Pergamon

Bioorganic & Medicinal Chemistry Letters 9 (1999) 2167–2172

BIOORGANIC & MEDICINAL CHEMISTRY LETTERS

DERIVATIVES OF (*R*)-2-AMINO-5-METHOXYTETRALIN: Antagonists and Inverse Agonists at the Dopamine D_{2A} Receptor.

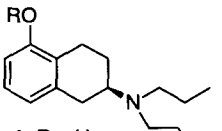
Berit Backlund Höök,[a] Cecilia Brege,[a] Tero Linnanen,[a] Åsa Mikaels,[b] Åsa Malmberg[b] and Anette M. Johansson,[a,*]

[a]*Organic Pharmaceutical Chemistry, Uppsala Biomedical Centre, Uppsala University, Box 574, SE-751 23 Uppsala, Sweden,* [b]*Dept. of Lead Generation, Preclinical R&D, Astra Arcus AB, SE-151 85 Södertälje, Sweden*

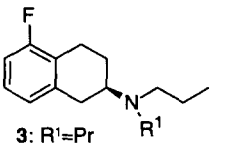
Received 9 April 1999; accepted 17 June 1999

Abstract: A series of *N*-arylmethyl substituted (*R*)-5-methoxy-2-(propylamino)tetralins has been prepared and evaluated for affinity and efficacy at dopamine (DA) D_{2A} receptors. The novel compounds appeared to be antagonists or inverse agonists. (*R*)-2-[(Benzyl)propylamino]-5-methoxytetralin (**7**) was characterized as a potent inverse agonists at DA D_{2A} receptors in a [^{35}S]GTPγS binding assay.

5-Oxygenated 2-aminotetralins have been of interest for medicinal chemists during the past 40 years. This is related to the various effects of 2-aminotetralin derivatives at dopamine (DA) receptors in the CNS.[1-4] The tetralin derivative **1** has in different biological assays been characterized as an antagonist as well as a partial agonist at DA D_2 receptors.[4,5] In addition, the corresponding methyl ether **2** was shown to be a partial DA D_2 receptor agonist with 4-fold higher affinity than **1**.[4] By substituting the 5-methoxy/5-hydroxy group in **1** or **2** for a fluorine, leading to **3**, does not change the intrinsic activity but decreased the affinity for D_{2A} receptors slightly.[4,6] Furthermore, by introducing a *N*-benzyl group in **3**, the inverse D_2 receptor agonist **4** is produced.[6] These previously reported results have now been utilized in the design of novel 2-aminotetralin-based D_{2A} receptor antagonists and inverse agonists.



1: R = H
2: R = Me



3: R^1=Pr
4: R^1=Bn

In this paper we present the synthesis and pharmacologial data of a novel series of *N*-arylmethyl substituted derivatives of **2**. The affinities of these novel compounds to high and low affinity sites at cloned human D_{2A} receptors were evaluated in binding studies *in vitro* using [^{3}H]quinpirole and [^{3}H]raclopride, respectively, as radioligands. The intrinsic activity of the compounds was determined in a [^{35}S]GTPγS binding assay. The novel compounds were characterized either as antagonists or inverse agonists at D_{2A} receptors, i.e. compounds characterized as antagonists did not affect basal [^{35}S]GTPγS binding whereas inverse agonists inhibited the basal binding. The *N*-benzyl-*N*-propyl substituted derivative **7** behaved as a potent inverse agonist by decreasing both the basal [^{35}S]GTPγS binding and the DA stimulated [^{35}S]GTPγS binding.

