Novel Benzothiazolin-2-one and Benzoxazin-3-one Arylpiperazine Derivatives with Mixed $5HT_{1A}/D_2$ Affinity as Potential Atypical Antipsychotics

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Since it was known that 5HT properties ($5HT_{1A}$ agonism or $5HT_{2A}$ antagonism) combined with D_2 antagonism may lead to atypical antipsychotic drugs, a series of 19 benzothiazolin-2-one and benzoxazin-3-one derivatives possessing the arylpiperazine moiety was prepared, and their binding profiles were investigated. All tested compounds displayed very high affinities for the $5HT_{1A}$ and D_2 receptors. Therefore, further pharmacological studies were carried out on selected compounds (**24**, **27**, **30**, **46**, and **47**). This evaluation in rats clearly revealed potent antipsychotic properties along with a decrease of extrapyramidal side effects. These derivatives are currently under preclinical development.

Introduction

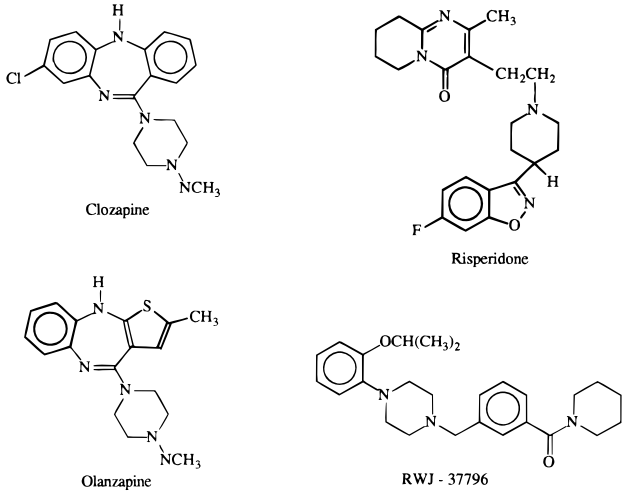
Schizophrenia is a group of illnesses that occurs in approximately 1% of the adult population. The schizophrenia is characterized by a spectrum of symptoms that typically include disordered thought, social withdrawal, and hallucinations. So far, there is no known cure, and the disease is chronic and generally progressive. However, the introduction of the neuroleptic phenothiazine chlorpromazine in 1952 initiated the area of pharmacotherapy in psychiatric medicine and has led to the marketing of dozens of clinically diverse antipsychotic drugs, belonging to different chemical families (i.e. thioxanthenes, butyrophenones, diphenylbutylpiperidines, etc.).[1]

Nevertheless, currently available antipsychotics drugs have several limitations. First, they are ineffective in many patients (neuroleptic nonresponders). Second, even when patients do respond, some aspects of psychopathology may benefit more than others: in many patients, positive symptoms may be alleviated while negative or deficit remain unresponsive.[2] Negative symptoms include alogia, flattened affect, anhedonia, asociability, and attentional impairment.[3,4] Third, classical antipsychotic drugs exhibit unwanted extrapyramidal side effects (EPS) and tardive dyskinesia.[5]

During the past few years, a second generation of antipsychotic agents has emerged; the term "atypical neuroleptic" originates from the clinical observation that the dibenzazepine clozapine exhibits antipsychotic activity in the absence of debilitating, short (e.g. dystonia) or long term (e.g. tardive dyskinesia) extrapyramidal side effects.[6]

Clozapine (Scheme 1) generally considered as the reference drug is characterized by a dual dopaminergic and serotoninergic mechanism of action:[7] a relatively weak dopamine D_2 receptor antagonism in vitro and in vivo,[8] but potentially important activities at other dopaminergic (D_1, D_4) receptors, at serotoninergic ($5HT_{2A}$, $5HT_3$, $5HT_{2C}$), adrenergic (α_1, α_2), histaminergic (H_1), and muscarinic receptors.[9−12]

Scheme 1



However, treatment with clozapine is associated with an increased risk of agranulocytosis,[13] which strongly limits its therapeutic use.

This finding with clozapine led to a major search for other pharmacological agents with mixed serotoninergic and dopaminergic activities such as risperidone[14] (Scheme 1), olanzapine,[15] or mixed $5HT_{1A}$ agonists/D_2 antagonists such as RWJ-37796[16] (Scheme 1).

Previous works in our laboratory[17−20] have established the bioisosteric equivalency of benzoxazolin-2-one and pyrocatechol. This concept led to the synthesis of amino ketones, amino alcohols, and amine derivatives of 6-(2-aminoethyl)benzoxazolin-2-one which exhibited high affinity for adrenergic and dopaminergic receptors. The expected advantages resulting from this bioisosteric

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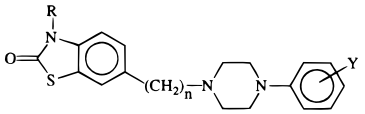
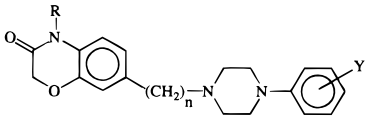
Scheme 2

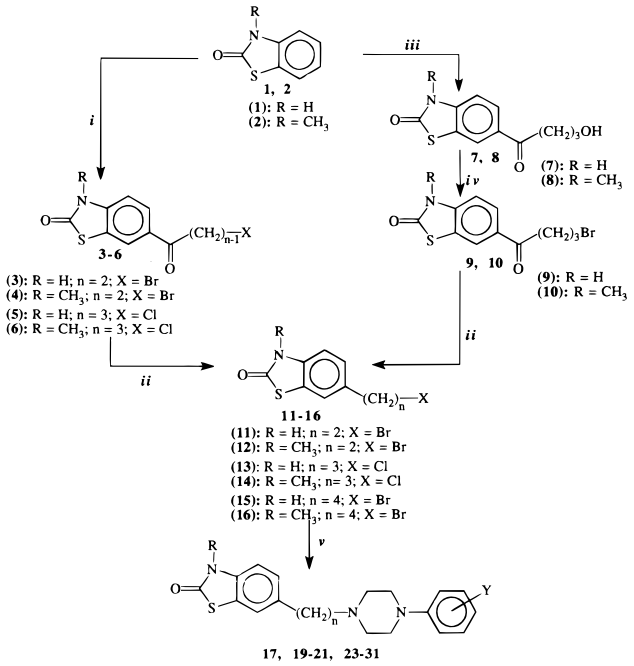


Benzothiazolin-2-one derivatives
Structure A



Benzoxazin-3-one derivatives
Structure B

Scheme 3[a]



