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(±)-Nantenine analogs as antagonists at human 5-HT_{2A} receptors: C1 and flexible congeners

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Abstract

C1 and flexible analogs of (±)-nantenine were synthesized and evaluated for antagonist activity at human 5-HT_{2A} receptors in a calcium mobilization assay. This work has resulted in the identification of the most potent 5-HT_{2A} antagonist known based on an aporphine. Our results also suggest that the C1 position may be a key site for increasing 5-HT_{2A} antagonist activity in this compound series. In addition, the structural rigidity of the aporphine core appears to be required for nantenine to function as a 5-HT_{2A} antagonist.

The monoamine neurotransmitter, serotonin (5-hydroxytryptamine, 5-HT, **1**) is known to play a significant role in the central nervous system (CNS) modulation of appetite, mood, body temperature and sleep in humans.¹ There are fourteen serotonin receptors presently known, of which all except one (5-HT₃) are G-protein coupled receptors.² Ligands for the 5-HT_{2A} receptor are constantly being developed as chemical tools to study the functional role of this receptor in hallucinations, depression, anxiety and psychosis.^{3–7} The role of 5-HT_{2A} receptor blockade in the neuropharmacological processes of addiction is also of growing interest.^{8–13}

Aporphines are a diverse group of tetracyclic alkaloids found in several plant species and have been found to show a range of interesting biological activities such as antiplasmodial, antihelminthic and antitumor activities.^{14–18} As a result of their biological activities, new and facile synthetic methodologies to prepare these compounds are always being explored.^{19, 20}

Others have reported the 5-HT_{2A} antagonist properties of the aporphine alkaloid nantenine (**2**).^{21, 22} This pharmacodynamic property seems to be relevant to its in vivo activity as an antagonist of the designer drug MDMA (methylenedioxymethamphetamine, “Ecstasy”).²³ Although aporphines have been evaluated as 5-HT_{1A}, α -adrenergic, and dopaminergic D1 and D2 ligands, very little SAR work has been performed on aporphines as 5-HT_{2A} antagonists.^{21, 24–28} Part of our program is geared towards understanding the importance of selective receptor blockade as well as multi-potent antagonism involving 5-HT_{2A} receptors in the antagonism of MDMA-induced effects. Aporphines may be a valuable structural template for such a study, given the apparent promiscuity of these compounds across various CNS targets including 5-HT subtypes. As such, we have embarked on a study to evaluate the 5-HT_{2A}

antagonist activity of aporphines using **2** as a lead molecule. In this communication, we report results on the synthesis and evaluation of C1 and less rigid nantenine analogs.

Preparation of C1 analogs commenced with readily available²⁹ 4-benzyloxy-3-methoxy phenethylamine (**3**) which was condensed with bromoacid (**4**) under standard peptide coupling conditions (Scheme 1). The amide (**5**) thus produced was cyclized under Bischler-Napieralski³⁰ conditions to afford imine **6**, which was immediately reduced to give **7** without purification (due to apparent instability of the imine).

Following protection of amine **7** as the ethyl carbamate, we were now in a position to attempt the direct biaryl cyclization³¹ of **8** to give the aporphine core - a key step in this synthetic route. Following similar methodology to that previously described³¹ with some optimization of reaction conditions, we obtained **9** in 76% yield. Hydrogenolysis of the C1 benzyl group gave key intermediate **10** which served as a precursor for the C1 alkyl intermediates **11a – 11f**.

Our plan at this juncture was to reduce the carbamate functionalities of **11a – 11f** with LAH to provide the C1 target analogs **12a – 12e** and nantenine (**2**).

However, when LAH reduction was attempted, cleavage of the carbamate group occurred, giving the corresponding secondary amines as major products. This result was surprising and is in need of further investigation. Nevertheless, subsequent reductive amination of the derived secondary amines with formaldehyde afforded the target analogs **12a – 12e** and **2**. The C1 benzyl derivative **13** was accessed via LAH reduction of **9**.