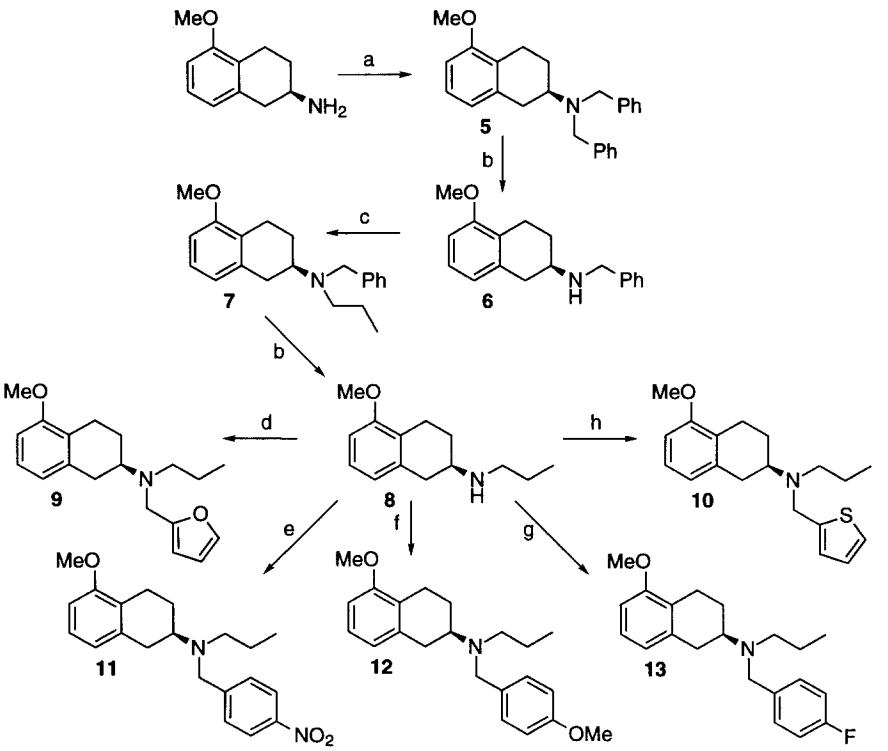
*e-mail: anette@bmc.uu.se FAX:+46-18-471 4024

0960-894X/99/$ - see front matter
PII: S0960-894X(99)00345-5

Synthesis.

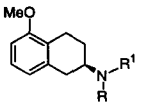
The novel derivatives were synthesized as shown in Scheme 1 and their physical data are summarized in Table 1. The stereochemical purity of (*R*)-2-amino-5-methoxytetralin (≥99 %ee) was determined indirectly by HPLC [Chiracel OD® column eluted with 2-propanol/iso-hexane (1:1)] after conversion of the amine to an amide by reaction with (*S*)-Mosher acid chloride.[7] (*R*)-2-Amino-5-methoxytetralin contained, however, about 6-7% of a methylated analog and was therefore first purified. To facilitate the purification, the mixture of primary amines was dialkylated with benzylbromide to give the dibenzylamines, which were chromatographed [SiO_2, ether/pentane (1:99) saturated with NH_3] to afford pure **5**. The purity of **5** was determined by GC to be > 99.5%. Monodebenzylation of **5**·HCl by catalytic hydrogenation afforded **6**, which was reductively alkylated with propanal and $NaCNBH_3$ to give the *N*-benzyl-*N*-propyl derivative **7**. The key-intermediate **8** was obtained from **7** by *N*-debenzylation. Five more *N*-arylmethyl derivatives were synthesized from **8** either by acylation followed by reduction (**9**, **10**) or by alkylation with the appropriate benzylhalide (**11-13**).

Scheme 1.



Reagents: a) $PhCH_2Br$, K_2CO_3, MeCN; b) H_2, Pd(C), HCl, MeOH; c) EtCHO, AcOH, MeOH, $NaCNBH_3$; d) i. 2-furoyl chloride, Et_3N, Et_2O, ii. $LiAlH_4$, THF ; e) 4-NO_2-$PhCH_2Br$, K_2CO_3, MeCN; f) 4-MeO-$PhCH_2Cl$, K_2CO_3, MeCN; g) 4-F-$PhCH_2Br$, K_2CO_3, MeCN; h) i. 2-thenoyl chloride, Et_3N, Et_2O, ii. $LiAlH_4$, THF.

Table 1. Physical Data of Some Novel (*R*)-2-Amino-5-methoxytetralin Derivatives.