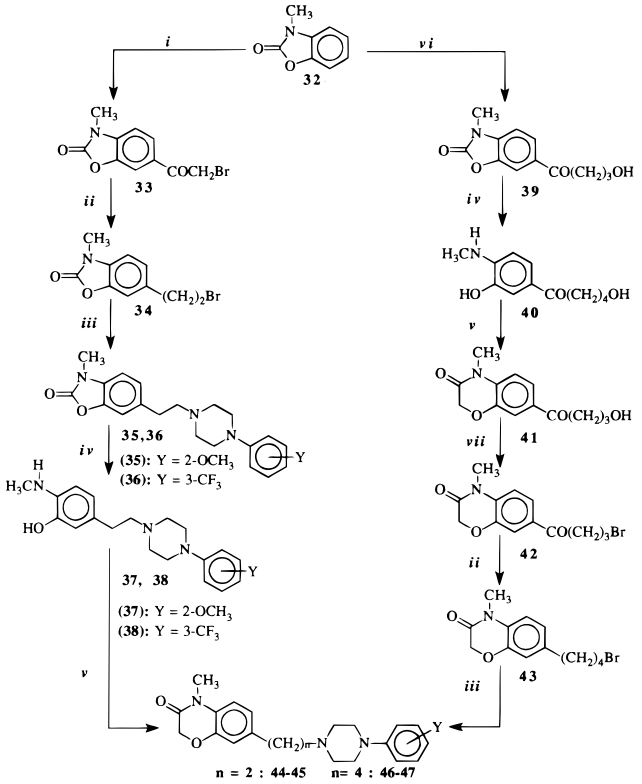
[a] (i) $AlCl_3$/DMF; $BrCH_2COCl$ or $Cl(CH_2)_2COCl$; (ii) $(C_2H_5)_3SiH$, CF_3CO_2H; (iii) PPA, $Cl(CH_2)_3COCl$; (iv) acetone, HBr; (v) arylpiperazine, acetone, $(C_2H_5)_3N$.

replacement were increases of the chemical stability and duration of action. On the basis of the well-established advantages of mixed $5HT_{1A}/D_2$ ligands in the search for atypical antipsychotics, we therefore combined the intrinsic D_2 affinity of these compounds with the phenylpiperazine pharmacophore, known as one of the most potent $5HT_{1A}$ pharmacophores.[21−23] These arylpiperazine derivatives showed important psychotropic and analgesic properties involving interaction at central serotonin (5HT) and dopamine (D_2) receptors. To increase the CNS activity and the metabolic stability, we therefore decide to replace the benzoxazolinone heterocycle with its sulfur bioisostere and with its benzoxazin-3-one homologue (structures A and B in Scheme 2). This article reports their synthesis and pharmacological evaluation as mixed $5HT_{1A}/D_2$ receptor ligands and atypical antipsychotic agents.

Chemistry

Scheme 3 illustrates the procedures used to synthesize benzothiazolin-2-one derivatives **17**, **19**−**21**, and **23**−**31**. From benzothiazolin-2-one (**1**) or 3-methylbenzothiazolin-2-one (**2**), 6-halogenoacyl **3**−**6**, and **9**−**10** and

Scheme 4[a]



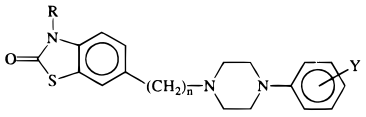
[a] (i) $AlCl_3$/DMF, $BrCH_2COCl$; (ii) $(C_2H_5)_3SiH$, CF_3CO_2H; (iii) arylpiperazine, acetone, $(C_2H_5)_3N$; (iv) NaOH, methanol; (v) ethyl bromoacetate, sodium ethanolate DMSO; (vi) $Cl(CH_2)_3COCl$, PPA; (vii) acetone, HBr.

6-halogenoalkyl **11**−**16** derivatives were obtained as previously described.[24−26] Compounds **11**−**16** were then reacted with the desired arylpiperazines in dry acetone in the presence of triethylamine to give the arylethylamino (**17, 19**−**21,** and **23**−**25**), the arylpropylamino (**26** and **27**), and the arylbutylamino (**28**−**31**) compounds. Phenolic compounds **18** and **22** were obtained by acidic cleavage of the corresponding methoxy ethers **19** and **24**. 4-Methylbenzoxazin-3-one derivatives **44** and **45** were obtained as shown in Scheme 4. Compounds **33** and **34** were synthesized by previously described procedures.[19,20] Compounds **35** and **36** were obtained by using the same conditions which were employed for the preparation of compounds **17**, **19**−**21**, and **23**−**25**. The heterocycle cleavage was carried out in aqueous medium in the presence of NaOH and led to *o*-aminophenols **37** and **38**. A one-pot reaction using sodium ethanolate, dimethyl sulfoxide, and ethyl bromoacetate gave the final compounds **44** and **45**. The synthetic approach employed for the preparation of the butylamino homologues is summarized in Scheme 4. The bromo ketone **42** was obtained from 3-methylbenzoxazolinone **32** according a previously described procedure.[27] Reduction of the ketone carbonyl group of compound **42** was carried out with the triethylsilane−trifluoroacetic acid reagent.[28] Condensation reaction with arylpiperazines was performed in dry acetone in the presence of triethylamine to give compounds **46** and **47**.

Pharmacological Results and Discussion

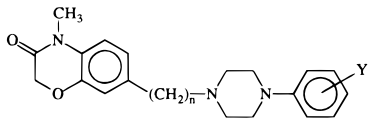
Affinities (IC_{50}'s, nM) for $5HT_{1A}$, $5HT_{1B}$, $5HT_{2A}$, $5HT_{2C}$, D_2, and α_1 receptors were determined for com-

