Structure–Activity Relationship Studies of Central Nervous System Agents. 13. 4-[3-(Benzotriazol-1-yl)propyl]-1-(2-methoxyphenyl)piperazine, a New Putative 5-HT_{1A} Receptor Antagonist, and Its Analogs[1]

Jerzy L. Mokrosz,* Maria H. Paluchowska, Ewa Chojnacka-Wójcik,† Małgorzata Filip,† Sijka Charakchieva-Minol, Anna Dereń-Wesołek,† and Maria J. Mokrosz

Department of Medicinal Chemistry and Laboratory of New Drugs, Institute of Pharmacology, Polish Academy of Sciences, 12 Smętna Street, 31-343 Kraków, Poland

Received February 23, 1994®

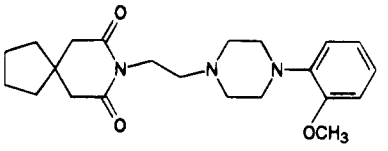
A new set of 4-alkyl-1-(*o*-methoxyphenyl)piperazines containing a terminal benzotriazole fragment were synthesized, and their 5-HT_{1A} and 5-HT_2 affinity was determined. It was shown that the benzotriazole moiety contributes to both the 5-HT_{1A} and 5-HT_2 receptor affinity. It was demonstrated in several behavioral models that 4-[3-(benzotriazol-1-yl)propyl]-1-(2-methoxyphenyl)piperazine (**11**) is a new, potent presynaptic and postsynaptic 5-HT_{1A} receptor antagonist. However, it is not selective for 5-HT_{1A} versus α_1 receptors.

Introduction

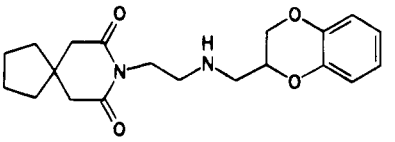
Of different subpopulations of the G-coupled serotonin (5-HT) receptors, in the past decade the utmost attention was focused on the 5-HT_{1A} subtype. A very large number of new 5-HT_{1A} ligands have been synthesized, and their receptor affinities and intrinsic activities have been reported in numerous papers. Fortunately, these issues have been extensively reviewed.[2–7] Although a number of selective 5-HT_{1A} receptor agonists have been developed, there have been only few candidates to date which can be regarded as potential selective antagonists at both presynaptic and postsynaptic 5-HT_{1A} receptors (Chart 1). Most of them (e.g., **1**–**4**), previously thought to act as 5-HT_{1A} receptor antagonists, are now classified as partial agonists.[7,8] Compound **5** seems to be the first highly selective and potent full 5-HT_{1A} receptor antagonist.[8,9] Its presynaptic and postsynaptic antagonism has been shown in electrophysiological, biochemical, and behavioral studies.[9–14] It has also been reported that (*S*)-5-fluoro-8-hydroxy-2-(di-*n*-propylamino)tetralin acts as a full 5-HT_{1A} receptor antagonist, but it has a low, 8-fold, D_2/5-HT_{1A} selectivity ratio.[15,16]

All the compounds shown in Chart 1 belong to the arylpiperazine class of 5-HT_{1A} ligands, except **2** which contains the 2-(aminomethyl)-benzodioxan moiety. It should be stressed that all these compounds contain a methoxy or a carbomethoxy groups which are commonly known acceptors of the hydrogen bond; moreover, such a structural feature of the 1-arylpiperazine fragment may be important for their 5-HT_{1A} receptor antagonism. Furthermore, the 2-(aminomethyl)benzodioxan portion of **2** mimics very well the structure of 1-(*o*-methoxyphenyl)piperazine. According to Glennon,[5] there appears to be no clear-cut relationship between the chain length of 4-substituted 1-arylpiperazines and their intrinsic activity. Although structural criteria for the intrinsic activity of long-chain arylpiperazines has been lacking, to date, it may be postulated that the electronic structure of the terminal cyclic amide fragment is responsible for their 5-HT_{1A} receptor antagonism. Indeed, in both **3** and **4**—potent postsynaptic 5-HT_{1A} receptor antagonists—there exists a strong conjugation of the amide group with the aromatic ring. Moreover, in light of recent findings of El-Bermawy et al.[17] and Mokrosz and Duszyńska,[18] it is also noteworthy that the terminal

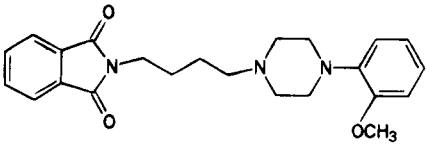
Chart 1. Structures of the Selected 5-HT_{1A} Receptor Antagonists and Compound **11**



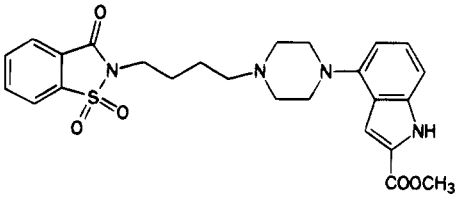
(1)



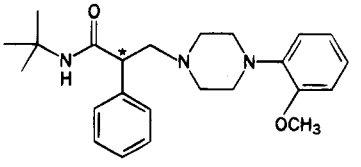
(2)



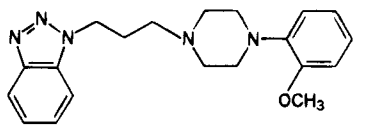
(3)



(4)



(5)



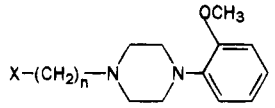
(11)

† Laboratory of New Drugs.

® Abstract published in *Advance ACS Abstracts,* June 15, 1994.

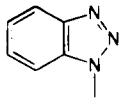
0022-2623/94/1837-2754$04.50/0

Chart 2. Structures of the Investigated Compounds

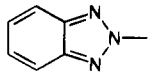


6 - 12, 15, 16

X: H,



1 - Bt



2 - Bt

n: 2, 3, 4

Table 1. Structures and 5-HT_{1A} and 5-HT_2 Receptor Binding Data of Compounds **6**–**12**, **15**, and **16**

	structure		$K_i \pm$ SEM, nM[b]		selectivity
no.	X	*n*	5-HT_{1A}	5-HT_2	5-HT_2/5-HT_{1A}
6	H	2	203 ± 10	3120 ± 350	15
7	H	3	249 ± 15	3290 ± 260	13
8	H	4	57 ± 4	2280 ± 280	40
9	1-Bt	2	42 ± 3	1620 ± 230	39
10	2-Bt	2	41 ± 9	1760 ± 200	43
11	1-Bt	3	15 ± 2	1040 ± 30	69
12	2-Bt	3	24 ± 2	1580 ± 120	66
15	1-Bt	4	17 ± 6	160 ± 25	9.4
16	2-Bt	4	4 ± 1	214 ± 19	54

[a] Abbreviations correspond to those shown in Chart 2. [b] Mean value from at least three independent experiments.

amide function of long-chain arylpiperazines is not important for the binding and that its removal yields high-affinity 5-HT_{1A} receptor agents.

