, 4H), 2.81(dd, 1H, J = 3.0, 16.5 Hz), 2.56 (m, 5H). MS(EI-LR): 531(M^+).

5.12. (*R*)-2,11-Diaminoaporphine (21)

To a solution of triflate **20** (71 mg, 0.13 mmol) in THF (10 mL) were added $Pd(OAc)_2$ (20 mg), *rac*-2,2-bis (diphenylphosphino)-1,1-binaphthyl (BINAP, 25 mg), benzophenone imine (0.100 mL, 0.60 mmol), Cs_2CO_3 (250 mg), and 4,7,13,16,21,24-hexaoxa-1,10-diazabicyclo[8.8.8]hexacosane (20 mg) under nitrogen. The mixture was heated to 65–70 °C with stirring overnight, and then the solvent was removed. The residue was diluted with CH_2Cl_2, washed with brine, dried, and concentrated. The crude product was purified by column chromatography (petroleum/EtOAc = 1:1, with 1% Et_3N) to yield the imine intermediate as a yellow solid (45 mg, 61%).

To a solution of the imine intermediate (45 mg, 0.075 mmol) in MeOH (10 mL) were added NH_2OH·HCl (78 mg, 1.1 mmol) and NaOAc (120 mg, 1.46 mmol). The reaction mixture was stirred overnight at rt, and the solvent was removed. The residue was diluted with 0.1 N NaOH solution, and extracted with CH_2Cl_2 (5× 20 mL). The combined organic layer was washed with brine, dried over anhydrous Na_2SO_4, and then evaporated. The residue was purified by column chromatography ($CHCl_3$/MeOH = 20:1 with 1% Et_3N), to yield diaminoaporphine **21** as a yellow solid (13 mg, 65%). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.31 (dd, 1H, J = 2.4 Hz), 7.01 (m, 1H), 6.69 (m, 2H), 6.41 (d, 1H, J = 2.1 Hz), 4.05 (br s, 2H), 3.60 (br s, 2H), 3.10 (m, 4H), 2.69 (m, 1H), 2.53 (m, 5H). MS (EI-LR) 265 (M^+).

5.13. (*R*)-2,3:[4,5-*b*]-10,11:[5,4-*m*]-Bis(2′-aminothiazolo)-aporphine ((−)-5)

Diamine **21** (42 mg, 0.16 mmol) and KSCN (92 mg, 0.90 mmol) were mixed in a solution of AcOH (5 mL). A solution of Br_2 (43 mg, 0.28 mmol) in AcOH (4.2 mL) was added dropwise. The resulting mixture was stirred overnight at rt and then evaporated. The residue was diluted with 28 mL of NH_4OH, and extracted with CH_2Cl_2 (5× 20 mL). The combined organic layer was washed with brine, dried over anhydrous Na_2SO_4, and then evaporated. The residue was purified by column chromatography ($CHCl_3$/CH_3OH = 25:1 with 1% Et_3N) to yield bisaminothiazole (−)-**5** as a white solid (17 mg, 28%). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.02 (s, 1H), 7.36 (d, 1H, J = 8.7 Hz), 6.69 (d, 1H, J = 8.4 Hz), 5.37 (br s, 2H), 4.47 (s, 2H), 3.69 (q, 1H), 3.10 (m, 3H), 2.60 (m, 5H), 2.38 (t, 1H); ^{13}C NMR (75 MHz, $CDC1_3$-CD_3OD) δ 167.8, 149.5, 146.7, 140.2, 134.3, 129.6, 128.6, 128.0, 125.7, 121.2, 116.2, 112.7, 111.7, 107.6, 61.3, 51.8, 43.0, 32.6, 27.8. MS (EI) 379 (M+). HRMS calcd for $C_{19}H_{17}N_5S_2$ (M^+) 379.0925; found 379.0920.

5.14. (*R*)-2-Hydroxy-10,11-(methylenedioxy)apomorphine (23)

Finely ground NaOH (0.452 g, 11.3 mmol) was added to a solution of **22**[32,33] (1.23 g, 3.2 mmol) in 30 mL of dry DMSO at rt under N_2 and stirred for 1 h. Methylene dibromide (0.290 mL, 4.2 mmol) was added, and the mixture was heated at 80 °C overnight. After cooling, the solution was diluted with ice water and extracted with EtOAc (5× 100 mL). The organic phase was combined, washed with brine, dried over anhydrous Na_2SO_4, and evaporated in vacuo. The residue was purified by column chromatography ($CHCl_3$/MeOH = 20:1) to yield the title compound **23**[32,33] (0.344 g, 36%). ^{1}H NMR (300 MHz, $CDC1_3$) δ 7.30 (d, 1H, J = 2.4 Hz), 6.67 (d, 1H, J = 8.1 Hz), 6.61 (d, 1H, J = 7.8 Hz), 6.38 (d, 1H, J = 2.4 Hz), 5.84 (d, 1H, J = 1.5 Hz), 5.79 (d, 1H, J = 1.2 Hz), 3.12 (m, 4H), 2.69 (t, 1H), 2.54 (m, 5H).