Compd	R	R^1	yield (%)	mp, (°C)	recrystn solvent[a]	$[\alpha]_D$ (deg)[b]	Anal.
5	CH_2Ph	CH_2Ph	69	212-214	A	+58	$C_{25}H_{27}NO{\cdot}HCl$
6	H	CH_2Ph	80	243-245[c]	B	+63[d]	$C_{18}H_{21}NO{\cdot}HCl$
7	Pr	CH_2Ph	80	193-194	B	+58	$C_{21}H_{27}NO{\cdot}HCl$
8	Pr	H	96	273-274	B	+72	$C_{14}H_{21}NO{\cdot}HCl$
9	Pr	2-furfuryl	41	181-183	-	+59	$C_{19}H_{25}NO_2{\cdot}HCl$
10	Pr	2-thenyl	61	197-198	-	+57	$C_{19}H_{25}NOS{\cdot}HCl$
11	Pr	CH_2Ph-4-NO_2	70	191-192	B	+52	$C_{21}H_{26}N_2O_3{\cdot}HCl$
12	Pr	CH_2Ph-4-OMe	40	155-157	A	+51	$C_{22}H_{29}NO_2{\cdot}HCl$
13	Pr	CH_2Ph-4-F	69	193-194	-	+55	$C_{21}H_{26}FNO{\cdot}HCl$

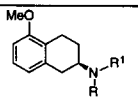
[a]Recrystallization solvent: (A) EtOH/ether; (B) MeOH/ether; [b](*c* 1.0, MeOH) measured at room temperature; [c]Litt mp 246-247 °C, see ref 13; [d]Litt $[\alpha]_D$ +61° (*c* 2.0, MeOH), see ref 13; +64.0° (*c* 2.0, MeOH), see ref 5.

In vitro radioligand binding and measurements of intrinsic activity.

The ability of the novel compounds to bind to and stimulate D_{2A} receptors was studied. The affinities of the compounds for the low and high-affinity sites at cloned human D_{2A} receptors were determined using *in vitro* receptor binding studies. The DA receptor antagonist [^{3}H]raclopride was used to label mainly the low-affinity site[8] and the high-affinity site was labelled by the DA receptor agonist [^{3}H]quinpirole.[9] The efficacy of the novel compounds at D_{2A} receptors was estimated by using two methods: (I) the calculated ratio between the affinity constants for the high- and low-affinity agonist site and (II) the G-protein activation-assay, [^{35}S]GTPγS binding. It has previously been shown that the ratio of the affinity constants for the high- and low-affinity agonist site correlates with the intrinsic activity of a compound.[6,10] Therefore, the calculated ratio between the affinity constants determined with [^{3}H]quinpirole and [^{3}H]raclopride was used as a measure of intrinsic activity. Furthermore, the intrinsic activity of the compounds was determined at D_{2A} receptors using the [^{35}S]GTPγS assay.[6,11] In this assay the ability of a compound to stimulate or inhibit the basal level of [^{35}S]GTPγS binding is a measurement of agonist or inverse agonist properties, respectively, whereas the ability of a compound to reverse a DA induced stimulation of [^{35}S]GTPγS binding is a measurement of antagonist properties. The affinities of the compounds for D_{2A} receptors are shown in Table 2 and the results from the [^{35}S]GTPγS binding assay are presented in Table 3.

The novel (*R*)-5-methoxy-2-aminotetralin derivatives display similar or lower D_{2A} receptor affinities compared to the parent compound **2**. The previously reported[3,12] importance of a *N*-propyl group for high affinity binding of 2-aminotetralin derivatives to D_{2A} receptors is also apparent in this series; the *N*-propyl group in **7** and **8** increased the affinity considerably in comparison to **5** and **6** which lack a *N*-propyl substituent. Interestingly, the exchange of a *N*-propyl group in **2** for a *N*-benzyl group decreased the affinity for the high affinity site of the D_{2A} receptors while the affinity for the low affinity site is almost unchanged. The electronic properties of the benzyl group does not dramatically influence the affinity to the D_{2A} receptors. However, the affinity of the heteroaromatic compounds **9** and **10** is slightly lower.

Table 2. Affinities of the Novel Derivatives to Cloned Human Dopamine D_{2A} Receptors Expressed in Ltk$^-$ Cells and Labelled by [^{3}H]Quinpirole and [^{3}H]Raclopride.