Table 1. Structures and Binding Assays Results ($IC_{50} \pm$ SEM in nM) of Compounds of General Structure **A**



compd	*n*	R	Y	5-HT_{1A}	5-HT_{1B}	5-HT_{2A}	5-HT_{2C}	D_2	α_1
17	2	H	H	3 ± 0.06	300 ± 25	40 ± 15	90 ± 10	100 ± 15	
18	2	H	2-OH	0. 7 ±0.09	260 ± 50	530 ± 200	520 ± 110	27 ± 6	
19	2	H	2-OCH_3	0.3 ± 0.09	20 ± 4	300 ± 50	300 ± 25	10 ± 4	0.6 ± 0.2
20	2	H	3-CF_3	3 ± 0.7	600 ± 180	200 ± 35	60 ± 7	60 ± 7	90 ± 10
21	2	CH_3	H	0.7 ± 0.08	200 ± 39	70 ± 7	2000 ± 300	1000 ± 120	>100
22	2	CH_3	2-OH	0.2 ± 0.03	31 ± 4	1100 ± 15	2000 ± 260	71 ± 22	
23	2	CH_3	4-F	40 ± 7	3000 ± 420	200 ± 27		1000 ± 120	30 ± 4
24	2	CH_3	2-OCH_3	2 ± 0.3	300 ± 55	500 ± 75	4000 ± 440	40 ± 9	10 ± 3
25	2	CH_3	3-CF_3	1 ± 0.1	70 ± 8	600 ± 110	>10000	200 ± 40	400 ± 60
26	3	H	2-OCH_3	11 ± 3	61 ± 5	290 ± 80	99 ± 15	6 ± 0.5	1 ± 0.3
27	3	CH_3	2-OCH_3	3 ± 0.7	360 ± 95	910 ± 170	560 ± 29	7 ± 4	6 ± 3
28	4	H	2-OCH_3	0.2 ± 0.02	200 ± 10	150 ± 25	320 ± 50	0.77 ± 0.07	0.82 ≠ 0.1
29	4	H	3-CF_3	43 + 9	400 ± 36	570 ± 20	11000 ± 700	57 ± 4	45 ± 9
30	4	CH_3	2-OCH_3	0.8 ± 0.07	300 ± 30	1000 ± 120	240 ± 10	10 ± 2	30 ± 4
31	4	CH_3	3-CF_3	2 ± 0.6	1000 ± 49	400 ± 38		50 ± 8	200 ± 22
clozapine				150 ± 40	2500 ± 290	3 ± 1	10 ± 3	140 ± 30	160 ± 25
haloperidol				1500 ± 100	>10000	30 ± 5	>10000	1.2 ± 0.1	20 ± 5

Table 2. Structures and Binding Assays Results ($IC_{50} \pm$ SEM in nM) of Compounds of General Structure **B**



compd	*n*	Y	5-HT_{1A}	5-HT_{1B}	5-HT_{2A}	5-HT_{2C}	D_2	α_1
44	2	2-OCH_3	0.2 ± 0.03	180 ± 50	600 ± 50	1200 ± 400	3 ± 1	0.5 ± 0.05
45	2	3-CF_3	0.7 ± 0.04	460 ± 135	620 ± 100	360 ± 50	150 ± 15	85 ± 5
46	4	2-OCH_3	0.8 ± 0.09		840 ± 150		5.2 ± 2	20 ± 4
47	4	3-CF_3	0.6 ± 0.07	280 ± 65	620 ± 35	800 ± 190	8 ± 3	30 ± 2

Table 3. α_1/5-HT_{1A} and D_2/5-HT_{1A} Selectivity Ratios

compd	α_1/5-HT_{1A}	D_2/5-HT_{1A}	compd	α_1/5-HT_{1A}	D_2/5-HT_{1A}
17		33	**27**	2	2.3
18		39	**28**	4.1	3.85
19	2	33	**29**	1.05	1.33
20	30	20	**30**	37.5	12.5
21		1429	**31**	100	25
22		355	**44**	2.5	15
23	0.75	25	**45**	123	214
24	5	20	**46**	251	6.25
25	400	200	**47**	50	13.3
26	0.09	0.55			

pounds of general structures A and B (Tables 1 and 2). The selectivity ratios α_1/5HT_{1A} and D_2/5HT_{1A} are reported in Table 3.

The expected affinities were associated with an agonist character at 5HT_{1A} receptors and an antagonist one at D_2 sites. The 5HT_{1A} agonism should lead to antianxiety properties comparable to those observed with buspirone and when associated with a D_2 blockade to antipsychotic properties with less extrapyramidal side effects. Thus, the combination of high and selective 5HT_{1A} affinity with D_2 affinity lends support to the "atypical" profile of the potentially antipsychotic compounds.

Structure−affinity relationships were first studied in the benzothiazolinonic series. Replacement of benzoxazolin-2-one with its sulfur analogue (i.e. compound 24) has led to the same binding profile. The obtained 5-HT_{1A} and 5-HT_{1B} affinities are quite similar. The α_1 affinity obtained with **24** is 5-fold less than that observed with its oxygene analogue, confering to the benzothiazolin-2-one derivatives a best 5-HT_{1A} versus α_1 selectivity ratio. Pharmacomodulations concerning the nitrogen substituent of the heterocyclic pivotal template, the length of the hydrocarbon chain linking this heterocycle with the arylpiperazine moiety, and the nature and place of the substitution group of the arylpiperazine moiety were studied. It was anticipated that the diverse number of synthesized compounds could lead to the selection of compounds presenting high and selective affinities for both the 5HT_{1A} and D_2 receptors and low affinity for α_1 sites. Comparison of the results obtained with the *N*-methyl derivatives (**21**, **22**, **24**, **25**, **27**, **30**, and **31**) with their nonmethylated analogues (i.e. **17**, **18**, **19**, **20**, **26**, **28**, and **29**, respectively) leads to the following observations:

(1) The two classes of compounds retain high and similar affinities at 5HT_{1A} and D_2 sites (0.1 to 1 nM at 5HT_{1A} sites and 1 to 1000 nM at D_2 receptors).

(2) The *N*-methyl derivatives generally possess lower affinities at the other 5HT receptors (i.e. 5HT_{2A} and 5HT_{2C}). This can be observed when **17** or **18** are compared with **21** or **22**.

(3) The undesirable α_1 affinity is lower in most cases with the *N*-methyl compounds as for example **19**, **28**, **29** compared with **24**, **30**, **31**. N-Methylation seems to have no or negligible influence on the D_2 affinity.

The length of the hydrocarbon linking chain bearing the arylpiperazine was modified since the high and

undesirable α_1 affinity of compounds **17**−**25** could be related to the arylethylamino structure. No significant modification concerning the α_1 affinity with compounds bearing three and four methylene units **(26**−**31)** was observed as shown when **19** and **24** are compared with **26** and **30**. Nevertheless, this result is not surprising since it is well-known that the arylpiperazine is also a good structure for α_1 affinity.[29] On the other hand, the length of the hydrocarbon side chain seems to influence D_2 affinity. Compounds with a 4-butylamino side chain (**30** and **31**) are respectively more potent at D_2 sites than their ethylamino homologues **24** and **25**. Finally, variation of the number of methylene units has no or negligible influence on $5HT_{1A}$ affinity. The nature of the substituent on the arylpiperazine pharmacophore is of importance for the $5HT_{1A}$, D_2, and α_1 affinities: no substitution leads to compounds with high $5HT_{1A}$ affinity **(17** and **21)** but with a low D_2 affinity. The (*o*-methoxyphenyl)piperazino derivatives (**19**, **28**, and **30**) exhibit better $5HT_{1A}$, D_2, and α1 affinity than their *m*-trifluoromethyl analogues (**20**, **29**, and **31**, respectively). In most cases D_2 and α_1 affinity of nonsubstituted and 3-CF_3 arylpiperazines is lower than that of the other substituted compounds. The (*o*-hydroxyphenyl)piperazines **(18** and **22)** exhibit high $5HT_{1A}$ and D_2 affinities. Replacement of the benzothiazolin-2-one heterocycle with the benzoxazin-3-one moiety (compounds **44**−**47**, Table 2) leads to high combined $5HT_{1A}$ (0.2−0.8) and D_2 (3−8) affinity, except for compound **45** (D_2 = 150).