5.15. (*R*)-2-(Trifluromethyl)sulfonyl-10,11-(methylenedioxy)apomorphine (24)

A solution of **23** (181 mg, 0.61 mmol) and Et_3N (0.15 mL, 1.06 mmol) in 20 mL of anhydrous CH_2Cl_2 was cooled to −30 °C. Tf_2O (0.125 mL, 0.75 mmol) in 0.5 mL of anhydrous CH_2Cl_2 was added dropwise. The reaction mixture was allowed to reach rt and stirred for 2 h. The reaction was quenched with cold water and the organic layer was separated, washed with brine, dried over anhydrous Na_2SO_4 and evaporated in vacuo. Purification by silica gel chromatography (petroleum/EtOAc = 2:1, 1% Et_3N) afforded the title compound **24** (0.247 g, 95%). ^{1}H NMR (300 MHz, $CDC1_3$) δ 7.85 (d, 1H, J = 2.4 Hz), 6.97 (d, 1H, J = 2.4 Hz), 6.76 (s, 2H), 6.15 (d, 1H, J = 1.2 Hz), 6.01 (d, 1H, J = 1.2 Hz), 3.14 (m, 4H), 2.77 (dd, 1H, J = 16.5, 2.7 Hz), 2.58 (m, 5H).

5.16. (*R*)-2-Amino-10,11-(methylenedioxy)apomorphine (25)

This compound was prepared from **24** by using a similar procedure as that of preparation of **18** in 74% yield ($CHCl_3$/MeOH = 20:1, 1% Et_3N). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.33 (d, J = 2.4 Hz, 1H), 6.73 (d, J = 7.8 Hz, 1H), 6.69 (d, J = 7.8 Hz, 1H), 6.41 (d, 1H, J = 2.1 Hz), 6.08 (d, J = 1.5 Hz, 1H), 5.95 (d, J = 1.5 Hz, 1H), 3.50 (br s, 2H), 3.10 (m, 4H), 2.50 (m, 6H).

5.17. (*R*)-2,3:[4,5-*b*]-2′-Aminothiazolo-10,11-(methylenedioxy)aporphine (26)

This compound was prepared from amine **25** in 91% yield by using a similar procedure as that of preparation of (−)-**4**. ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.19 (s, 1H), 6.75 (d, 1H, J = 7.8 Hz), 6.68 (d, 1H, J = 8.1 Hz), 6.09 (d, 1H, J = 0.9 Hz), 5.97 (d, 1H, J = 1.2 Hz), 5.41 (br s, 2H), 3.18 (m, 4H), 2.60 (m, 6H).

5.18. (*R*)-2,3:[4,5-*b*]-2′-Aminothiazolo-10,11-dihydroxyaporphine ((−)-6)

To a solution of **26** (100 mg, 0.29 mmol) in 10 mL of anhydrous CH_2Cl_2, cooled to −78 °C, was added dropwise a solution of BBr_3 (1 M in CH_2Cl_2, 5 mL). The mixture was stirred at −78 °C for 2 h, then at rt overnight. After cooling to −78 °C again, 10 mL MeOH was added dropwise. The mixture was stirred at rt for 2 h, and evaporated. The residue was redissolved in MeOH, and evaporated again. After repeating this procedure twice, the residue was recrystallized from anhydrous MeOH to give the title compound (−)-**6** as pale yellow solid (45.2 mg, 32%). MS (EI) 339 (M^+). ^{1}H NMR (300 MHz, CD_3OD-d_4) δ 8.71 (s, 1H), 6.82 (d, 1H, J = 8.1 Hz), 6.77 (d, 1H, J = 7.8 Hz), 3.18 (m, 4H), 2.60 (m, 6H). Anal. Calcd for $C_{18}H_{17}N_3O_2S$. 2.5HBr: C, 39.91; H, 3.63; N, 15.48; found: C, 39.79; H, 4.03; N, 15.59.