			K_i (nM)[a]			
			[^{3}H]Quinpirole	[^{3}H]Raclopride		Ratio
Compd	R	R^1	(D_{2Ahigh})	(D_{2Alow})	n_H[b]	(D_{2Alow}/D_{2Ahigh})
2	Pr	Pr	6.35±0.15[c]	46.0±3.6	1.06±0.11	7
5	CH_2Ph	CH_2Ph	>5000	>5000	-	-
6	H	CH_2Ph	1310±140	1010±40	0.83±0.03	0.8
7	Pr	CH_2Ph	75.0±3.9	65.6±0.3	1.05±0.05	0.9
8	Pr	H	82.3±13.9	114±6	1.06±0.02	1
9	Pr	2-furfuryl	118±18	125±9	0.83±0.04	1
10	Pr	2-thenyl	339±93	152±14	0.90±0.08	0.5
11	Pr	CH_2Ph-4-NO_2	215±59	59.2±6.8	0.90±0.00	0.3
12	Pr	CH_2Ph-4-OMe	64.1±5.8	22.0±1.8	0.94±0.00	0.3
13	Pr	CH_2Ph-4-F	69.7±15.2	43.8±10.2	0.90±0.10	0.6
Dopamine			1.89±0.25[d]	759±4[e]	0.65±0.02[e]	400
Haloperidol			0.16±0.03[d]	0.44±0.01[e]	1.03±0.02[e]	3
3[f]			26.0±1.7	201±12	0.89±0.02	8
4[f]			179±61	415±41	0.86±0.06	2

[a]For experimental details see refs 6, 8 and 9. The K_i values are means ± standard errors of two to three experiments. [b]Hill coefficients are given for [^{3}H]Raclopride binding where high and low affinity agonist states can be determined; [c]From ref 4; [d]From ref 9; [e]From ref 14;. [f]From ref 6.

The affinity of the novel derivatives for the high affinity site of the D_{2A} receptor was shown to be about 10-200 times lower when compared to that of **2**. However, at the low affinity site of the D_{2A} receptor several of the compounds have similar affinity as **2**. Thus, the calculated ratio of the affinity constants for the high and low affinity agonist sites at the D_{2A} receptors were lower for all the novel compounds when compared with **2**. The ratios were ≤ 1 indicating an absence of or a low intrinsic activity at the D_{2A} receptors. Interestingly, several of the compounds (**6, 7, 10-13**) were shown to have higher affinity for the low affinity site then for the high affinity site: **11** had the highest selectivity for the low affinity site (D_{2Alow}/D_{2Ahigh}=0.3) and **12** was the most potent derivative at the low affinity site (K_i=22 nM).

These results are in agreement with the results obtained in the [^{35}S]GTPγS binding assay. None of the novel derivatives displayed any intrinsic activity at the D_{2A} receptors in this assay. Compounds **7**, **9**, **12** and **13** inhibited both the basal [^{35}S]GTPγS binding and the DA induced stimulation of [^{35}S]GTPγS binding and were characterized as inverse agonists. The *N*-benzyl-*N*-propyl derivative **7** is the most potent inverse agonist of the novel derivatives being almost as efficacious as haloperidol in the [^{35}S]GTPγS binding assay. In comparison with the previously published inverse agonist **4**,[6] **7** has higher affinity at both D_{2A} receptor binding sites and is a more efficacious inverse agonist. Also when the corresponding *N,N*-dipropyl derivatives **2** and **3** are compared, the C5-methoxy substituted **2** has higher potency and more antagonistic properties than the C5-fluoro derivative **3**. Derivatives **8** and **11** did not display any intrinsic activity but inhibited the DA stimulated [^{35}S]GTPγS binding and were therefore classified as D_{2A} receptor antagonists. The 2-thenyl derivative **10** also seemed to be a

Table 3. Effects of the Novel Derivatives on [^{35}S]GTPγS Binding to Cell Membranes Expressing Dopamine D_{2A} Receptors.