To begin to evaluate the role of molecular rigidity on the 5-HT_{2A} activity of nantenine, we prepared the benzyltetrahydroisoquinoline **16** and tertiary amine **18** (Scheme 2). Thus, following similar procedures as in Scheme 1, the readily available amide **14** was cyclized under Bischler-Napieralski conditions. Subsequently, the imine prepared was reduced and then subjected to reductive amination conditions providing the *seco*-ring C derivative **16**. Borane reduction of amide **14** followed by *N*-alkylation gave compound **18**. All compounds were characterized with routine spectroscopic techniques including ¹H NMR, ¹³C NMR and HRMS.

Compounds **2**, **12a–12e**, **13**, **16** and **18** were evaluated for functional activity at human 5-HT_{2A} receptors using a calcium mobilization assay.³² Results are presented in Table 1. The less rigid analogs, ie compounds **16** and **18** had significantly reduced antagonist activities as compared to nantenine, suggesting that the structural rigidity of the aporphine nucleus is required for 5-HT_{2A} activity. Increasing the C1 alkyl chain length by one carbon (compound **12a**) had little effect on 5-HT_{2A} antagonist activity. However, incremental additions of 2, 3 and 4 carbons gave a progressive increase in 5-HT_{2A} antagonist activity (**12b**, **12c**, **12d**). Replacement of the C1 methyl group of nantenine with a methylenecyclopropyl moiety (compound **12e**) resulted in a twelve-fold enhancement in activity as compared to nantenine. Interestingly, the C1 benzyl analog was found to be a negative allosteric modulator (IC₅₀ = 4600 nM). In keeping with attributes of successful CNS agents,³³ the ClogP values of the C1 analogs represent reasonable starting points for simultaneous optimization of pharmacodynamic properties and blood-brain barrier penetrability in this compound series.

In conclusion, our biological results indicate that increasing the length of the C1 alkyl chain beyond two carbon atoms, results in an increase in 5-HT_{2A} antagonist activity in this series of aporphines. The C1 site may thus be a key position for further structural modifications to increase 5-HT_{2A} antagonist activity. Compound **12e**³⁴ was the most active compound identified and showed a 12-fold increase in activity as compared to nantenine. This compound is the most potent 5-HT_{2A} antagonist known with an aporphine skeleton. Our work has also identified a low activity negative allosteric modulator, **13**. We are continuing to explore the

SAR of other nantenine-derived aporphines at human 5-HT_{2A} receptors and will report our findings in due course.