5.19. Established stable expression of cell lines

The rat 5-HT_{1A} receptor gene, human D_1 receptor gene, and human D_2 receptor gene were individually cloned into pcDNA3.0 vector. The 5-HT_{1A} construct was then transfected into CHO cells. The D_1 and D_2 receptors were transfected to HEK293 cells, respectively. G418 at 800 μg/ml was used for selection. Monoclonal transfected cells were isolated and maintained in medium containing Ham's F12 nutrient mixture (Gibco), 10% fetal bovine serum, 100 U/ml penicillin, 100 U/ml streptomycin, and 200 μg/ml G418 at 37 °C and 5% CO_2.

To confirm the success of transfection, the saturation binding experiment that the expression of 5-HT_{1A} receptor in the CHO cell line is 1.5531 ± 0.2803 nmol/g protein with a K_d value of 1.2058 nM, expression for D_1 is 10.67 nmol/g protein with a K_d value of 1.31 ± 0.16 nM. The K_d for D_2 is 0.06 nM.

5.20. Radioligand binding assays

The affinity of *compounds* to the D_1 and D_2 dopamine receptors, and the 5-HT_{1A} receptor was determined by competition binding assays. Membrane homogenates of 5-HT_{1A}-CHO cells, D_1- or D_2-HEK293 cells were prepared as described previously.[26,29] Duplicated tubes were incubated at 30 °C for 50 min with increasing concentrations of respective compound and with 0.7 nM [^{3}H]8-OH-DPAT (for 5-HT_{1A} receptor), [^{3}H]SCH23390 (for D_1 dopamine receptors), or [^{3}H]spiperone (for dopamine D_2 receptor) in a final volume of 200 μL binding buffer containing 50 mM Tris, 4 mM $MgCl_2$, pH 7.4. Nonspecific binding was determined by parallel incubations with either 10 μM WAY100635 for 5-HT_{1A}, SCH23390 for D_1, or spiperone for D_2 dopamine receptors, respectively. The reaction was started by addition of membranes (15 ng/tube), and stopped by rapid filtration through Whatman GF/B glass fiber filter and subsequent washing with cold buffer (50 mM Tris, 5 mM EDTA, pH 7.4) using a Brandel 24-well cell harvester. Scintillation cocktail was added and the radioactivity was determined in a MicroBeta liquid scintillation counter. The IC_{50} and K_i values were calculated by nonlinear regression (PRISM, Graphpad, San Diego, CA) using a sigmoidal function.

Acknowledgments

This work was financially supported by grants from Chinese National Science Foundation (30672517, to A.Z.), Shanghai Commission of Science and Technology (07pj14104, to A.Z. and X.Z.), and grant from Ministry of Science and Technology (2007AA022163, to X.Z.). Support from Chinese Academy of Sciences and Shanghai Institute of Materia Medica was also appreciated. We also thank Professors John L. Neumeyer and Ross J. Baldessarini for their instructive discussion during this work and reevaluation of some of the synthetic compounds. Thebaine and morphine alkaloid were kindly provided by the neuropharmacological laboratory, SIMM.

Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bmc.2008.05.077.