As a general remark concerning compounds of general structure **A** and **B**, it can be stated that they possess a multireceptorial binding profile and most of them suffer from a lack of selectivity and specificity. They represent, however, precious pharmacological tools for the studies of 5HT receptors and in particular for a better knowledge of the pharmacological activities generally related to $5HT_{1A}$ receptors.

The nature of the interactions with $5HT_{1A}$, $5HT_{2A}$, D_2, and α1 receptors was determined for some of the synthesized compounds. Compounds **24, 25, 28, 30**, and **47** were tested for their ability to induce forepaw treading in rats, indicating a stimulation of the $5HT_{1A}$ receptors subtypes. None of these compounds behave in vivo in rats like $5HT_{1A}$ agonists since they were unable to induce forepaw treading at doses active in models predictive of an antipsychotic activity; in the same conditions, the 5-HT_{1A} agonist, 8-OH-DPAT, at the dose of 8 mg/kg ip, induced forepaw treading (77% of the maximal possible score, $p < 0.001$).

Nevertheless, the same compounds, i.e. **24, 25, 28, 30**, and **47**, showed clear antagonist action at $5HT_{2A}$ receptor subtype since they significantly inhibited the head-twich response induced by 5-hydroxytryptophan (5HTP) in mice. The antagonism was nearly complete at the dose of 1 mg/kg ip for compounds **24** and **28** (respectively 94% and 83% of antagonism, $p < 0.01$), at the dose of 4 mg/kg ip for compound **25** (91% of antagonism, $p < 0.01$) and at the dose of 8 mg/kg ip for compounds **30** and **47** (respectively 78% and 97% of antagonism, $p < 0.01$). Tested in the same conditions, the 5-HT_2 antagonist, cyproheptadine, at the dose of 16 mg/kg ip, produced an antagonism of 83% ($p > 0.05$).

Compounds **24**, **30**, and **47** were tested for their ability to antagonize the effects of a low dose of a direct D_2 dopaminergic agonist, apomorphine, in mice.

Compounds **24** and **47** completely blocked the stereotypies and the climbing at the dose of 1 mg/kg ip (100% of antagonism), while compound **30** was active at the dose of 8 mg/kg ip (100% of antagonism), indicating that the tested compounds behave like D2 antagonists. It must be noted that none of these compounds were able to reduce the apomorphine-induced hypothermia since they intrinsically induced hypothermia in mice; tested in the same conditions, haloperidol (0.25 mg/kg ip) completely abolished both stereotypies and climbing (100% of antagonism), and significantly reduced the apomorphine-induced hypothermia (2.2 °C, $p < 0.01$).

Some of the synthesized compounds were examined for potential antipsychotic activity. Compounds **22**, **24**, **27**, **28**, **30**, **46**, and **47** were tested for their ability to antagonize the hyperlocomotion induced by *d*-amphetamine in mice. The compounds were tested at two doses which weakly reduced the spontaneous locomotor activity. All the tested compounds significantly antagonized the d-amphetamine induced hyperlocomotion. The doses which reduced by approximately 50% the locomotion in comparison with the amphetamine treated animals were 0.25 mg/kg ip for compound **46** (46% of antagonism, $p < 0.01$), 0.50 mg/kg ip for compound **30** (54% of antagonism, $p < 0.05$), 1 mg/kg ip for compounds **24** and **27** (respectively 57% and 52% of antagonism, $p < 0.01$), 2 mg/kg ip for compound **22** (56% of antagonism, $p < 0.01$), and 4 mg/kg ip for compounds **28** and **47** (respectively 68% and 61%, $p < 0.01$). In the same experimental conditions, clozapine reduced by 48% ($p < 0.01$) and haloperidol by 67% ($p < 0.01$) the amphetamine-induced hyperactivity, at the respective doses of 4 and 0.125 mg/kg ip.

In addition, compounds **24** and **46** were also tested in rats, using the same paradigm. After oral administration, both compounds significantly ($p < 0.05$) reduced the hyperactivity by 50% at the doses of 2 and 4 mg/kg po, respectively 63% and 58% of antagonism for compounds **24** and **46**; the antagonism was complete (103% and 108%) at the respective doses of 8 and 16 mg/kg po for compounds **24** and **46** ($p < 0.01$). In comparison, haloperidol completely blocked the hyperactivity ($p < 0.001$) at the dose of 1 mg/kg po (179% of antagonism).

These behavioral tests predictive of an antipsychotic activity were based on the dopaminergic hypothesis of schizophrenia.

Some of the synthesized compounds were tested for their ability to antagonize the scratching induced by mescaline, a serotonergic compound, in mice.

Compound **18** (2 and 4 mg/kg ip), **24** (0.25 and 1 mg/kg ip), **26** (4 mg/kg ip), **27** (2 mg/kg ip), **46** (1 mg/kg ip), and **47** (2 and 8 mg/kg ip) significantly ($p < 0.05$) reduced the scratching induced by mescaline (Table 4). In comparison, at the dose of 4 mg/kg ip, the atypical antipsychotic drug clozapine completely blocked the scratching induced by mescaline, in accordance with the interaction with serotoninergic receptors.

Five compounds were tested in the tail suspension test (TST) which allows to screen different psychotropic drugs including antidepressants, anxiolytics, and neuroleptics. In this test derived from the forced swim test,

Table 4. In Vivo Antagonism of the Effects of Mescaline in Mice

	18		**24**		**26**	**27**	**46**	**47**		clozapine
dose (mg/kg ip)	2	4	0.25	1	4	2	1	2	8	4
antagonism of mescaline-induced scratching (% vs controls)[a]	71% **	97***	92***	100***	96**	73*	81*	60 ns	87**	99***

[a] $*p < 0.05$. $**p < 0.01$. $***p < 0.001$. $n = 10$ animals per group.