	Stimulation or Inhibition of Basal [^{35}S]GTPγS Binding[a]		
Compd	Dose (μM)	(%)[b]	+ 100 μM dopamine[c]
Dopamine	100	16.0 ± 2.0	
2	1	2.3 ± 0.3**	10.0 ± 3.7
	10	3.4 ± 0.2***	6.6 ± 3.0*
	100	3.0 ± 1.3	2.9 ± 2.8*
5	1	-2.5 ± 2.5	14.4 ± 2.0
	10	NT[d]	NT[d]
	100	NT[d]	NT[d]
6	1	-1.4 ± 0.5	15.8 ± 4.1
	10	0.9 ± 1.0	13.4 ± 2.8
	100	1.6 ± 1.5	13.0 ± 1.7
7	1	-0.2 ± 1.4	13.6 ± 5.7
	10	-0.5 ± 1.9	9.2 ± 5.1
	100	-9.0 ± 1.8*	-7.4 ± 1.0**
8	1	-1.7 ± 1.8	14.2 ± 3.3*
	10	-1.9 ± 2.2	11.2 ± 2.6*
	100	-4.3 ± 2.4	4.9 ± 3.4**
9	1	-1.5 ± 1.4	10.6 ± 4.7
	10	-0.4 ± 1.2	9.5 ± 2.5**
	100	-9.6 ± 3.0*	-0.4 ± 2.5***
10	1	2.0 ± 1.1	14.3 ± 3.8
	10	0.6 ± 0.4	11.4 ± 2.8
	100	-0.5 ± 0.7	10.9 ± 1.9
11	1	-2.0 ± 1.7	13.9 ± 2.2
	10	0.9 ± 0.9	11.9 ± 2.7*
	100	3.7 ± 2.6	10.7 ± 2.6*
12	1	0.5 ± 0.2	10.8 ± 2.9
	10	-0.8 ± 1.8	5.1 ± 1.2**
	100	-6.6 ± 1.1*	1.4 ± 3.7**
13	1	-0.9 ± 1.1	12.3 ± 2.9
	10	1.3 ± 0.3	10.1 ± 0.5
	100	-5.2 ± 0.4**	2.0 ± 1.0
Haloperidol	1	-2.2 ± 1.7	-3.1 ± 1.6*
	10	-4.1 ± 1.1*	-2.8 ± 1.3*
	100	-12.1 ± 1.3**	-10.5 ± 1.6*
3[e]	100	3.1 ± 1.6	10.5 ± 1.3***
4[f]	1	3.2 ± 0.8	3.5 ± 7.4
	10	1.0 ± 2.2	4.3 ± 7.9
	100	-8.4 ± 0.4*	-3.1 ± 0.1

[a]For experimental details see ref 6. The values are given as percent stimulation or inhibition (means ± standard errors of three to four independent experiments) of basal [^{35}S]GTPγS binding. [b]The compounds were tested alone. An asterisk indicates statistical significance difference as compared with basal value zero: *p <0.05, ** p <0.01 (Student's paired t-test). [c]The compounds were tested together with dopamine (100 μM). An asterisk indicates statistical significance difference as compared with dopamine stimulation: *p <0.05, ** p <0.01, *** p <0.001 (Student's paired t-test). [d]NT=not tested, this is due to solubility problems. [e]From ref 6. [f]n=2.

antagonist at D_{2A} receptors although no significant values were obtained. Compounds **5** and **6** had low affinity at the D_{2A} receptors and were inactive in the [^{35}S]GTPγS binding assay. Consistent with literature data, both haloperidol and **4** behaved as inverse agonists at D_{2A} receptors,[6] while **2** might be a partial agonist[4] because it slightly stimulated the basal [^{35}S]GTPγS binding and inhibited the DA induced stimulation of [^{35}S]GTPγS binding.

Conclusion.

