Pergamon

Bioorganic & Medicinal Chemistry Letters 11 (2001) 655–658

BIOORGANIC & MEDICINAL CHEMISTRY LETTERS

Influence of Chain Length and *N*-Alkylation on the Selective Serotonin Receptor Ligand 9-(Aminomethyl)-9,10-dihydroanthracene

Scott P. Runyon,[a] Jason E. Savage,[b,c] Mohamed Taroua,[a] Bryan L. Roth,[b,c] Richard A. Glennon[a] and Richard B. Westkaemper[a,*]

[a]*Department of Medicinal Chemistry, School of Pharmacy, Virginia Commonwealth University, R.B. Smith Building, 410 North 12th Street, PO Box 980540, Richmond 23298-0540, VA 23298, USA*

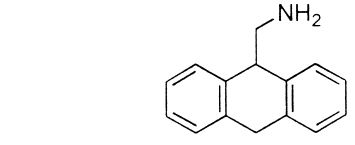
[b]*Departments of Biochemistry and Psychiatry, Case Western Reserve University School of Medicine, Cleveland, OH 44106, USA*

[c]*The NIMH Psychoactive Drug Screening Program, USA*

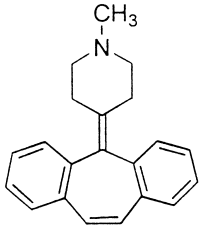
Received 17 July 2000; accepted 28 December 2000

Abstract—Comparison of the serotonin 5-HT_{2A} receptor affinities of chain lengthened and *N*-alkylated analogues of the novel ligand 9-aminomethyl-9,10-dihydroanthracene (AMDA) and a structurally similar prototypical tricyclic amine imipramine suggests that the two agents bind to the receptor in different fashions. The demonstration that AMDA is highly selective for serotonin receptors (5-HT_{2A}, $K_i = 20$ nM; 5-HT_{2C}, $K_i = 43$ nM) versus the dopamine D_2 receptor ($K_i > 10{,}000$ nM), as well as the serotonin and norepinephrine transporters ($K_i > 10{,}000$ nM) further suggests that AMDA and the nonselective ligand imipramine interact with these target macromolecules in different ways.

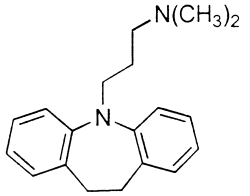
We have recently described 9-(aminomethyl)-9,10-dihydroanthracene (AMDA, **1a**) as the parent member of a potentially new class of 5-HT_{2A} receptor antagonists.[1] With the exception of its two aromatic rings and basic nitrogen, it is remarkably devoid of the pharmacophore features usually associated with selective, high affinity receptor ligands. Although AMDA was initially conceived and evaluated to test a theoretical receptor model without explicit consideration of other known 5-HT ligands,[2] it became apparent that the 9-(aminomethyl)-9,10-dihydroanthracene nucleus bears a general similarity to at least two classes of known, nonselective serotonin receptor ligands: tricyclic antidepressants and antipsychotic agents. Both classes of agents are tricyclic amines consisting of two aromatic groups flanking a non-aromatic central ring that bears an alkylamino substituent as in AMDA. If AMDA were to share a common mode of binding with either class, given the multiple neurochemical actions of classical tricyclic amines, enthusiasm for further development based on the AMDA skeleton would be significantly diminished. We have recently demonstrated that AMDA and a conformationally restricted representative of the classical tricyclic amine class, cyproheptadine (**2**), probably bind to 5-HT_{2A} receptors in different fashions.[3]



AMDA, **1a**



cyproheptadine, **2**



imipramine, **3c**

Cyproheptadine is a useful model to study potential similarities and differences between classical tricyclic amines and AMDA derivatives, principally because it is a relatively rigid structure. However, most clinically useful classical tricyclic amines are conformationally flexible. Since chain length and *N*-alkylation are two

*Corresponding author. Fax: +1-804-828-7625; e-mail: richard.westkaemper@vcu.edu

0960-894X/01/$ - see front matter
PII: S0960-894X(01)00023-3

major structural features that clearly distinguish AMDA from conformationally flexible classical tricyclic amines, we investigated the influence of chain length and *N*-alkylation in the AMDA series compared to imipramine (**3c**), a prototypical, nonselective, conformationally flexible tricyclic amine. To this end, we synthesized, examined geometric properties of, and evaluated chain lengthened and *N*-alkylated analogues of AMDA and demethylated derivatives of imipramine for comparison.