In the present paper we have analyzed a new set of model arylpiperazines, **9**–**12**, **15**, and **16** (Chart 2, Table 1). The benzotriazole ring system has been chosen as a terminal fragment of 4-alkyl-1-(*o*-methoxyphenyl)-piperazines, due to the following advantages: firstly, a strong conjugation—similar to that found in **3** or **4**—exists within the ring system; secondly, the system is devoid of any additional functional groups, and the only effects of stabilization may stem from either π-electron or local dipole–dipole interactions at the receptor.

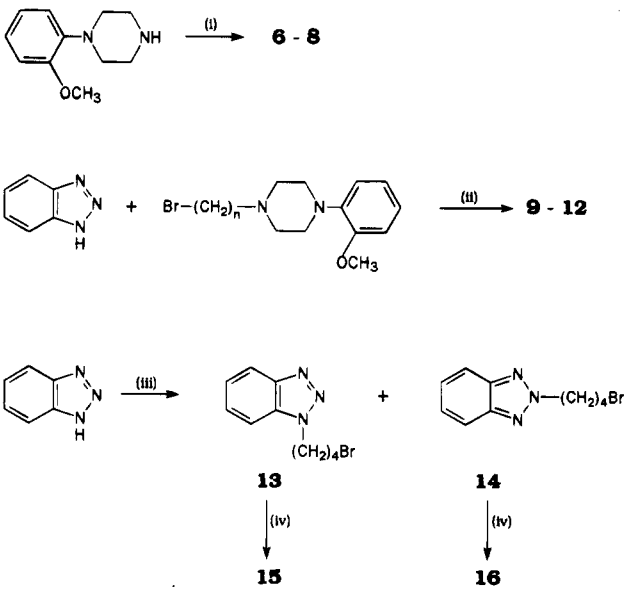
Chemistry

Compounds **6**–**12**, **15**, and **16** (Table 1) were obtained by a simple one- or two-step condensation with a yield of 18–74% (Chart 3). All of the compounds had satisfactory elemental analyses, and their mass spectra and/or ^1H NMR spectra were consistent with the proposed structures. The ^1H NMR spectra differentiated clearly between 1- and 2-benzotriazole isomers regarding the characteristic pattern of their spectra. The resonance signal of the 2-BtCH_2 methylene group was shifted downfield by 0.1–0.2 ppm in relation to that observed for isomer 1-BtCH_2. Moreover, proton signals of the 2-benzotriazolyl fragments formed a diagnostic, symmetrical pattern in the spectrum.[19]

Radioligand Binding Studies

The affinity of compounds **6**–**12**, **15**, and **16** for central 5-HT_{1A} and 5-HT_2 receptors in vitro was assessed on the basis of their ability to displace [^3H]-8-OH-DPAT and [^3H]ketanserin, respectively. The obtained results are summarized in Table 1. In order to analyze the effect of the terminal benzotriazole moiety on the 5-HT_{1A} and 5-HT_2 receptor affinity of derivatives **6**–**12**, **15**, and **16**, we chose the appropriate 4-*n*-alkyl-1-(*o*-methoxyphenyl)piperazines **6**–**8** as reference compounds.

Chart 3



Replacement of the hydrogen atom in **6**–**8** with 1- or 2-benzotriazole fragments resulted in a significantly higher affinity of **6**–**12**, **15**, and **16** for both 5-HT_{1A} and 5-HT_2 receptors (Table 1). In general, the investigated benzotriazole derivatives showed a lower 5-HT_2 than 5-HT_{1A} affinity, and the most significant 5-HT_2/5-HT_{1A} selectivity ratio was found for the isomer pair **11** and **12** (Table 1).

In Vivo Experiments

The Behavioral Syndrome Induced by 8-OH-DPAT in Reserpinized Rats. 8-OH-DPAT injected in a dose of 5 mg/kg produced a flat body posture and reciprocal forepaw treading in reserpine-pretreated rats, the maximum behavioral score being 14.8 and 14.5, respectively (Table 2). Compounds **11** (4–16 mg/kg) and **16** (2–16 mg/kg) antagonized dose-dependently the 8-OH-DPAT-induced symptoms, having produced an almost complete blockade of the forepaw treading (both compounds) and flat body posture (only **16**) after the highest dose used. Compounds **12** (4–16 mg/kg) and **15** (8–16 mg/kg) reduced the forepaw treading but failed to inhibit the flat body posture produced by 8-OH-DPAT. At the same time, all four compounds (**11**, **12**, **15**, and **16**), injected in doses of 2–16 mg/kg to normal rats, did not yield any component of the 8-OH-DPAT-induced syndrome (data not shown).

The Lower Lip Retraction (LLR) Induced by 8-OH-DPAT in Rats. 8-OH-DPAT (1 mg/kg ip) induced LLR, the maximum possible score being 87–93%. The effect of 8-OH-DPAT was dose-dependently antagonized by compound **11** (2–16 mg/kg), whereas derivatives **12**, **15**, and **16** (2–16 mg/kg) practically did not affect the LLR induced by 8-OH-DPAT (Table 2). Moreover, compounds **11**, **12**, and **15**, given alone in doses of 2–16 mg/kg, had no activity in that test; only

Table 2. The Effect of Compounds **11**, **12**, **15**, **16**, and **5** on the 8-OH-DPAT-Induced Behavior in Reserpine-Pretreated Rats[a] (A) and on the 8-OH-DPAT-Induced Lower Lip Retraction (LLR)[b] (B) and Induction of LLR by the Investigated Compounds in Rats[c] (C)

		A: mean ± SEM behavioral score		mean ± SEM LLR score	
treatment	dose, mg/kg	flat body posture	forepaw treading	B	C
vehicle	–	14.8 ± 0.2	14.3 ± 0.3	2.6 ± 0.2	0.1 ± 0.1
11	2	14.8 ± 0.3	13.4 ± 0.8	2.3 ± 0.3	0.1 ± 0.1
	4	11.5 ± 1.2	5.8 ± 1.7[d]	1.1 ± 0.2[d]	0.0 ± 0.0
	8	8.0 ± 2.0[d]	3.7 ± 1.5[d]	0.7 ± 0.3[d]	0.1 ± 0.1
	16	6.1 ± 0.3[d]	2.6 ± 0.9[d]	0.9 ± 0.2[d]	0.6 ± 0.2
vehicle	–	14.2 ± 0.4	14.5 ± 0.3	2.6 ± 0.2	0.1 ± 0.1
12	2	NT	NT	2.3 ± 0.2	0.2 ± 0.2
	4	13.8 ± 0.5	8.0 ± 1.9[d]	1.8 ± 0.3	0.2 ± 0.2
	8	11.8 ± 1.5	4.8 ± 1.1[d]	1.8 ± 0.4	0.4 ± 0.3
	16	12.4 ± 0.9	4.0 ± 1.1[d]	2.2 ± 0.2	0.3 ± 0.2
vehicle	–	14.8 ± 0.3	14.0 ± 0.4	2.8 ± 0.1	0.2 ± 0.1
15	2	NT	NT	2.2 ± 0.2	0.2 ± 0.1
	4	11.0 ± 1.2	10.3 ± 1.8	2.1 ± 0.2	0.3 ± 0.2
	8	8.2 ± 1.4	3.6 ± 1.3[d]	1.9 ± 0.2	0.1 ± 0.1
	16	12.7 ± 0.8	6.1 ± 1.5[d]	2.4 ± 0.2	0.7 ± 0.3
vehicle	–	14.2 ± 0.4	13.8 ± 0.8	2.8 ± 0.1	0.1 ± 0.1
16	1	14.2 ± 0.4	13.6 ± 0.5	NT	NT
	2	9.3 ± 2.5	5.6 ± 1.8[d]	2.5 ± 0.2	0.3 ± 0.1
	4	11.7 ± 1.3	5.1 ± 1.8[d]	2.3 ± 0.2	1.0 ± 0.4
	8	5.2 ± 0.8[d]	2.4 ± 0.9[d]	2.3 ± 0.3	1.5 ± 0.2[d]
	16	2.6 ± 1.4[d]	1.3 ± 0.6[d]	2.4 ± 0.2	1.9 ± 0.3[d]
vehicle[e]	–	15.0 ± 0.0	13.2 ± 0.9	2.8 ± 0.4	0.1 ± 0.1
5[c]	10	9.7 ± 1.0[d]	1.0 ± 0.3[d]	0.8 ± 0.3[d]	0.1 ± 0.1