These results indicate that both the C5-substituent and the *N*-substituents of (*R*)-2-aminotetralins are of importance for the affinity and intrinsic activity at D_{2A} receptors. The affinity for the D_{2A} receptors is gradually increased by changing the C5-substituent in (*R*)-2-(dipropylamino)tetralin from a hydrogen (D_{2Ahigh}:31.7 nM; D_{2Alow}:554 nM[6]) to a fluorine and then to a methoxy group, the main increase being observed at the low affinity site. At least one *N*-propyl group seems to be of crucial importance for high affinity to the D_{2A} receptors. The previously reported C5-fluoro and the novel C5-methoxy series of derivatives show the same trends in changes in affinity and intrinsic activity at D_{2A} receptors when the *N*-propyl, the *N*,*N*-dipropyl and the *N*-benzyl-*N*-propyl analogues are compared. An increase in affinity mainly at the high affinity D_{2A} receptor site is observed when a second *N*-propyl group is introduced, whereas for the corresponding *N*-benzyl-*N*-propylamines the affinity for the high affinity site is decreased and the intrinsic activity is reduced, thereby changing the profile towards inverse agonism.

Acknowledgment.

The authors are grateful to Emil Schwan for skillful technical assistance. Financial support and (*R*)-2-amino-5-methoxytetralin were obtained from Astra Arcus AB.

References and Notes.

1. Hacksell, U.; Johansson, A.M.; Karlén, A.; Svensson, K.; Grol, C.J. In "Chirality and Biological Activity", Eds. H. Frank, B. Holmstedt, and B. Testa, Alan R. Liss, Inc., N.Y., **1989**, pp 247-266.
2. Wikström, H. *Prog. Med. Chem.* **1992**, *29*, 185-216.
3. Malmberg, Å.; Nordvall, G.; Johansson, A.M.; Mohell, N.; Hacksell, U. *Mol. Pharmacol.* **1994**, *46*, 299-312.
4. Malmberg, Å.; N. Mohell, Backlund Höök, B.; Johansson, A.M.; Hacksell, U.; Nordvall, G. *Eur. J. Pharmacol.* **1998**, *346*, 299-307.
5. Karlsson, A.; Björk, L.; Pettersson, C.; Andén, N.-E.; Hacksell, U. *Chirality*, **1990**, *2*, 90-95.
6. Malmberg, Å.; Backlund Höök, B.; Johansson, A.M.; Hacksell, U. *J. Med. Chem.* **1996**, *39*, 4421-4429.
7. Ward, D.E.; Rhee, C.K. *Tetrahedron Lett.* **1991**, *32*, 7165-7166. The (*S*)-Mosher acid (Fluka, no 65369) had a stereochemical purity of ≥99 %ee.
8. Malmberg, Å.; Jackson, D.M.; Eriksson, A.; Mohell, N. *Mol. Pharmacol.* **1993**, *43*, 749-754.
9. Malmberg, Å. and Mohell, N.; *J. Pharmacol. Exp. Ther.* **1995**, *274*, 790-797.
10. a) De Lean, A.; Stadel, J.M.; Lefkowitz, R.J. *J. Biol. Chem.* **1980**, *255*, 7108-7117, b) Kent, R.S.; De Lean, A.; Lefkowitz, R.J. *Mol. Pharmacol.* **1980**, *17*, 14-23, c) Freedman, S.B.; Harley, E.A.; Iversen, L.L. *Br. J. Pharmacol.* **1988**, *93*, 437-445, d) Lahti, R.A.; Figur, L.M.; Piercey, M.F.; Ruppel, P.L.; Evans, D.L. *Mol.Pharmacol.* **1992**, *42*, 432-438.
11. a) Lazareno, S.; Farries, T.; Birdsall, N.J.M. *Life Sci.* **1993**, *52*, 449-456, b) Wieland, T.; Jakobs, K.H. In *Heterotrimeric G proteins*; Iyengar, R., Ed.; Academic Press, Inc.: San Diego, CA, 1994; pp 3-13. c) Gardner, B.; Strange, P.G. *Biochem. Soc. Trans.* **1995**, *23*, 91S.
12. a) Hacksell, U.; Svensson, U.; Nilsson, J.L.G.; Hjorth, S.; Carlsson, A.; Wikström, H.; Lindberg, P.; Sanchez, D.; *J. Med. Chem.* **1979**, *22*, 1469-1475.
13. McDermed, J.D.; McKenzie, G.M.; Freeman, H.S. *J. Med. Chem.* **1976**, *19*, 547-549.
14. Malmberg, Å.; Mikaels, Å.; Mohell, N. *J. Pharmacol. Exp. Ther.* **1998**, *285*, 119-126.