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References and Notes

- Nichols DE, Nichols CD. Chem Rev 2008;108:1614. [PubMed: 18476671]
- Costall B, Naylor RJ. Curr Drug Targets 2004;3:27.
- Schmidt CJ, Sorensen SM, Kehne JH, Carr AA, Palfreyman MG. Life Sci 1995;56:2209. [PubMed: 7791509]
- Olivier B, Mos J, van Oorschot R, Hen R. Pharmacopsychiatry 1995;282(Suppl):80. [PubMed: 8614705]
- Pae CU, Serretti A, Patkar AA, Masand PS. CNS Drugs 2008;22:367. [PubMed: 18399707]
- Moller HJ. Expert Opin Pharmacother 2005;6:803. [PubMed: 15934906]
- Graeff FG, Guimaraes FS, De Andrade TG, Deakin JF. Pharmacol., Biochem Behav 1996;54:129. [PubMed: 8728550]
- Auclair A, Drouin C, Cotecchia S, Glowinski J, Tassin JP. Eur J Neurosci 2004;20:3073. [PubMed: 15579162]
- Herin DV, Liu S, Ullrich T, Rice KC, Cunningham KA. Psychopharmacology 2005;178:505. [PubMed: 15765263]
- Szucs RP, Frankel PS, McMahon LR, Cunningham KA. Behav Neurosci 2005;119:1173. [PubMed: 16300424]
- Zaniewska M, McCreary AC, Przegalinski E, Filip M. Eur J Pharmacol 2007;571:156. [PubMed: 17617403]
- Lanteri C, Salomon L, Torrens Y, Glowinski J, Tassin JP. Neuropsychopharmacology 2008;33:1724. [PubMed: 17805311]
- Levin ED, Slade S, Johnson M, Petro A, Horton K, Williams P, Rezvani AH, Rose JE. Eur J Pharmacol 2008;600:93. [PubMed: 18950618]
- Rasoanaivo P, Ratsimamanga-Urverg S, Rafatro H, Ramanitrahambola D, Palazzino G, Galeffi C, Nicoletti M. Planta Med 1998;64:58. [PubMed: 17253219]
- Ayers S, Zink DL, Mohn K, Powell JS, Brown CM, Murphy T, Brand R, Pretorius S, Stevenson D, Thompson D, Singh SB. Planta Med 2007;73:296. [PubMed: 17309014]
- Abdalla S, al-Khalil S, Afifi F. Gen Pharmacol 1991;22:253. [PubMed: 1905248]
- Stevigny C, Bailly C, Quetin-Leclercq J. Curr Med Chem Anticancer Agents 2005;5:173. [PubMed: 15777224]
- Huang RL, Chen CC, Huang YL, Ou JC, Hu CP, Chen CF, Chang C. Planta Med 1998;64:212. [PubMed: 9581516]
- Hamamoto H, Shiozaki Y, Nambu H, Hata K, Tohma H, Kita Y. Chem Eur J 2004;10:4977.
- Orito K, Uchiito S, Satoh Y, Tatsuzawa T, Harada R, Tokuda M. Org Lett 2000;2:307. [PubMed: 10814309]
- Indra B, Matsunaga K, Hoshino O, Suzuki M, Ogasawara H, Ishiguro M, Ohizumi Y. Can J Physiol Pharmacol 2002;80:198. [PubMed: 11991230]
- Tsuchida H, Ohizumi Y. Eur J Pharmacol 2003;477:53. [PubMed: 14512098]
- Fantegrossi WE, Kiessel CL, Leach PT, Van Martin C, Karabenick RL, Chen X, Ohizumi Y, Ullrich T, Rice KC, Woods JH. Psychopharmacology 2004;173:270. [PubMed: 14740148]
- Zhang A, Zhang Y, Branfman AR, Baldessarini RJ, Neumeyer JL. J Med Chem 2007;50:171. [PubMed: 17228858]

25. Si YG, Gardner MP, Tarazi FI, Baldessarini RJ, Neumeyer JL. *Bioorg Med Chem Lett* 2007;17:4128. [PubMed: 17543523]
26. Cannon JG, Flaherty PT, Ozkutlu U, Long JP. *J Med Chem* 1995;38:1841. [PubMed: 7783115]
27. Ivorra MD, Valiente M, Martinez S, Madrero Y, Noguera MA, Cassels BK, Sobarzo EM, D'Ocon P. *Planta Med* 2005;71:897. [PubMed: 16254819]
28. Westkaemper RB, Yousif M, Teitler M, Glennon RA. *Med Chem Res* 1992;2:482.
29. Batra S, Sabnis YA, Rosenthal PJ, Avery MA. *Bioorg Med Chem* 2003;11:2293. [PubMed: 12713840]
30. Wang YC, Georghiou PE. *Org Lett* 2002;4:2675. [PubMed: 12153207]
31. Lafrance M, Blaquiére N, Fagnou K. *Chem Commun* 2004:2874.
32. *Calcium Mobilization Human 5-HT_{2A} Receptor Functional Assays*. Stably expressed human 5-HT_{2A} receptor in CHO-K1 cells (ATCC), were used for the calcium mobilization functional assay. The calcium 4 dye assays (Molecular Devices, Sunnyvale, CA) were run according to manufacturer's specifications. Briefly, wells of black clear-bottom 96-well tissue culture-treated plates were seeded with 20,000 cells the afternoon before assay. On the day of assay, the cells were incubated with the calcium indicator dye for 1 hr @ 37 °C. The test compounds were preincubated with the cells during the last 15 min of the dye incubation. The plate was then placed into a FlexStation pre-warmed to 37 °C. Basal or unstimulated fluorescence intensity was recorded for 13 sec followed by the addition of 5-HT (antagonist and K_e assays). Fluorescence intensity was recorded for an additional 47 s. The effect of test compound was determined by subtracting the minimum from the maximum fluorescence recorded for each well during the 47 s recording period. Thus, each well served as its own control. All samples were run in duplicate.
33. Pajouhesh H, Lenz GR. *Neuro Rx* 2005;2:541. [PubMed: 16489364]
34. *Compound 12e*. ¹H NMR (CDCl₃, 500 MHz): δ 0.12 (m, 2H), 0.48 (m, 2H), 1.15 (m, 1H), 2.50–2.54 (obscured, 2H), 2.53 (s, 3H), 2.67 (dd, J = 3.2, 16.3 Hz, 1H), 2.97 (m, 2H), 3.03 (dd, J = 5.7, 11.4 Hz, 1H), 3.13 (m, 1H), 3.40 (dd, J = 7.6, 10.0 Hz, 1H), 3.71 (dd, J = 7.0, 10.0 Hz, 1H), 3.86 (s, 3H), 5.96 (d, J = 1.1 Hz, 1H), 5.98 (d, J = 1.1 Hz, 1H), 6.57 (s, 1H), 6.74 (s, 1H), 8.07 (s, 1H); ¹³C NMR (CDCl₃, 125 MHz): δ 3.1, 3.4, 11.0, 29.1, 35.1, 44.0, 53.2, 55.8, 62.5, 77.8, 100.8, 108.2, 109.5, 110.4, 125.9, 127.1, 127.5, 128.5, 130.6, 143.3, 146.2, 146.3, 152.1; HRESIMS Calcd for C₂₃H₂₅NO₄: 379.1784. Found: 379.1783.