[a] Reserpine (1 mg/kg sc) and the investigated compounds (ip) were administered 18 h and 30 min, respectively, before 8-OH-DPAT (5 mg/kg ip). [b] The investigated compounds were administered ip 30 min before 8-OH-DPAT (1 mg/kg ip). [c] The investigated compounds were administered ip 45 min before the test. [d] $p < 0.01$ vs vehicle. [e] Data for comparison taken from ref 12. NT = not tested.

Table 3. The effect of Compounds **11**, **12**, **15**, **16**, and **5** on the 8-OH-DPAT-Induced Hypothermia in Mice[a]

	Δt ± SEM, °C			
treatment (dose), mg/kg	15 min	30 min	45 min	60 min
vehicle + vehicle	−0.1 ± 0.1	0.0 ± 0.1	0.2 ± 0.2	0.1 ± 0.2
vehicle + 8-OH-DPAT (5)	−1.2 ± 0.2[c]	−1.1 ± 0.2[c]	−0.8 ± 0.2[c]	−0.6 ± 0.2[c]
11 (1) + 8-OH-DPAT (5)	−1.5 ± 0.3[c]	−1.1 ± 0.2[c]	−0.6 ± 0.2[c]	−0.3 ± 0.2
11 (2) + 8-OH-DPAT (5)	−0.4 ± 0.3[d]	−0.2 ± 0.2[e]	−0.1 ± 0.1	−0.1 ± 0.1
11 (4) + 8-OH-DPAT (5)	−0.3 ± 0.2[e]	−0.1 ± 0.2[e]	0.0 ± 0.2[d]	0.1 ± 0.2
11 (8) + 8-OH-DPAT (5)	0.3 ± 0.1[e]	0.4 ± 0.1[e]	0.7 ± 0.2[e]	0.6 ± 0.1[d]
vehicle + vehicle	−0.2 ± 0.1	0.0 ± 0.1	0.0 ± 0.1	0.0 ± 0.1
vehicle + 8-OH-DPAT (5)	−1.0 ± 0.1[b]	−1.1 ± 0.2[c]	−1.0 ± 0.2[c]	−0.8 ± 0.2[b]
12 (4) + 8-OH-DPAT (5)	−1.8 ± 0.2[c,e]	−1.4 ± 0.3[c]	−0.9 ± 0.2[b]	−0.6 ± 0.3
12 (8) + 8-OH-DPAT (5)	−2.0 ± 0.5[c]	−1.5 ± 0.5[c]	−0.9 ± 0.3[b]	−0.7 ± 0.3
12 (16) + 8-OH-DPAT (5)	−2.6 ± 0.3[c,e]	−2.0 ± 0.4[c]	−1.3 ± 0.3[c]	−0.9 ± 0.3[b]
vehicle + vehicle	0.0 ± 0.1	0.1 ± 0.1	0.0 ± 0.1	0.1 ± 0.1
vehicle + 8-OH-DPAT (5)	−1.1 ± 0.4[c]	−1.6 ± 0.4[c]	−1.2 ± 0.4[c]	−1.0 ± 0.3
15 (4) + 8-OH-DPAT (5)	−3.9 ± 0.2[c,e]	−4.2 ± 0.3[c,e]	−3.8 ± 0.5[c,e]	−3.2 ± 0.6[c,e]
15 (8) + 8-OH-DPAT (5)	−3.9 ± 0.3[c,e]	−4.1 ± 0.3[c,e]	−3.5 ± 0.3[c,e]	−2.7 ± 0.2[c,e]
15 (16) + 8-OH-DPAT (5)	−5.3 ± 0.3[c,e]	−5.6 ± 0.4[c,e]	−5.6 ± 0.5[c,e]	−5.2 ± 0.6[c,e]
vehicle + vehicle	−0.2 ± 0.1	−0.2 ± 0.1	−0.3 ± 0.1	−0.3 ± 0.1
vehicle + 8-OH-DPAT (5)	−1.4 ± 0.3[c]	−1.5 ± 0.3[c]	−1.2 ± 0.3[c]	−1.0 ± 0.3[b]
16 (2) + 8-OH-DPAT (5)	−2.1 ± 0.5[c]	−1.9 ± 0.4[c]	−1.2 ± 0.3[c]	−0.9 ± 0.2
16 (4) + 8-OH-DPAT (5)	−2.7 ± 0.3[c,e]	−2.3 ± 0.3[c]	−1.8 ± 0.3[c]	−1.3 ± 0.4
16 (8) + 8-OH-DPAT (5)	−2.4 ± 0.3[c,e]	−2.2 ± 0.4[c]	−1.6 ± 0.4[b]	−1.1 ± 0.3
vehicle + vehicle[f]	0.3 ± 0.1	0.2 ± 0.1	0.1 ± 0.2	0.1 ± 0.3
vehicle + 8-OH-DPAT (5)[f]	−1.3 ± 0.3[c]	−1.6 ± 0.2[c]	−1.8 ± 0.4[c]	−1.4 ± 0.3[c]
5 (10) + 8-OH-DPAT (5)[f]	0.2 ± 0.2[e]	0.0 ± 0.2[e]	0.3 ± 0.2[e]	0.1 ± 0.2[e]

[a] The investigated compounds were administered ip 30 min before 8-OH-DPAT. The absolute mean initial body temperatures were within a range of 36.1 ± 0.5 °C. [b] $p < 0.05$ vs vehicle + vehicle. [c] $p < 0.01$ vs vehicle + vehicle. [d] $p < 0.05$ vs vehicle + 8-OH-DPAT. [e] $p < 0.01$ vs vehicle + 8-OH-DPAT. [f] Data for comparison taken from ref 12.

derivative **16** (4–16 mg/kg) induced LLR in rats, the maximum score being 63% after the highest used dose (Table 3).