References and notes

1. Neumeyer, J. L. In *The Chemistry and Biology of Isoquinoline Alkaloids*; Phillipson, J. D., Roberts, M. F., Zenk, M. H., Eds.; Springer-Verlag: Berlin, Germany, 1985; pp 146–170.
2. Zhang, A.; Zhang, Y.; Branfman, A. R.; Baldessarini, R. J.; Neumeyer, J. L. *J. Med. Chem.* **2007**, *50*, 171–181.
3. Zhang, A.; Neumeyer, J. L.; Baldessarini, R. J. *Chem. Rev.* **2007**, *107*, 274–302.
4. Mathiessen, A.; Wright, C. R. A. *Proc. R. Soc., Ser. B.* **1869**, *17*, 455–460.
5. Mathiessen, A.; Wright, C. R. A. *Proc. R. Soc., Ser. B.* **1869**, *17*, 460–462.
6. Neumeyer, J. L.; Neustadt, B. R.; Weinhardt, K. K.; Aporphines, V. *J. Pharm. Sci.* **1970**, *59*, 1850–1852.
7. Neumeyer, J. L.; Baldessarini, R. J.; Booth, R. G., 6th ed.. In *Burger's Medicinal Chemistry and Drug Discovery*; Abraham, D. J., Ed.; John Wiley & Sons: New York, 2003; vol. 6, pp 711–714. Chapter 12.
8. Kolls, B. J.; Stacy, M. *Clin. Neuropharmacol.* **2006**, *29*, 292–301.
9. Subramony, J. A. *Mol. Pharm.* **2006**, *3*, 380–385.
10. Si, Y.; Gardner, M. P.; Tarazi, F. I.; Baldessarini, R. J.; Neumeyer, J. L. *J. Med. Chem.* **2008**, *51*, 983–987.
11. Zhang, A.; Csutoras, C.; Zong, R.; Neumeyer, J. L. *Org. Lett.* **2005**, *7*, 3239–3242.
12. Garrido, J. M.; Delerue-Matos, C.; Borges, M. F. M.; Macedo, T. R. A.; Oliveira-Brett, A. M. *Bioelectrochemistry* **2002**, *55*, 113–114.
13. Vernier, V. G., 5th ed.. In *Burger's Medicinal Chemistry and Drug Discovery*; Wolff, M. E., Ed.; John Wiley & Sons: New York, 1996; vol. 3, pp 43–98. Chapter 37.
14. Moller, J. C.; Oertel, W. H. *Expert. Rev. Neurother.* **2005**, *5*, 581–586.
15. Reichmann, H.; Brecht, M. H.; Koster, J.; Kraus, P. H.; Lemke, M. R. *CNS Drugs* **2003**, *17*, 965–973.
16. Zhang, A.; van Vliet, L. A.; Neumeyer, J. L. *Tetrahedron Lett.* **2003**, *44*, 6459–6462.
17. Zhang, A.; Xiong, W.; Hilbert, J. E.; DeVita, E. K.; Bidlack, J. M.; Neumeyer, J. L. *J. Med. Chem.* **2004**, *47*, 1886–1888.
18. van Vliet, L. A.; Rodenhuis, N.; Wikstrom, H. *J. Med. Chem.* **2000**, *43*, 3549–3557.
19. Liu, D.; Venhuis, B. J.; Wikstrom, H. V.; Dijkstra, D. *Tetrahedron* **2007**, *63*, 7264–7270.
20. Berney, D.; Schuh, K. *Helv. Chim. Acta* **1982**, *65*, 1305–1308; Schneider, C. S.; Mierau, J. *J. Med. Chem.* **1987**, *30*, 494–498.
21. Schneider, F.; Gerold, M.; Bernauer, K. *Helv. Chim. Acta* **1973**, *56*, 759–773.
22. Schneider, C.; Merz, H.; Sobotta, R.; Bauer, R.; Mierau, J.; Schingnitz, G. Ger. Offen. DE 3624607 A1, 1988, 13pp.
23. Zhang, A.; Li, F.; Ding, C.; Yao, Q.; Knapp, B. I.; Bidlack, J. M.; Neumeyer, J. L. *J. Med. Chem.* **2007**, *50*, 2747–2751.
24. Coop, A.; Janetka, J. W.; Lewis, J. W.; Rice, K. C. *J. Org. Chem.* **1998**, *63*, 4392–4396.
25. Coop, A.; Lewis, J. W.; Rice, K. C. . *J.Org. Chem.* **1996**, *61*, 6774.
26. Si, Y.; Gardner, M. P.; Tarazi, F.; Baldessarini, R. J.; Neumeyer, J. L. . *Bioorg. Med. Chem. Lett.* **2007**, *17*, 4128–4130.
27. Si, Y.; Neumeyer, J. L. *Synthesis* **2007**, *24*, 3787–3790.
28. Sajiki, H.; Mori, A.; Mizusaki, T.; Ikawa, T.; Maegawa, T.; Hirota, K. *Org. Lett.* **2006**, *8*, 987–990.
29. Csutoras, C.; Zhang, A.; Zhang, K.; Kula, N. S.; Baldessarini, R. J.; Neumeyer, J. L. *Bioorg. Med. Chem.* **2004**, *12*, 3553–3559.
30. Hedberg, M. H.; Jansen, J. M.; Nordvall, G.; Hjorth, S.; Unelius, L.; Johansson, A. M. *J. Med. Chem.* **1996**, *39*, 3491–3502.
31. Hedberg, M. H.; Linnanen, T.; Jansen, J. M.; Nordvall, G.; Hjorth, S.; Unelius, L.; Johansson, A. M. *J. Med. Chem.* **1996**, *39*, 3503–3513.
32. Gao, Y.; Baldessarini, R. J.; Kula, N. S.; Neumeyer, J. L. *J. Med. Chem.* **1990**, *33*, 1800–1805.
33. Neumeyer, J. L.; Gao, Y.; Kula, N. S.; Baldessarini, R. J. *J. Med. Chem.* **1990**, *33*, 3122–3124.