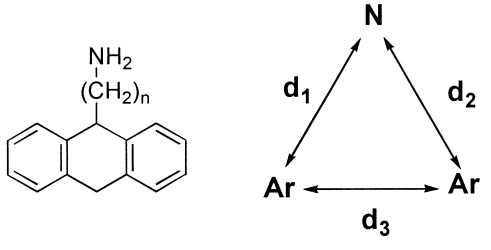
The data suggest (Table 1) that there may be two optimal chain lengths for high 5-HT_{2A} receptor affinity in the AMDA series. These are represented by an embedded phenylethyl fragment ($n=1$) (AMDA, **1a**) and a phenylbutyl chain length ($n=3$) as found in compound **6a** and the classical tricyclic antidepressants.

Several pharmacophore models for 5-HT_{2A} receptors have been proposed based on the SARs of known antagonists.[4,5] Typically, the essential geometric characteristics are described by the distances between two aromatic rings ($d_3=4.6$–7.3 Å) and the distances between each aromatic ring and the basic amine nitrogen ($d_1=5.2$–8.4 Å, $d_2=5.7$–8.5 Å). The corresponding dimensions for the minimum energy conformation of AMDA (**1a**) are similar to existing agents with respect to the aromatic rings ($d_3=4.9$ Å) but deviate substantially in that it is not symmetrical with respect to the two amine-ring distances ($d_1=3.8$ Å, $d_2=5.2$ Å), d_1 being much shorter than is considered optimal. The distance parameters for compound **6a** are quite similar to those of classical antidepressants including cyproheptadine (**2**) and imipramine (**3c**). Thus, unlike AMDA (**1a**), compound **6a** is consistent with existing pharmacophores. Crystal structures of flexible tricyclic amines like imipramine usually show an extended aminoalkyl chain[6] but folded conformers that more closely resemble AMDA are energetically accessible. Molecular dynamics simulations of imipramine suggest that folded conformers with decreased phenyl-to-N distances (range 4.0–7.5 Å) do occur.[7] Thus, classical tricyclic amines can attain an AMDA-like configuration. The series, where $n=0$–3, includes the 'phenylethylamine' skeleton of AMDA and the 'phenylbutylamine' skeleton of imipramine. Affinity is highest for the phenylethylamine and 'phenylbutylamine' skeletons. Interpreted in terms of receptor binding modes, there are two limiting possibilities. AMDA (**1a**) and **6a** might bind in a similar fashion with **6a** attaining an AMDA-like conformation. Alternatively, the aromatic moieties of AMDA and **6a** might occupy different regions of the receptor allowed or enforced by the extension of the **6a** side chain, that is **6a** binds in a manner similar to imipramine. Since different classes of 5-HT_{2A} ligands show different effects with respect to *N*-alkylation,[8] we explored a parallel series of *N*-alkylated derivatives of AMDA (**1a**–**1e**), the chain lengthened form of AMDA (**6a**–**6c**), and imipramine (**3a**–**3c**) to test this hypothesis.

N-Methylation of AMDA (Table 2) progressively decreases affinity. In contrast, mono- and dimethylation of the imipramine-like compound **6a** and of the imipramine analogue **3a** has little effect on affinity.