Table 5. Tail Suspension Test in Mice

	18	**24**	**26**	**30**	**46**	haloperidol	clozapine
dose (mg/kg ip)	4	1	4	8	1	0.5	4
duration of immobility (% vs controls)[a]	+92**	+89***	+99**	+36*	+124**	+65***	+90***

[a] $*p < 0.05$. $**p < 0.01$. $***p < 0.001$. $n = 10$ animals per group.

Table 6. In Vivo Antagonism of the Effects of SKF 10047 and of PCP by Compound **24** in Rats

	24			haloperidol	
dose (mg/kg po)	0.5	2	8	1	2
antagonism of SKF 10047-induced hypermotility (% vs controls)[a]	39*	56**	71****	94***	
antagonism of PCP-induced stereotypies (% vs controls)[a]	12	2	21		87***

[a] $*p < 0.05$. $**p < 0.01$. $***p < 0.001$. $n = 12$ animals per group.

Table 7. Effects of Compound **24** on Sidman Avoidance Performance in Rats

	24			haloperidol	clozapine
dose (mg/kg po)	8	16	32	1	32
number of avoidance responses (% change from control)[a]	−12 ns	−38**	−78***	−86***	−62**
number of shocks (absolute change from control)[a]	+7.3 ns	+23.8 ns	+87.8***	+99.7***	+71.1**
number of escape failure (absolute change from control)[a]	+5.8 ns	+18.5 ns	+63.6**	+84.6***	+53.8*

[a] $*p < 0.05$. $**p < 0.01$. $***p < 0.001$. $n = 10$ animals per group.

neuroleptics increased the duration of immobility while they had no clear effects on the power of movements (Table 5).

The different arylpiperazine derivatives tested thus showed clear activity in different tests predictive of an antipsychotic activity: these models were based on the dopaminergic and the serotonergic hypothesis of schizophrenia. Compounds **24**, **25**, **30**, and **31** were tested for their sedative potential using an interaction test with the barbital-induced sleep. Compounds **24** (2 mg/kg ip) and **25** (16 mg/kg ip) weakly increased the duration of the sleep, respectively 50% and 70% (ns) of increase in comparison with barbital treated animals (non significant). On the contrary, compounds **30** and **31** dramatically increased the duration of sleep (+400% for compound **30** at the dose of 8 mg/kg ip and +360% for compound **31** at the doses of 64 mg/kg ip, $p < 0.001$). This latter effect was similar to the one induced by the typical antipsychotic haloperidol at the dose of 0.5 mg/kg ip (Table 6).

Compound **24** seems very promising since it was active at low doses and was interestingly devoid of clear sedative effects at dose active in these models and thus was tested in other models, based on the σ and the NMDA hypothesis of schizophrenia.

Compound **24** showed a clear dose dependent antagonism of the hyperlocomotion induced by the σ agonist, SKF 10047, and of the stereotypies induced by the phencyclidine (PCP).

Finally compound **24** was investigated after oral administration in rats to evaluate its effects in the Sidman avoidance test.

Compound **24** dose-dependently decreased the number of avoidance responses (Table 7). This effect was associated only at the highest dose with a significant increase in the number of shocks received and in the number of escape failures (i.e. decrease in the ability of animals to escape shocks). For comparison, haloperidol decreased the avoidance response and increased the escape failure at low doses.

To qualify a putative antipsychotic agent as atypical, its low propensity to induce extrapyramidal side effects (EPS) has to be demonstrated.

Compounds **24** and **47** were tested at pharmacological doses (1 and 2 mg/kg ip, respectively) and at high doses (32 and 64 mg/kg ip) in rats for their potential ability to induce catalepsy indicative of EPS production in human.

Both compounds did not induce catalepsy in rats at doses active in models predictive of an antipsychotic activity. Compound **24** increased significantly catalepsy at the dose of 32 mg/kg while compound **47** was devoid of any catalepsigenic effects.

The amphetamine-induced stereotypies are thought to result from dopaminergic activation in the striatum. Compounds **24** and **30** which were previously shown to antagonize the hyperactivity induced by *d*-amphetamine (resulting from DA activation in the limbic system) were tested for their ability at the same dose to block the stereotypies induced by amphetamine.

Compound **24** neither blocked the stereotypies induced by amphetamine in mice (0.25 and 1 mg/kg ip) nor those induced in rats (8 mg/kg PO); these doses were previously found active on amphetamine-induced hyperlocomotion.

Conclusion

We have designed and synthesized novel original ligands with mixed affinities for the $5HT_{1A}$ and D_2 receptors for which the expected profiles were indeed obtained. Moreover, we have obtained potent and selective pharmacological tools in the field of central

$5HT_{1A}$ ligands. Five compounds (**24, 27**, **30**, **46**, and **47**) which could be qualified as potential "atypical" antipsychotics emerged in this series as they do not induce any (or only a low level of) cataleptic behavior which is indicative of EPS.

Experimental Section

Chemistry. Compounds were characterized by elemental analyses and IR and ^{1}H NMR spectra. IR spectra were recorded on a Perkin-Elmer 297 spectrophotometer, using KBr tablets; the frequencies are expressed in cm^{-1}. The ^{1}H NMR spectra were obtained on a Brücker WP 80 SY (80 MHz) spectrometer, with Me_4Si as the internal standard and with $CDCl_3$ or DMSO-d_6 as solvent; the chemical shifts are reported in δ units and the coupling constants in hertz. Melting points were determined using a Büchi SMP-20 apparatus and are uncorrected. Elemental analyses were determined by the CNRS center of analysis in Vernaison (France). Elemental analyses were within 0.4% of the theoretical values. Compounds **3**−**16**, **32**−**34**, and **39**−**42** were prepared according to published procedures.[18,19,25,28]

General Procedure for the Synthesis of the Arylpiperazinoalkylbenzothiazolin-2-one (17, 19−21, 23−31) or benzoxazolin-2-one (35, 36) Derivatives. The method adopted for the synthesis of 6-(2-(4-phenylpiperazin-1-yl)ethyl)-benzothiazolin-2-one hydrochloride **(17)** is described. A stirred solution of 2.60 g (0.01 mol) of 6-(2-bromoethyl)benzothiazolin-2-one **(11)**, 0.96 g (0.01 mol) of 1-phenylpiperazine, and 1.01 g (0.01 mol) of triethylamine in 150 mL of dry acetone was heated under reflux for 72h and then concentrated in vacuo. The residue was triturated in 50 mL of a 5% aqueous solution of HCl. The resulting precipitate was filtered, washed with H_2O, treated with a 10% aqueous solution of Na_2CO_3, and extracted with EtOAc. The combined organic layers were dried (K_2CO_3) and evaporated. The resulting residue was dissolved in absolute ethanol, and dry HCl was bubbled into the solution. The formed solid was filtered, dried, and recrystallized from 95% EtOH to give 2.25 g of **17** (60% yield): mp >250 °C; ^{1}H NMR (DMSO-d_6) δ 3.00−4.10 (m, 12H, CH_2-CH_2 + piperazine); 6.80−7.50 (m, 8H, aromatic); 11.80 (s, 1H, NH^+); 12.00 (s, 1H, NH). Anal. ($C_{19}H_{21}N_3OS\cdot1HCl\cdot{}^1/_2H_2O$) C, H, N.