Figure 1.
Structures of serotonin (1) and Nantenine (2)

Scheme 1.

Reagents and conditions: (a) GDI, THF, rt, 20 h, 90% (b) PCl_5 , DCM, 0 °C - rt, 96% (c) NaBH_4 , MeOH, 0 °C, 4h, 99% (d) ethyl chloroformate, K_2CO_3 , DCM, rt, 12h, 76% (e) $\text{Pd}(\text{OAc})_2$, ligand A, K_2CO_3 , $(\text{CH}_3)_3\text{CCOOH}$, DMA, 130 °C, 17h, 76% (f) $\text{H}_2/\text{Pd-C}$, MeOH-THF, rt, 38h, 75% (g) alkyl bromide, K_2CO_3 , acetone, reflux 12 h (h) LiAlH_4 , THF, 0 °C, 5h, 91% (i) aq. HCHO , $\text{NaBH}(\text{OAc})_3$, DCM, rt, 24h, 96% (j) LiAlH_4 , Et_2O , 0 °C, 5h, 91%

Scheme 2.

Reagents and conditions: (a) PCl_5 , DCM, 0°C - rt (b) NaBH_4 , MeOH, rt, (c) aq. HCHO, NaBH (OAc) $_3$, DCM, rt, 62% over 3 steps (d) $\text{BF}_3 \cdot \text{OEt}_2$, $\text{BH}_3 \cdot \text{THF}$, THF, 88%

Table 1Apparent affinity of nantenine analogs at human 5-HT_{2A} receptors and ClogP values³²

Compds	R	K _e (nM) ^a	CLogP ^b
2	CH ₃	850 (±5.8)	3.60
12a	–CH ₂ CH ₃	890 (±430)	4.13
12b	–(CH ₂) ₂ CH ₃	297 (±130)	4.66
12c	–(CH ₂) ₃ CH ₃	274 (±80)	5.19
12d	–(CH ₂) ₄ CH ₃	171 (±50)	5.72
12e	–CH ₂ -cyclopropyl	68 (±8)	4.58
13	–CH ₂ Ph	4600 ^c	5.37
16	-	>10,000	3.82
18	-	5180 ^d	3.69
Ketanserin	-	32 ^{c,d}	-

^aK_e (apparent affinity) values are means of at least two experiments carried out in duplicate; standard deviation is given in parentheses.^bDetermined with ChemBioDraw Ultra 11.0^cIC₅₀ determined in the presence of 5-HT EC₈₀^dExperiment run once