The Hypothermia Induced by 8-OH-DPAT in Mice. 8-OH-DPAT (5 mg/kg ip) decreased the rectal body temperature in mice, the maximum hypothermic effect being observed at 30 min after administration. Compound **11** (2–8 mg/kg) reduced dose-dependently the 8-OH-DPAT-induced hypothermia, and at the highest dose of 8 mg/kg the effect was completely abolished. Contrariwise, compounds **12** (4–16 mg/kg), **15** (4–16 mg/kg), and **16** (2–8 mg/kg) potentiated and prolongated the hypothermia induced by 8-OH-DPAT (Table 3). On the other hand, compounds **11** (1–8 mg/kg) and **12** (4–16 mg/kg) given alone did not show any activity nor did they change the body temperature of mice, whereas

Table 4. The Effect of the Compounds **11**, **12**, **15**, **16**, and **5** on Body Temperature in Mice[a]

treatment	dose, mg/kg	Δt ± SEM, °C				
		30 min	45 min	60 min	75 min	90 min
vehicle	–	−0.1 ± 0.1	−0.2 ± 0.2	0.1 ± 0.2	−0.1 ± 0.2	−0.1 ± 0.2
11	1	0.0 ± 0.1	0.1 ± 0.1	0.1 ± 0.2	0.0 ± 0.2	0.1 ± 0.2
	2	−0.3 ± 0.1	−0.1 ± 0.1	0.2 ± 0.1	0.2 ± 0.2	−0.1 ± 0.2
	4	−0.4 ± 0.1	−0.2 ± 0.1	0.1 ± 0.2	0.0 ± 0.2	−0.2 ± 0.1
	8	−0.4 ± 0.2	−0.2 ± 0.2	0.1 ± 0.1	0.2 ± 0.1	0.2 ± 0.2
vehicle	–	−0.1 ± 0.1	0.1 ± 0.1	0.1 ± 0.2	0.1 ± 0.2	0.1 ± 0.2
12	4	−0.1 ± 0.1	0.3 ± 0.2	0.1 ± 0.2	0.2 ± 0.2	0.1 ± 0.2
	8	−0.1 ± 0.2	0.2 ± 0.2	0.4 ± 0.2	0.3 ± 0.3	0.3 ± 0.2
	16	−0.2 ± 0.1	0.3 ± 0.1	0.3 ± 0.1	0.4 ± 0.2	0.2 ± 0.2
vehicle	–	−0.2 ± 0.1	0.0 ± 0.1	0.0 ± 0.2	−0.2 ± 0.1	−0.2 ± 0.2
15	4	−1.3 ± 0.3[c]	−1.4 ± 0.3[c]	−0.9 ± 0.3	−0.8 ± 0.3	−0.7 ± 0.3
	8	−2.4 ± 0.3[c]	−2.3 ± 0.4[c]	−2.1 ± 0.3[c]	−1.6 ± 0.4[c]	−1.3 ± 0.3[c]
	16	−3.0 ± 0.3[c]	−3.3 ± 0.4[c]	−3.3 ± 0.5[c]	−2.9 ± 0.5[c]	−2.5 ± 0.5[c]
vehicle	–	0.0 ± 0.1	0.0 ± 0.1	0.1 ± 0.1	0.0 ± 0.1	0.2 ± 0.1
16	2	−0.1 ± 0.1	0.0 ± 0.2	−0.2 ± 0.2	−0.3 ± 0.2	0.1 ± 0.2
	4	−0.9 ± 0.1[c]	−0.8 ± 0.1[c]	−0.9 ± 0.1[c]	−0.8 ± 0.3[b]	−0.8 ± 0.2[b]
	8	−2.8 ± 0.3[c]	−2.5 ± 0.4[c]	−2.0 ± 0.4[c]	−2.0 ± 0.4[c]	−1.4 ± 0.3[c]
vehicle[d]	–	−0.2 ± 0.1	−0.2 ± 0.1	−0.3 ± 0.1	NT	NT
5[d]	10	0.2 ± 0.1	0.2 ± 0.2	0.2 ± 0.2	NT	NT

[a] The investigated compounds were administered ip 30 min before the test. The absolute mean initial body temperatures were within a range of 36 ± 0.5 °C. [b] $p < 0.05$ vs vehicle. [c] $p < 0.01$ vs vehicle. [d] Data for comparison taken from ref 12. NT = not tested.

under the same conditions compounds **15** (4−16 mg/kg) and **16** (2−8 mg/kg) produced hypothermia in a dose-dependent manner (Table 4).

Discussion

Extension of reference 4-*n*-alkyl-1-(*o*-methoxyphenyl)-piperazines **6**−**8** with a 1- or 2-benzotriazole fragment results in a significantly higher 5-HT_{1A} affinity of **9**−**12**, **15**, and **16** (Table 1). The data presented in Table 1 indicate that the terminal benzotriazole system contributes to the observed 5-HT_2/5-HT_{1A} selectivity ratio of derivatives **9**−**12** in relation to that observed for the respective reference compounds **6** and **7**. It should also be stressed that specific isomers of the benzotriazole ring system have no effect on the 5-HT_2/5-HT_{1A} selectivity ratio, except for the pair of derivatives **15** and **16**.

It has been well-established that the length of the alkyl chain which separates the arylpiperazine portion from the terminal phthalimido moiety is of great importance for the 5-HT_{1A} affinity of some long-chain 1-arylpiperazines.[17,20,21] Elongation of the chain from two to four carbon atoms enhances the 5-HT_{1A} affinity of *N*-phthalimidoalkyl derivatives of 1-(2-methoxyphenyl)piperazine by at least 3 orders of magnitude.[20,21] By contrast, the effect of the chain length in two series of the investigated derivatives either is negligible (cf. **9**, **11**, and **15**) or does not exceed factor 10 (cf. **10**, **12**, and **16**, Table 1). Our findings are coherent with the results of El-Bermawy et al.,[17] who reported that the excision of the terminal amide function in compound **3** and its analogs reflects an almost negligible effect of the chain length on the 5-HT_{1A} affinity.

The results of our in vivo experiments clearly suggest that the investigated compounds show a different intrinsic activity at 5-HT_{1A} receptors. Our results indicate that compound **11** behaves like a functional antagonist at both pre- and postsynaptic 5-HT_{1A} receptors, whereas compound **16** appears to be a weak, partial agonist of 5-HT_{1A} receptors in the used in vivo tests.