While the number of different alkylation patterns is limited in this series, the results suggest that, at least with respect to nitrogen substitution, the short and long chain series bind differently. One possible explanation for the difference between AMDA and **6** with respect to

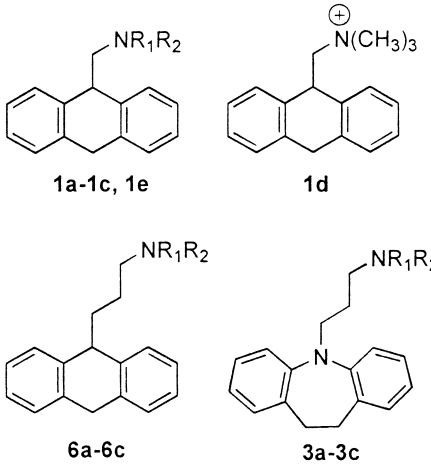
Table 1. The effect of chain length on 5-HT_{2A} receptor affinity and molecular geometry



Compound	*n*	d_1 (Å)[a]	d_2 (Å)[a]	d_3 (Å)[a]	K_i (nM)[b]
4	0	3.7	3.7	4.9	12,000
1a	1	3.8	5.2	4.9	20
5	2	5.2	6.2	4.9	480
6a	3	6.0	7.6	4.9	32
Imipramine	—	6.5	7.2	4.8	160
Cyproheptadine	—	6.1	6.1	4.9	1.6

[a]Distances measured for chain extended conformers.
[b][^{3}H]Ketanserin labeled cloned 5-HT_{2A} sites. Values represent the mean of computer-derived K_i estimates (using LIGAND) of quadruplicate determinations. Standard errors typically range between 15 and 25% of the K_i value.

Table 2. The effect of chain length on 5-HT_{2A} receptor affinity and molecular geometry



R_1	R_2	Compound	K_i (nM)[a] 1	6	3
H	H	**a**	20	32	392
CH_3	H	**b**	52	13	160
CH_3	CH_3	**c**	540	22	140
—	—	**d**	4000	—	—
CH_2Ph	H	**e**	721	—	—

[a][^{3}H]Ketanserin labeled, cloned 5-HT_{2A} sites. Values represent the mean of computer-derived K_i estimates (using LIGAND) of quadruplicate determinations. Standard errors typically range between 15 and 25% of the K_i value.

N-alkylation is that, due to the proximity of the tricyclic ring system and the steric bulk of the methyl groups, dimethyl AMDA preferentially adopts a conformation with a buried ammonium ion NH. While **6** can adopt a folded conformation that places the nitrogen within a region of space similar to that for AMDA, dimethylation similarly buries the ammonium ion NH. The steric clash of a folded dimethyl **6c** can be relieved by adopting a more chain extended and necessarily less AMDA-like conformation.

Classical tricyclic amines such as imipramine bind with high affinity to several neurotransmitter receptors and transporters (e.g., 5-HT_{2A} D_2, SERT, and NET).[9,10] Ligand SAR and ligand–receptor docking studies suggest that the imipramine-related and AMDA-related compounds interact with the 5-HT_{2A} receptor differently. This suggests that AMDA may behave differently with respect to other neurotransmitter receptors and binding sites. Table 3 shows the results of a preliminary selectivity study. Given the high degree of receptor sequence homology, it is not surprising that the AMDA does not differentiate between 5-HT_{2A} and 5-HT_{2C} sites. It is, however, quite remarkable that AMDA shows at least 500-fold selectivity for 5-HT_{2A} receptors versus D_2 receptors, and versus serotonin (SERT) and norepinephrine (NET) transporters.

The results suggest that AMDA and imipramine most likely bind to the 5-HT_{2A} receptor in different fashions, though the chain-lengthened aminoalkyl dihydroanthracene **6a** may be more imipramine-like in binding mode. While *N*-methylation decreases the 5-HT_{2A} receptor affinity of AMDA, *N*-methylation increases the affinity in the imipramine and chain-lengthened AMDA series. The fact that AMDA has a high degree of selectivity and imipramine does not suggests that AMDA and imipramine bind to the D_2 receptors, SERT, and NET differently as well. Thus, AMDA behaves quite differently from classical tricyclic amines and may be a suitable template for the construction of structurally novel, selective 5-HT_2 receptor antagonists.