6-(2-(4-(2-Methoxyphenyl)piperazin-1-yl)ethyl)-benzothiazolin-2-one Hydrochloride (19). Recrystallized from H_2O (70% yield): mp >250 °C; ^{1}H NMR (DMSO-d_6) δ 2.20 (m, 12H, CH_2CH_2 and piperazine moiety); 3.75 (s, 3H, OCH_3); 6.80−7.50 (m, 7H, aromatic); 11.80 (s, 2H, NH + NH^+). Anal. (C_{20} $H_{23}N_3O_2S\cdot1HCl$) C, H, N.

6-(2-(4-(3-Trifluoromethylphenyl)piperazin-1-yl)ethyl-)benzothiazolin-2-one (20). Recrystallized from 1-propanol to give 1.83 g (45% yield): mp 132 °C; ^{1}H NMR (DMSO-d_6) δ 2.39−3.50 (m, 12H, CH_2CH_2 + piperazine); 6.96−7.60 (m, 7H, aromatic); 11.75 (s, 1H, NH). Anal. (C_{20} $H_{20}F_3N_3OS$) C, H, N.

3-Methyl-6-(2-(4-phenyl)piperazin-1-yl)ethyl)benzothiazolin-2-one Hydrochloride (21). Recrystallized from 95% EtOH (77% yield): mp 226−228 °C; ^{1}H NMR (DMSO-d_6) δ 3.20−3.30 (m, 8H, $CH_2CH_2N(CH_2)_2$); 3.35−3.40 (m, 7H, $N(CH_2)_2$ + NCH_3); 7.00−7.60 (m, 8H, aromatic); 11.00 (s, 1H, NH^+). Anal. ($C_{20}H_{23}N_3OS\cdot1HCl$) C, H, N.

3-Methyl-6-(2-(4-(4-fluorophenyl)piperazin-1-yl)ethyl)-benzothiazolin-2-one Hydrochloride (23). Recrystallized from absolute EtOH (45% yield): mp 232−235 °C; ^{1}H NMR (DMSO-d_6) δ 3.25−3.80 (m, 12H, $ArCH_2CH_2$ piperazine); 3.40 (s, 3H, NCH_3); 6.70−7.60 (m, 7H, aromatic); 11.90 (s, 2H, NH^+). Anal. ($C_{20}H_{22}FN_3OS\cdot2HCl$) C, H, N.

3-Methyl-6-(2-(4-(2-methoxyphenyl)piperazin-1-yl)-ethyl)benzothiazolin-2-one Hydrochloride (24). Recrystallized from MeOH (55% yield): mp >250 °C; ^{1}H NMR (DMSO-d_6) δ 3.10−3.60 (m, 12H, CH_2CH_2 + piperazine); 3.40 (s, 3H, NCH_3); 3.85 (s, 3H, OCH_3); 6.95−7.60 (m, 7H, aromatic); 11.30 (s, 1H, NH^+). Anal. ($C_{21}H_{25}N_3O_2S\cdot1HCl$) C, H, N.

3-Methyl-6-(2-(4-(3-trifluoromethylphenyl)piperazin-1-yl)ethyl)benzothiazolin-2-one Hydrochloride (25). Recrystallized from absolute EtOH (50% yield): mp 236−237 °C; ^{1}H NMR (DMSO-d_6) δ 3.20−3.85 (m, 12H, CH_2CH_2 + piperazine); 3.40 (s, 3H, NCH_3); 7.00−7.55 (m, 7H, aromatic); 11.30 (s, 1H, NH^+). Anal. ($C_{21}H_{22}F_3N_3OS\cdot1HCl$) C, H, N.

6-(3-(4-(2-Methoxyphenyl)piperazin-1-yl)propyl)-benzothiazolin-2-one Hydrochloride (26). Recrystallized from absolute EtOH (65% yield): mp 224−225 °C; ^{1}H NMR (DMSO-d_6) δ 2.05 (m, 2H, CH_2); 2.65 (t, J = 7.30 Hz, 2H, $ArCH_2$); 3.15 (m, 6H, $CH_2N(CH_2)_2$); 3.50 (m, 4H, $(CH_2)_2N$); 3.80 (s, 3H, OCH_3); 7.00−7.50 (m, 7H, aromatic); 11.10 (s, 1H, NH^+); 11.95 (s, 1H, NH). Anal. ($C_{21}H_{25}N_3O_2S\cdot1HCl$) C, H, N.

3-Methyl-6-(3-(4-(2-methoxyphenyl)piperazin-1-yl)-propyl)benzothiazolin-2-one Hydrochloride (27). Recrystallized from absolute EtOH (40% yield): mp 214−216 °C; ^{1}H NMR (DMSO-d_6) δ 2.10 (m, 2H, CH_2); 2.70 (t, J = 7.30 Hz, 2H, $ArCH_2$); 3.10 (m, 6H, $CH_2N(CH_2)_2$); 3.40 (s, 3H, NCH_3); 3.50 (m, 4H, $(CH_2)_2N$); 3.80 (s, 3H, OCH_3) 7.00−7.60 (m, 7H, aromatic); 11.25 (s, 1H, NH^+). Anal. ($C_{22}H_{27}N_3O_2S\cdot1HCl$) C, H, N.

6-(4-(4-(2-Methoxyphenyl)piperazin-1-yl)butyl)-benzothiazolin-2-one (28). Recrystallized from toluene (55% yield): mp 125−127 °C; ^{1}H NMR (DMSO-d_6) δ 1.60−1.80 (m, 4H, CH_2CH_2); 2.30−2.50 (m, 2H, $ArCH_2$); 3.00−3.40 (m, 10H, CH_2N + piperazine); 3.80 (s, 3H, OCH_3); 6.80−7.60 (m, 7H, aromatic); 11.90 (s, 1H, NH). Anal. ($C_{22}H_{27}N_3O_2S$) C, H, N.