A high potency of **11** at pre- and postsynaptic 5-HT_{1A} sites was demonstrated in several behavioral models. It was found previously that the 8-OH-DPAT-induced flat body posture and forepaw treading in reserpinized rats, as well as the lower lip retraction in rats were mediated by postsynaptic 5-HT_{1A} receptors.[22−24] However, the 8-OH-DPAT-induced hypothermia in mice was mediated by presynaptic 5-HT_{1A} receptors.[25−27] All the responses induced by 8-OH-DPAT in the above tests were completely blocked by **11** (Tables 2 and 3). Moreover, **11** did not produce any effect which would be characteristic of its agonistic properties at 5-HT_{1A} receptors. This means that **11** given alone did not induce the 5-HT behavioral syndrome, nor did it evoke the lower lip retraction in rats (Table 2) or hypothermia in mice (Table 4). A complete blocking effect of **11** was observed at doses of 4−8 mg/kg; hence its activity was essentially the same as that of the most prominent 5-HT_{1A} antagonist (*S*)-WAY-100135 (**5**).[10] In fact, compound **5** behaved like a functional antagonist at both pre- and postsynaptic 5-HT_{1A} sites.[9−12,14] A fully blocking effect of **5** on the 8-OH-DPAT-induced syndrome, lower lip retraction in rats and hypothermia in mice, as well as other responses mediated by 5-HT_{1A} receptors were observed after administration of doses of 3−10 mg/kg (cf. also Tables 2−4).[10−12] Moreover, it should be added that the observed similarities between **11** and **5** in the discussed behavioral models are justified, if one considers—at the first stage of approximation—their affinity for 5-HT_{1A} sites and the 5-HT_2/5-HT_{1A} selectivity ratio. Both compounds are highly potent 5-HT_{1A} ligands, with a similar 5-HT_2/5-HT_{1A} selectivity [cf. K_i = 15 and 1040 nM for **11** (Table 1) vs IC_{50} = 15.5 ± 4.6 and 1393 ± 400 nM for **5**[9] at 5-HT_{1A} and 5-HT_2 sites, respectively]. The preliminary studies on the receptor binding profile indicated that compound **11** showed significant α_1 adrenergic (K_i = 69 ± 11 nM) and low dopamine D_1 and D_2 receptors affinity (K_i = 2880 ± 140 and 696 ± 62 nM, respectively).[28] However, the observed α_1/5-HT_{1A} = 4.6 selectivity ratio for compound **11** is relatively low, the presented in vivo effects are fully comparable to **5**, which is a known selective, full 5-HT_{1A} antagonist.

Although compound **16** exhibits an even higher 5-HT_{1A} affinity than its structural analog **11**, as well

as a similar 5-HT_2/5-HT_{1A} selectivity ratio (Table 1), the results of the in vivo experiments indicate that **16** can be classified as a partial agonist of the 5-HT_{1A} receptors. Like 8-OH-DPAT, compound **16** induces lower lip retraction in rats (Table 2) and hypothermia in mice (Table 4). In contrast to 8-OH-DPAT, compound **16** does not produce flat body posture or forepaw treading in rats. Moreover, it inhibits the behavioral syndrome induced by 8-OH-DPAT in reserpinized rats (Table 2). A similar profile in those tests was earlier reported for ipsapirone, which is known as a typical partial agonist of 5-HT_{1A} receptors.[23,29,30]

Surprisingly, compounds **12** and **15**, which are isomeric counterparts of **11** and **16**, respectively, are practically inactive in the tests used, except of **10** which showed similar hypothermic effect to compound **11** (Tables 2–4); however, they show an essentially identical, high 5-HT_{1A} affinity (Table 1).

Conclusions

The results presented above indicate that the terminal benzotriazole ring system stabilizes effectively the 5-HT_{1A} receptor–ligand complex. The most striking findings of our study are that the structurally close derivatives **11**, **12**, **15**, and **16** show a different intrinsic activity at 5-HT_{1A} receptors and that compound **11** is the only one which, in the behavioral tests used, behaves like a potent pre- and postsynaptic antagonist of 5-HT_{1A} receptors. This phenomenon has not been clearly explained, as yet. Moreover, at the present stage of knowledge, there are no sufficient data available on the structure of full (pre- and postsynaptic) 5-HT_{1A} antagonists, as only two of such compounds, i.e., **5** and (*S*)-5-fluoro-8-hydroxy-2-(di-*n*-propylamino)tetralin, have been found to date. Therefore, any conclusions on the structure–activity relationships are only speculative. However, the reported stereoselectivity of **5**[8,9] permits us to assume that the steric relations between the protonation center, 1-(2-methoxyphenyl) substituent, and the phenyl ring attached to the ethyl chain, which are characteristic of the active isomer *S*, may be responsible for its 5-HT_{1A} antagonism.

In conclusion, it may be inferred that the 5-HT_{1A} antagonism of **11** and its receptor selectivity are still subject to confirmation by some more detailed investigations. Further behavioral, electrophysiological, and biochemical studies are presently in progress.

Experimental Section

Chemistry. Melting points were determined on a Boetius apparatus and are uncorrected. Electron impact mass spectra (70 eV) were taken with an LKB 2091 instrument. ^{1}H NMR spectra were obtained on a Varian EM-360L (60 MHz) spectrometer in $CDCl_3$ solution with Me_4Si as an internal standard. Elemental analyses were performed in the Institute of Organic Chemistry PAN (Warsaw, Poland) and were within ±0.3% of the theoretical values.

Benzotriazole and 1-(*o*-methoxyphenyl)piperazine were commercial products (Aldrich). The catalyst (KF/Al_2O_3) was prepared according to Yamawaki et al.[31] TLC was performed on silica gel plates (Kieselgel 60F_{254}, Merck).

General Procedure A. Preparation of Compounds 6–8. A mixture of the appropriate 1-arylpiperazine (3 mmol), anhydrous K_2CO_3 (1.2 g), alkyl bromide (4 mmol),and acetone (20 mL) was stirred for 24 h at room temperature. Then the inorganic salt was filtered off and acetone was evaporated to dryness. The residue was dissolved in *n*-hexane, filtered off again, and purified using a column chromatography (Al_2O_3/$CHCl_3$ for **6**, SiO_2/ethyl acetate for **7**, and SiO_2/$CHCl_3$–CH_3OH, 9:1, for **8**). Free bases were dissolved in acetone (5–7 mL), treated with an excess of diethyl ether saturated with dry, gaseous HCl, and kept in refrigerator to give colorless crystalline products.

4-Ethyl-1-(2-methoxyphenyl)piperazine (6): yield 74%; oil; ^{1}H NMR δ 1.1 (t, 3 H, CH_3, J = 7), 2.5 (m, 6 H, 3 CH_2), 3.1 (m, 4 H, 2 CH_2), 3.85 (s, 3 H, OCH_3), 6.95 (s, 4 H arom). **6·2HCl:** mp 205–207 °C (Me_2CO). Anal. ($C_{13}H_{20}N_2O$·2HCl) C, H, N.

1-(2-Methoxyphenyl)-4-*n*-propylpiperazine (7): yield 56%; oil; ^{1}H NMR δ 0.95 (t, 3 H, CH_3, J = 7), 1.6 (m, 2 H, CH_2), 2.5 (m, 6 H, 3 CH_2), 3.2 (m, 4 H, 2 CH_2), 3.9 (s, 3 H, OCH_3), 7.0 (s, 4 H arom). **7·2HCl:** mp 193–195 °C (Me_2CO). Anal. ($C_{14}H_{22}N_2O$·2HCl) C, H, N.

4-*n*-Butyl-1-(2-methoxyphenyl)piperazine (8): yield 36%; oil; ^{1}H NMR δ 1.4 (cluster, 7 H, $CH_2CH_2CH_3$), 2.5 (m, 6 H, 3 CH_2), 3.2 (m, 4 H, 2 CH_2), 3.9 (s, 3 H, OCH_3), 7.0 (s, 4 H arom), **8·2HCl·0.5H_2O:** mp 210–212 °C (Me_2CO). Anal. ($C_{15}H_{24}N_2O$·2HCl·0.5H_2O) C, H, N.