Affinity Determinations

Binding assays and data analysis were performed as previously described using [^{3}H]ketanserin as the radioligand and stably transfected NIH3T3 cells expressing the 5-HT_{2A} receptor (GF-62 cells).[11] Full details for measurements of other receptors and transporters can be found at the NIMH Psychoactive Drug Screening Program web site (http://pdsp.cwru.edu/pdsp.htm).

Table 3. Receptor and transporter selectivities of AMDA (**1a**) and classical tricyclic agents

	K_i (nM)				
Comppound	5-HT_{2A}[a]	5-HT_{2C}[b]	D_2[c]	SERT[d]	NET[e]
AMDA (**1a**)	20	43	>10,000	>10,000	>10,000
Imipramine (**3c**)	94	160	726	5	16
Cyproheptadine (**2**)	3	11	112	4100	290

[a][^{3}H]ketanserin.
[b][^{3}H]mesulergine.
[c][^{3}H]spiperone.
[d][^{3}H]paroxetine.
[e][^{3}H]nisoxetine radioligands.

Ligand Synthesis

9-Aminomethyl-9,10-dihydroanthracene (**1a**) was prepared as previously described.[12] 9-(*N*-Methylaminomethyl)-9,10-dihydroanthracene (**1b**), 9-(*N*,*N*-dimethylaminomethyl)-9,10-dihydroanthracene (**1c**), and 9-(*N*-benzylaminomethyl)-9,10-dihydroanthracene (**1e**) were prepared by reduction (BH_3·THF) of the amides obtained by treatment of the acid chloride with methyl-, dimethyl-, and benzylamine. The quaternary ammonium bromide (**1d**) was obtained by treatment of **1c** with methyl bromide.[13]

Imipramine (**3c**) and desmethyl imipramine (**3b**) were purchased as the HCl salts. 5-(3-Aminopropyl)-(10,11)-dihydro-5*H*dibenz[*b,f*]azepine hydrochloride (**3a**)[13] was prepared by condensation of iminodibenzyl with 3-chloropropionyl chloride followed by reduction of the resulting amide[14] with BH_3·THF. The resulting 5-(3-chloropropyl)-10,11-dihydro-5*H*-dibenz[*b,f*]azepine[13] was treated with potassium phthalimide in anhydrous DMF and converted to the primary amine (**3a**)[13] using $NaBH_4$ in 2-propanol.[15]

9-Amino-9,10-dihydroanthracene oxalate (**4**) was prepared through a modified Hofmann rearrangement of 9,10-dihydroanthracene-9-carboxamide[1] using very mild conditions. [Bis(trifluoroacetoxy)iodo]benzene[16] in aqueous acetonitrile conveniently converted 9,10-dihydroanthracene-9-carboxamide to 9-amino-9,10-dihydroanthracene (**4**) in 59% yield.

9-(2-Aminoethyl)-9,10-dihydroanthracene oxalate (**5**) was prepared by condensation of 9-anthraldehyde with nitromethane in methylene chloride to provide *trans*-1-(9-anthryl)-2-nitroethylene,[17] which was reduced to 9-(2-aminoethyl)anthracene using LAH/THF. The target molecule (**5**) was then prepared through reduction using sodium metal in liquid ammonia.[18]

9-(2-Aminopropyl)-9,10-dihydroanthracene oxalate (**6a**), 9-[3-(*N*-methylaminopropyl)]-9,10-dihydroanthracene oxalate (**6b**), and 9-[3-(*N*,*N*-dimethylaminopropyl]-9,10-dihydroanthracene (**6c**) were prepared by the reduction (BH_3·THF) of the amides obtained by treatment of the acid chloride of 9-(2-carboxyethyl)-9,10-dihydroanthracene[19] with ammonia gas, methylamine, and dimethylamine, respectively.