6-(4-(4-(3-Trifluoromethylphenyl)piperazin-1-yl)butyl-)benzothiazolin-2-one (29). Recrystallized from 95% EtOH (46% yield): mp 129−130 °C; ^{1}H NMR (DMSO-d_6) δ 1.40−1.60 (m, 4H, CH_2CH_2); 2.40−2.50 (m, 4H, $ArCH_2$ + CH_2N); 3.00−3.40 (m, 8H, piperazine); 7.00−7.50 (m, 7H, aromatic); 11.80 (s, 1H, NH). Anal. ($C_{22}H_{24}F_3N_3OS$) C, H, N.

3-Methyl-6-(4-(4-(2-methoxyphenyl)piperazin-1-yl)-butyl)benzothiazolin-2-one Hydrochloride (30). Recrystallized from absolute EtOH (48% yield): mp 228−230 °C; ^{1}H NMR (DMSO-d_6) δ 1.50−2.00 (m, 4H, CH_2CH_2); 2.50−3.00 (m, 4H, NCH_2 + $ArCH_2$); 3.30−3.70 (m, 11H, NCH_3 + piperazine); 3.80 (s, 3H, OCH_3); 6.80−7.50 (m, 7H, aromatic); 11.20 (s, 2H, NH^+). Anal. ($C_2H_{29}N_3O_2S\cdot2HCl$) C, H, N.

3-Methyl-6-(4-(4-(3-trifluoromethyl)phenylpiperazin-1-yl)butyl)benzothiazolin-2-one Hydrochloride (31). Recrystallized from toluene (67% yield): mp 182−184 °C; ^{1}H NMR (DMSO-d_6) δ 1.10−1.40 (m, 4H, CH_2CH_2); 1.60−2.00 (m, 4H, NCH_2 + $ArCH_2$); 3.20−3.70 (m, 11H, NCH_3 + piperazine); 6.90−7.70 (m, 7H, aromatic); 13.40 (s, 2H, NH^+). Anal. ($C_{23}H_{26}F_3N_3OS\cdot2HCl$) C, H, N.

3-Methyl-6-(2-(4-(2-methoxyphenyl)piperazin-1-yl)-ethyl)benzoxazolin-2-one (35). Recrystallized from cyclohexane (65% yield): mp 137−138 °C; ^{1}H NMR ($CDCl_3$) δ 2.80 (m, 8H, $CH_2CH_2N(CH_2)_2$); 3.15 (m, 4H, $(CH_2)_2N$); 3.40 (s, 3H, NCH_3); 3.90 (s, 3H, OCH_3); 6.80−7.20 (m, 7H, aromatic). Anal. ($C_{21}H_{25}N_3O_3$) C, H, N.

3-Methyl-6-(2-(4-(3-trifluoromethylphenyl)piperazin-1-yl)ethyl)benzoxazolin-2-one (36). Recrystallized from cyclohexane (70% yield): mp 110 °C; ^{1}H NMR ($CDCl_3$) δ 2.70 (m, 8H, $CH_2CH_2N(CH_2)_2$); 3.30 (m, 4H, $(CH_2)_2N$); 3.40 (s, 3H, NCH_3); 6.90−7.30 (m, 7H, aromatic). Anal. ($C_{21}H_{22}F_3N_3O_2$) C, H, N.

Synthesis of the 6-(2-(4-(2-hydroxyphenyl)piperazin-1-yl)ethyl)benzothiazolin-2-one Derivatives (18, 22). The method adopted for the synthesis of 6-(2-(4-(2-hydroxyphenyl)-piperazin-1-yl)ethyl)benzothiazolin-2-one **(18)** is described. A stirred solution of 4.00 g (0.01 mol) of 6-(2-(4-(2-methoxyphenyl)piperazin-1-yl)ethyl)benzothiazolin-2-one hydrochloride **(19)** in 100 mL of a 47% aqueous solution of HBr was heated under reflux for 48 h. The formed precipitate was filtered, washed with acetone, and dissolved in 100 mL of H_2O. The solution was made basic with a 20% aqueous solution of NaOH and extracted with $CHCl_3$. The organic layer was washed with H_2O, dried over $CaCl_2$, and evaporated in vacuo. The residue was dissolved in dry acetone, and HCl was bubbled to give the hydrochloride salt which was filtered, dried, and recrystallized from absolute EtOH to give 1.76 g of pure **18** (45%

yield): mp 164−166 °C; ^{1}H NMR (DMSO-d_6) δ 3.00−3.30 (m, 8H, $CH_2CH_2N(CH_2)_2$); 3.50 (m, 4H, $(CH_2)_2N$)); 5.80 (s, 1H, NH); 6.75−7.50 (m, 7H, aromatic); 11.20 (s, 1H, OH); 12.00 (s, 1H, NH^+). Anal. ($C_{19}H_{21}N_3O_2S$·1HCl) C, H, N.

3-Methyl-6-(2-(4-(2-hydroxyphenyl)piperazin-1-yl)-ethyl)benzothiazolin-2-one hydrochloride (22). Recrystallized from absolute EtOH (64% yield): mp > 260 °C; ^{1}H NMR (DMSO-d_6) δ 3.10−3.60 (m, 12H, CH_2CH_2 + piperazine); 3.40 (s, 3H, NCH_3); 6.05 (s, 1H, OH); 7.00−7.60 (m, 7H, aromatic); 11.50 (s, 1H, NH^+). Anal. ($C_{21}H_{25}N_3O_2S$·1HCl) C, H, N.

General Procedure for the Synthesis of the Aminophenol Derivatives (37, 38). The method adopted for the synthesis of 5-(2-(4-(2-methoxyphenyl)piperazin-1-yl)ethyl)-2-(methylamino)phenol **(37)** is described. A mixture of 36.7 g (0.1 mol) of 3-methyl-6-(2-(4-(2-methoxyphenyl)piperazin-1-yl)-ethyl)benzoxazolin-2-one **(35)**, 300 mL of 10% aqueous solution of NaOH, and 150 mL of MeOH was heated under reflux for 3 h. After filtration, the filtrate was made acidic with a 6 N aqueous solution of HCl and filtered. The filtrate was then made basic with a 10% aqueous solution of Na_2CO_3; the resulting precipitate was filtered, washed with H_2O, dried, and recrystallized from toluene to give 19.40 g of pure **37** (57% yield): mp 172−174 °C; ^{1}H NMR (DMSO-d_6) δ 2.40−2.60 (m, 8H, $CH_2CH_2N(CH_2)_2$); 2.65 (s, 3H, NCH_3); 3.00 (m, 4H, $N(CH_2)_2$); 3.75 (s, 3H, OCH_3); 6.35−7.90 (m, 7H, aromatic); 9.10 (s, 1H, OH). Anal. ($C_{20}H_{27}N_3O_2$) C, H, N.