General Procedure B. Preparation of Derivatives 9–12. To the sodium butoxide solution prepared from sodium (0.23 g, 10 mmol) and *n*-butanol (50 mL) were added benzotriazole (1.19 g, 10 mmol) and the appropriate 1-(ω-bromoalkyl)-4-arylpiperazine (5 mmol). The reaction mixture was refluxed for 1 h and left overnight at room temperature. Then the solvent was evaporated and the residue was treated with benzene (100 mL) and 20% K_2CO_3 (10 mL). The organic layer was separated, washed with water, and dried over anhydrous K_2CO_3. The solvents were evaporated, and the mixture of 1- and 2-benzotriazole isomers was separated using a silica gel chromatography (Chromatotron) and ethyl acetate–*n*-hexane (1:1) as an eluent. Hydrochloride salts were prepared according to the general procedure A.

4-[2-(Benzotriazol-1-yl)ethyl]-1-(2-methoxyphenyl)piperazine (9): yield 18%; oil; R_f 0.23 (EtOAc–*n*-hexane, 1:1); ^{1}H NMR δ 3.0 (cluster, 10 H, 5 CH_2), 3.8 (s, 3 H, OCH_3), 4.85 (t, 2 H, CH_2Bt, J = 7), 6.9 (s, 4 H arom), 7.5 (m, 3 H, Bt H-5,6,7), 8.1 (m, 1 H, Bt H-4); MS m/e 337 (13, M^+). **9·2HCl·H_2O:** mp 205–207 °C (Me_2CO). Anal. ($C_{19}H_{23}N_5O$·2HCl·H_2O) C, H, N.

4-[2-(Benzotriazol-2-yl)ethyl]-1-(2-methoxyphenyl)piperazine (10): yield 39%; mp 109–110 °C (EtOAc–*n*-hexane, 1:1); R_f 0.38 (EtOAc–*n*-hexane, 1:1); ^{1}H NMR δ 2.7 (m, 4 H, 2 CH_2), 3.1 (cluster, 6 H, 3 CH_2), 3.75 (s, 3 H, OCH_3), 4.95 (t, 2 H, CH_2Bt, J = 7), 6.8 (s, 4 H arom), 7.3 (m, 2 H, Bt H-5,6), 7.9 (m, 2 H, Bt H-4,7); MS m/e 337 (21, M^+). **10·2HCl:** mp 219–220 °C (Me_2CO–Et_2O, 4:1). Anal. ($C_{19}H_{23}N_5O$·2HCl) C, H, N.

4-[3-(Benzotriazol-1-yl)propyl]-1-(2-methoxyphenyl)piperazine (11): yield 61%; mp 82–84 °C (EtOAc–*n*-hexane, 1:1); R_f 0.40 (EtOAc); ^{1}H NMR δ 2.3 (cluster, 8 H, 4 CH_2), 3.0 (m, 4 H, 2 CH_2), 3.8 (s, 3 H, OCH_3), 4.7 (m, 2 H, CH_2Bt), 6.9 (s, 4 H arom), 7.4 (m, 3 H, Bt H-5,6,7), 8.0 (m, 1 H, Bt H-4); MS m/e 351 (29, M^+). **11·2HCl·2H_2O:** mp 184–186 °C (Me_2CO). Anal. ($C_{20}H_{25}N_5O$·2HCl·2H_2O) C, H, N.

4-[3-(Benzotriazol-2-yl)propyl]-1-(2-methoxyphenyl)piperazine (12): yield 18%; mp 63–65 °C (EtOAc–*n*-hexane, 1:1); R_f 0.53 (EtOAc); ^{1}H NMR δ 2.4 (cluster, 8 H, 4 CH_2), 2.95 (m, 4 H, 2 CH_2), 3.75 (s, 3 H, OCH_3), 4.8 (m, 2 H, CH_2Bt), 6.8 (s, 4 H arom), 7.2 (m, 2 H, Bt H-5,6), 7.75 (m, 2 H, Bt H-4,7); MS m/e 351 (77, M^+). **12·2HCl:** mp 153–155 °C (Me_2CO). Anal. ($C_{20}H_{25}N_5O$·2HCl) C, H, N.

Preparation of Derivatives 13 and 14. A mixture of benzotriazole (1.19 g, 10 mmol), 1,4-dibromobutane (6.5 g, 30 mmol), KF/Al_2O_3 catalyst (10 g), potassium iodide (0.1 g), and acetonitrile (100 mL) was refluxed for 1 h and left overnight at room temperature. Then the inorganic precipitate was filtered off, the solvent was evaporated, and the mixture of isomers was separated using column chromatography (SiO_2/$CHCl_3$–*n*-hexane, 1:1, and *n*-hexane).

1-(4-Bromobutyl)benzotriazole (13): yield 35%; oil; R_f 0.69 (EtOAc–*n*-hexane, 1:1); ^{1}H NMR δ 2.2 (m, 4 H, CH_2CH_2), 3.4 (t, 2 H, CH_2Br, J = 6), 4.7 (t, 2 H, CH_2Bt, J = 6), 7.7 (m, 3 H, Bt H-5,6,7), 8.05 (m, 1 H, Bt H-4).

2-(4-Bromobutyl)benzotriazole (14): yield 22%; oil; R_f 0.87 (EtOAc–*n*-hexane, 1:1); ^{1}H NMR δ 2.05 (m, 4 H, CH_2-

CH_2), 3.45 (t, 2 H, CH_2Br, J = 6), 4.8 (t, 2 H, CH_2Bt, J = 6), 7.35 (m, 2 H, Bt H-5,6), 8.0 (m, 2 H, Bt H-4,7).

General Procedure C. Preparation of Compounds 15 and 16. A mixture of **13** or **14** (1.45 g, 6 mmol), *o*-(methoxyphenyl)piperazine (1.12 g, 6 mmol), anhydrous K_2CO_3 (1.6 g), and *n*-butanol (50 mL) was refluxed for 3 h and left overnight at room temperature. Then the inorganic salts were filtered off, the solvent was evaporated, and the oily residue was purified using a column chromatography SiO_2/$CHCl_3$–methanol, 49:1). Hydrochloride salts were prepared according to the general procedure A.

4-[4-(Benzotriazol-1-yl)butyl]-1-(2-methoxyphenyl)piperazine (15): yield 38%; oil; R_f 0.27 ($CHCl_3$–MeOH, 49:1); ^{1}H NMR δ 2.0 (cluster, 10 H, 5 CH_2), 3.0 (m, 4 H, 2 CH_2), 3.85 (s, 3 H, OCH_3), 4.7 (t, 2 H, CH_2Bt, J = 7), 6.95 (s, 4 H arom), 7.45 (m, 3 H, Bt H-5,6,7), 8.05 (m, 1 H, Bt H-4); MS m/e 365 (46, M^+). **15·2HCl:** mp 178–181 °C (Me_2CO–MeOH, 4:1). Anal. ($C_{21}H_{27}N_5O$·2HCl) C, H, N.