Acknowledgements

This work was supported by United States Public Health Service Grant MH57969 (RBW), MH57635, MH01366 (BLR), the NIMH Psychoactive Drug Screening Program (BLR), and DA01642 (RAG).

References

1. Westkaemper, R. B.; Runyon, S. P.; Bondarev, M. L.; Savage, J. E.; Roth, B. L.; Glennon, R. A. *Eur. J. Pharmacol.* **1999**, *380*, R5.
2. Westkaemper, R. B.; Dukat, M.; Glennon, R. A. *Med. Chem. Res.* **1992**, *1*, 401.
3. Westkaemper, R. B.; Runyon, S. P.; Bondarev, M. L.; Savage, J. E.; Roth, B. L.; Glennon, R. A. *Bioorg. Med. Chem. Lett.*, in press.
4. Mokrosz, M. J.; Strekowski, L.; Kozak, W. X.; Duszynska, B.; Bojarski, A. J.; Klodzinska, A.; Czarny, A.; Cegla, M. T.; Deren-Wesooek, A.; Chojnacka-Wojcik, E.; Dove, S.; Mokrosz, J. L. *Arch. Pharm. (Weinheim)* **1995**, *328*, 659.
5. Andersen, K.; Liljefors, T.; Gundertofte, K.; Perregaard, J.; Bogeso, K. P. *J. Med. Chem.* **1994**, *37*, 950.
6. Post, M. L.; Kennard, O.; Horn, A. S. *Nature* **1974**, *252*, 493.
7. Heimstad, E.; Edvardsen, O.; Ferrin, T. E.; Dahl, S. G. *Eur. Neuropsychopharmacol.* **1991**, *1*, 127.
8. Glennon, R. A.; Dukat, M.; el-Bermawy, M.; Law, H.; De los Angeles, J.; Teitler, M.; King, A.; Herrick-Davis, K. *J. Med. Chem.* **1994**, *37*, 1929.
9. Toll, L.; Berzetei-Gurske, I. P.; Polgar, W. E.; Brandt, S. R.; Adapa, I. D.; Rodriguez, L.; Schwartz, R. W.; Haggart, D.; O'Brien, A.; White, A.; Kennedy, J. M.; Craymer, K.; Farrington, L.; Auh, J. S. *NIDA Res. Monogr.* **1998**, *178*, 440.
10. Richelson, E.; Pfenning, M. *Eur. J. Pharmacol.* **1984**, *104*, 277.
11. Westkaemper, R. B.; Hyde, E. G.; Choudhary, M. S.; Khan, N.; Gelbar, E. I.; Glennon, R. A.; Roth, B. L. *Eur. J. Med. Chem.* **1999**, *34*, 441.
12. Fouché, J. *Bull. Soc. Chim. Fr.* **1970**, 1376.
13. Hulinska, H.; Polivka, Z.; Jilek, J.; Sindelar, K.; Holubek, J.; Svatek, E.; Matousova, O.; Budesinski, M.; Frycova, H.; Protiva, M. *Collect. Czech. Chem. Commun.* **1988**, *53*, 1820.
14. Woodward, R. W.; Craig, J. C. *J. Lab. Comp. Radiopharm.* **1979**, *16*, 321.
15. Osby, J. O.; Martin, M. G.; Ganem, B.; Ganem, M. *Tetrahedron Lett.* **1984**, *25*, 2093.
16. Radhakrishna, A. S.; Parham, M. E.; Riggs, R. M.; Loudon, M. G. *J. Org. Chem.* **1979**, *44*, 1746.
17. Becker, H. D.; Sorensen, H.; Sandros, K. *J. Org. Chem.* **1986**, *51*, 3223.
18. Rabideau, P. W.; Wetzel, D. M.; Young, D. M. *J. Org. Chem.* **1984**, *49*, 1544.
19. Daub, G. H.; Doyle, C. *J. Am. Chem. Soc.* **1952**, *74*, 4449.