4-[4-(Benzotriazol-2-yl)butyl]-1-(2-methoxyphenyl)piperazine (16): yield 41%; oil; R_f 0.32 (EtOAc); ^{1}H NMR δ 2.0 (cluster, 10 H, 5 CH_2), 3.05 (m, 4H, 2 CH_2), 3.85 (s, 3 H, OCH_3), 4.8 (t, 2 H, CH_2Bt, J = 7), 6.95 (s, 4 H arom), 7.4 (m, 2 H, Bt H-5,6), 7.85 (m, 2 H, Bt H-4,7); MS m/e 365 (21, M^+). **16·2HCl:** mp 201–203 °C (Me_2CO–MeOH, 4:1). Anal. ($C_{21}H_{27}N_5O$·2HCl) C, H, N.

Radioligand Binding Experiments. Radioligand binding studies were performed in the rat brain using the following structures: hippocampus (5-HT_{1A}) and cortex (5-HT_2) according to the published procedures.[32,33] Radioligands used were [^{3}H]-8-OH-DPAT (190 Ci/mmol, Amersham) and [^{3}H]ketanserin (60 Ci/mmol, NEN Chemicals) for 5-HT_{1A} and 5-HT_2 receptors, respectively. K_i values were determined from at least three competition binding experiments in which 10–14 drug concentrations run in triplicates were used.

In Vivo Experiments. The experiments were performed on male Wistar rats (220–260 g) or male Albino-Swiss mice (20–25 g). The animals were kept at room temperature (20 ± 1 °C) on a natural day–night cycle (September–December) and were housed under standard laboratory conditions. They had free access to food (Bacutil pellets) and tap water before the experiment. Each experimental group consisted of six to eight animals *per* dose, and all animals were used only once.

8-Hydroxy-2-(di-*n*-propylamino)tetralin (8-OH-DPAT, Research Biochemicals Inc.), reserpine (Ciba), and the investigated dihydrochloride salts of **11**, **12**, **15**, and **16** were used in the form of freshly prepared aqueous solutions.

The Behavioral Syndrome Induced by 8-OH-DPAT in Reserpinized Rats. The rats were individually placed in cages 5 min before injection of 8-OH-DPAT (5 mg/kg ip). Observation sessions, lasting 45 s each, began 3 min after 8-OH-DPAT injection and were repeated every 3 min. Reciprocal forepaw treading and flat body posture were scored using a ranked intensity scale, where 0 = absent, 1 = equivocal, 2 = present, and 3 = intense. The maximum score, summed up over five observation periods, amounted to 15 for each symptom/animal.[22] Reserpine (1.0 mg/kg sc) and **11**, **12**, **15**, or **16** (ip) were administered at 18 h and 30 min before 8-OH-DPAT, respectively. The effect of **11**, **12**, **15**, or **16** given alone on the behavior of normal rats was estimated in an independent experiment, and observations began 3 min after treatment and were repeated every 3 min for a period of 1 h.

The Lower Lip Retraction (LLR) Induced by 8-OH-DPAT in Rats. The LLR was assessed according to the method described by Berendsen at al.[23] The rats were individually placed in cages, having been scored three times (at 15, 30, and 45 min after administration of 8-OH-DPAT) as follows: 0 = lower incisors not visible, 0.5 = partly visible, 1 = completely visible. The maximum score, summed, amounted to 3 for each rat. Compounds **11**, **12**, **15**, or **16** were administered ip 30 min before 8-OH-DPAT (1 mg/kg, ip). The induction of LLR by **11**, **12**, **15**, or **16** given alone was tested in a separate experiment, and animals were scored 45, 60, and 75 min after the treatment.

The Hypothermia Induced by 8-OH-DPAT in Mice. The rectal body temperature in mice (measured with an Ellab thermometer) was recorded at 15, 30, 45, and 60 min after the injection of 8-OH-DPAT (5.0 mg/kg ip). Compounds **11**, **12**, **15**, or **16** were administered 30 min before 8-OH-DPAT. The effects of **11**, **12**, **15**, or **16** given alone on the rectal body temperature were measured at 30, 45, 60, 75, and 90 min after the treatment in an independent experiment. The results are expressed as a change in the body temperature (Δt) with respect to the basal body temperature, as measured at the beginning of the experiment.

Statistics. The obtained data were analyzed by Dunnett's test.

Acknowledgment. We are grateful to the Polish State Committee for Scientific Research (Grant No. KBN 4-1449-91-01, 1991–1994) for financial support of this research.

References

(1) Previous paper: Mokrosz, J. L.; Chojnacka-Wójcik, E.; Dereń-Wesołek, A.; Kłodzińska, A.; Maćkowiak, M.; Bielecka, Z.; Paluchowska M. H. Structure-activity relationship studies of CNS agents. XII. 1-[4-(4-Aryl-1-piperazinyl)butyl]-3,4-dihydro-2(1H)-quinolinones: New 5-HT_{1A}, 5-HT_2 and D_2 ligands with a potential antipsychotic activity. *Drug Des. Disovery*, in press.

(2) Glennon, R. A.; Westkaemper, R. B.; Bartyzel, P. In *Serotonin Receptor Subtypes: Basic and Clinical Aspects*; Peroutka, S. J., Ed.; Wiley-Liss: New York, 1991; pp 19–64.

(3) Glennon, R. A.; Dukat, M. Serotonin receptors and their ligands: A lack of selective agents. *Pharmacol. Biochem. Behav.* **1991**, *40*, 1009–1017.

(4) Nelson, D. L. Structure-activity relationships at 5-HT_{1A} receptors: Binding profiles and intrinsic activity. *Pharmacol. Biochem. Behav.* **1991**, *40*, 1041–1051.

(5) Glennon, R. A. Concepts for the design of 5-HT_{1A} serotonin agonists and antagonists. *Drug Dev. Res.* **1992**, *26*, 251–274.

(6) Glennon, R. A.; Dukat M. 5-HT Receptor ligands – update 1992. *Curr. Drugs: Serotonin* **1992**, 1–45.

(7) Cliffe, I. A.; Fletcher, A.; Dourish, C. T. The evolution of silent selective 5-HT_{1A} receptor antagonists. *Curr. Drugs: Serotonin* **1992**, 99–124.

(8) Fletcher, A.; Cliffe, I. A.; Dourish, C. T. Silent 5-HT_{1A} receptor antagonists. *Trends Pharmacol. Sci.* **1993**, *14*, 441–448.

(9) Cliffe, I. A.; Brightwell, C. I.; Fletcher, A.; Forster, E. A.; Mansell, H. L.; Reilly, Y.; Routledge, C.; White, A. C. *(S)-N*-tert.-Butyl-3-(4-(2-methoxyphenyl)piperazin-1-yl)-2-phenylpropanamide [*(S)*-WAY-100135]: A selective antagonist at presynaptic and postsynaptic 5-HT_{1A} receptors. *J. Med. Chem.* **1993**, *36*, 1509–1510.

(10) Fletcher, A.; Bill, D. J.; Bill, S. J.; Cliffe, I. A.; Dover, G. M.; Forster, E. A.; Haskins, J. T.; Jones, D.; Mansell, H. L. Reilly, Y. WAY 100135 – A novel, selective antagonist at presynaptic and postsynaptic 5-HT(1A) receptors. *Eur. J. Pharmacol.* **1993**, *237*, 283–292.

(11) Lejeune, F.; Rivet, J. M.; Gobert, A.; Canton, H.; Millan, M. J. WAY 100,135 and (−)-tertatolol act as antagonist at both 5-HT_{1A} autoreceptors and postsynaptic 5-HT_{1A} receptors in vivo. *Eur. J. Pharmacol.* **1993**, *240*, 307–310.

(12) Przegaliński, E.; Filip, M.; Budziszewska, B.; Chojnacka-Wójcik, E. Antagonism of (+)-WAY 100135 to behavioural, hypothermic and corticosterone effects induced by 8-OH-DPAT. *Pol. J. Pharmacol.* **1994**, *46*, 21–27.

(13) Routledge, C.; Gurling, J.; Forster, E. A.; Wright, I.; Fletcher, A.; Dourish, C. T. Antagonism of presynaptic and postsynaptic 5-HT_{1A} receptors in vivo by the selective 5-HT_{1A} receptor antagonist, WAY 100135. *Br. J. Pharmacol.* **1992**, *107*, 5P.

(14) Routledge, C.; Gurling, J.; Wright, I.; Dourish, C. T. Neurochemical profile of the selective 5-HT_{1A} receptor antagonist: WAY 100135 an in vivo microdialysis study. *Eur. J. Pharmacol.* **1993**, *239*, 195–202.

(15) Bjork, L.; Cornfield, L. J.; Nelson, D. L.; Hillver, S. E.; Anden, N. E.; Lewander, T.; Hacksell, U. Pharmacology of the novel 5-hydroxytryptamine$_{1A}$ receptor antagonist *(S)*-5-fluoro-8-hydroxy-2-(dipropyl-amino)tetralin: Inhibition of *(R)*-8-hydroxy-2-(dipropylamino)tetralin-induced effects. *J. Pharmacol. Exp. Ther.* **1991**, *258*, 58–65.

(16) Hillver, S. E.; Bjork, L.; Li, Y. L.; Svensson, B.; Ross, R.; Anden, N. E.; Hacksell, U. *(S)*-5-Fluoro-8-hydroxy-2-(dipropylamino)-tetralin: A putative 5-HT_{1A} receptor antagonist. *J. Med. Chem.* **1990**, *33*, 1541–1544.

(17) El-Bermawy, M.; Raghupathi, R.; Ingher, S. P.; Teitler, M.; Maayani, S.; Glennon, R. A. 4-[4-(1-Noradamantanecarboxamido)butyl]-1-(2-methoxyphenyl)piperazine: A high-affinity 5-HT_{1A}-selective agent. *Med. Chem. Res.* **1992**, *2*, 88–95.

(18) Mokrosz J. L.; Duszyńska, B. Structure-activity relationship studies of CNS agents. Part 7. The effect of the imidazole fragment in 2-substituted 1-[3-(4-aryl-1-piperazinyl)propyl]imidazoles on their interaction modes with 5-HT_{1A} and 5-HT_2 receptors. *Pol. J. Pharmacol. Pharm.* **1992**, *44*, 527–538.

(19) Katritzky, A. R.; Rachwal, S.; Rachwal, B. The chemistry of benzotriazole. Part 3. The aminoalkylation of benzotriazole. *J. Chem Soc,. Perkin Trans. 2*, **1987**, 799–804.
(20) Glennon, R. A.; Naiman, N. A.; Pierson, M. E.; Smith, J. D.; Ismaiel, A. M.; Titeler, M.; Lyon, R. A. N-(Phthalimidialkyl) derivatives of serotonergic agents: A common interaction at 5-HT_{1A} serotonin binding sites? *J. Med. Chem.* **1989**, *32*, 1921–1926.
(21) El-Bermawy, M. A.; Lotter, H.; Glennon, R. A. Comparative molecular field analysis of the binding of arylpiperazines at 5-HT_{1A} serotonin receptors. *Med. Chem. Res.* **1992**, *2*, 290–297.
(22) Tricklebank M. D.; Forler, C.; Fozard, J. R. The involvement of subtypes of the 5-HT_1 receptor and catecholaminergic system in the behavioural response to 8-hydroxy-2-(di-*n*-propylamino)-tetralin in the rat. *Eur. J. Pharmacol.* **1985**, *106*, 271–282.
(23) Berendsen, H. H. G.; Jenck, F.; Broekkamp, C. L. E. Selective activation of 5-HT_{1A} receptor induces lower lip retraction in the rat. *Pharmacol. Biochem. Behav.* **1989**, *33*, 821–827.
(24) Berendsen, H. H. G.; Broekkamp, C. L. E.; Van Delft, A. M. L. Antagonism of 8-OH-DPAT-induced behaviour in rats. *Eur. J. Pharmacol.* **1990**, *187*, 97–103.
(25) Goodwin, G. M.; De Souza, R. J.; Green, A. R. The pharmacology of the hypothermic response in mice to 8-hydroxy-2-(di-*n*-propylamino)tetralin (8-OH-DPAT): A model of presynaptic 5-HT_1 function. *Neuropharmacology* **1985**, *24*, 1187–1194.
(26) Martin, K. F.; Heal, D. J. 8-OH-DPAT-induced hypothermia in rodents: A specific model of 5-HT_{1A} autoreceptor function. In *Serotonin: Molecular Biology, Receptors and Functional Effects*; Fozard, J. R., Saxena, P. R., Eds.; Birkhauser Verlag: Basel, 1991; pp 483–490.
(27) Martin, K. F.; Phillips, I.; Hearson, M.; Prow, M. R.; Heal, D. J. Characterization of 8-OH-DPAT-induced hypothermia in mice as a 5-HT_{1A} autoreceptor response and its evaluation as a model to selectivity identify antidepressants. *Br. J. Pharmacol.* **1992**, *107*, 15–21.
(28) Przegaliński, E.; Filip, M. Unpublished results.
(29) Maj, J.; Chojnacka-Wójcik, E.; Tatarczyńska, E.; Kłodzińska, A. Central action of ipsapirone, a new anxiolytic drug, on serotoninergic, noradrenergic and dopaminergic functions. *J. Neural Transm.* **1987**, *70*, 1–17.
(30) Smith, J. M.; Peroutka, S. J. Differential effects of 5-hydroxytryptamine$_{1A}$ selective drugs on the 5-HT behavioral syndrome. *Pharmacol. Biochem. Behav.* **1986**, *24*, 1513–1519.
(31) Yamawaki, J.; Ando, T.; Hanafusa, T. *N*-Alkylation of amides and *N*-heterocycles with potassium fluoride on alumina. *Chem. Lett.* **1981**, 1143–1145.
(32) Middlemiss, D. N.; Fozard, J. R. 8-Hydroxy-2-(di-*n*-propylamino)-tetralin discriminates between subtypes of the 5-HT_1 recognition site. *Eur. J. Pharmacol.* **1983**, *90*, 151–153.
(33) Bojarski, A. J.; Cegła, M. T.; Charakchieva-Minol, S.; Mokrosz, M. J.; Maćkowiak, M,; Misztal, S.; Mokrosz, J. L. Structure-activity relationship studies of CNS agents. Part 9. 5-HT_{1A} and 5-HT_2 receptor affinity of some 2- and 3-substituted 1,2,3,4-tetrahydro-β-carbolines. *Pharmazie* **1993**, *48*, 289–294.