5-(2-(4-(3-Trifluoromethylphenyl)piperazin-1yl)ethyl)-2-methylaminophenol (38). Recrystallized from MeOH (59% yield): mp 158−161 °C; ^{1}H NMR (DMSO-d_6) δ 3.45−3.75 (m, 11H, $CH_2CH_2N(CH_2)_2$ + NCH_3); 4.20 (m, 4H, $N(CH_2)_2$); 4.50 (s, 1H, NH); 7.30−8.50 (m, 7H, aromatic); 9.00 (s, 1H, OH). Anal. ($C_{20}H_{24}N_3O$) C, H, N.

7-(4-Bromobutyl)-4-methyl-3-oxo-2,3-dihydro[1,4]-benzoxazine (43). **42** (12.50 g, 0.08 mol) was dissolved in CF_3COOH (70 mL), and under stirring at room temperature, triethylsilane (39.90 mL, 0.25 mol) was added dropwise. After 20 h, the mixture was poured onto ice. The resulting precipitate was filtered, washed with H_2O, dried, and recrystallized from cyclohexane to give 17.90 g of pure **43** (60% yield): mp 41−42 °C; ^{1}H NMR (DMSO-d_6) δ 1.75 (m, 4H, CH_2CH_2); 2.50 (t, J = 6.00 Hz, 2H, $ArCH_2$); 3.50 (t, J = 6.00 Hz, 2H, CH_2Br); 4.60 (s, 2H, $COCH_2O$); 6.80−7.20 (m, 3H, aromatic). Anal. ($C_{13}H_{16}BrNO_2$) C, H, N.

General Procedure for the Synthesis of the 3-Oxo-2,3-dihydro[1,4]benzoxazine Derivatives (44, 45). The method adopted for the synthesis of 7-(2-(4-(2-methoxyphenyl)piperazin-1-yl)ethyl)-4-methyl-3-oxo-2,3-dihydro[1,4]benzoxazine (**44)** is described. **37** (3.40 g, 0.01 mol) was dissolved in DMSO (120 mL). Under stirring, 0.68 g (0.01 mol) of sodium ethanolate was added and then dropwise 1.10 mL (0.01 mol) of ethyl bromoacetate. The mixture was stirred at room temperature for 3 h and then poured onto ice water and extracted with EtOAc. The organic layer was dried over K_2CO_3, filtered, and evaporated in vacuo. The resulting oil was recrystallized from absolute EtOH to give 1.10 g of pure **44** (30% yield): mp 117−119 °C; ^{1}H NMR (DMSO-d_6) δ 3.10−3.40 (m, 11H, $CH_2CH_2N(CH_2)_2$ + NCH_3); 3.60 (m, 4H, $N(CH_2)_2$); 3.80 (s, 3H, OCH_3); 4.65 (s, 2H, $COCH_2O$); 6.90−7.10 (m, 7H, aromatic). Anal. ($C_{22}H_{27}N_3O_3$) C, H, N.

7-(2-(4-(3-Trifluoromethylphenyl)piperazin-1-yl)ethyl)-4-methyl-3-oxo-2,3-dihydro[1,4]benzoxazine Hydrochloride (45). Recrystallized from acetone (30% yield): mp 241−243 °C; ^{1}H NMR (DMSO-d_6) δ 3.00−3.50 (m, 11H, $CH_2CH_2N(CH_2)_2$ + NCH_3); 3.60−4.00 (m, 4H, $N(CH_2)_2$); 4.70 (s, 2H, $COCH_2O$); 6.90−7.10 (m, 7H, aromatic); 11.50 (s, 1H, NH^+). Anal. ($C_{22}H_{24}F_3N_3O_2$·1HCl) C, H, N.

7-(4-(4-(2-Methoxyphenyl)piperazin-1-yl)butyl)-4-methyl-3-oxo-2,3-dihydro[1,4]benzoxazine (46). Starting from **43**, compound **46** was synthesized using the same procedure as for **17** and recrystallized from absolute EtOH to give 2.80 g of pure material (70% yield): mp 90−92 °C; ^{1}H NMR (DMSO-d_6) δ 1.50 (m, 4H, CH_2CH_2); 2.45−2.70 (m, 8H, $ArCH_2$ + $CH_2N(CH_2)_2$); 3.25 (s, 3H, NCH_3); 3.40 (m, 4H, $(CH_2)_2N$); 3.80 (s, 3H, OCH_3); 4.60 (s, 2H, $COCH_2O$); 6.80−7.10 (m, 7H, aromatic). Anal. ($C_{24}H_{31}N_3O_3$) C, H, N.

7-(4-(4-(3-Trifluoromethylphenyl)piperazin-1-yl)butyl)-4-methyl-3-oxo-2,3-dihydro[1,4]benzoxazine (47). Starting from **43**, compound **47** was synthesized using the same procedure as for **17**. The hydrochloride salt was obtained by bubbling HCl in dry acetone, and recrystallization from absolute EtOH yielded 3.40 g of pure material (70% yield): mp 205 °C; ^{1}H NMR (DMSO-d_6) δ 1−60 (m, 4H, CH_2CH_2); 2.75−3.50 (m, 15H, $ArCH_2$ + CH_2 piperazine + NCH_3); 4.60 (s, 2H, $COCH_2O$); 6.90−7.40 (m, 7H, aromatic); 11.10 (s, 1H, NH^+). Anal. ($C_{24}H_{28}F_3N_3O_2$·1HCl) C, H, N.

Binding Studies. $5HT_{1A}$ Receptor. Binding was determined using membranes prepared from bovine hippocampus. The receptor was labeled with 0.5 nM [^{3}H]-8-hydroxydipropylaminotetralin (8-OH-DPAT) by incubation at 25 °C for 30 min with 11 concentrations of the test compounds (10^{-9}−10^{-4} M). Nonspecific binding was determined using 10^{-5} M buspirone.[30] Competition experiments were analyzed using the iterative nonlinear least-squares curve-fitting program Inplot 4, graphpad; IC_{50} values were calculated using the Cheng−Prusoff equation.[31]

$5HT_{1B}$ Receptor. Binding was determined using membranes prepared from rat frontal cortex. The receptor was labeled with 2 nM [^{3}H]-5-hydroxytryptamine by incubation at 25 °C for 30 min. Nonspecific binding was determined using 10^{-5} M propranolol.[32] Competition experiments were analyzed using the iterative nonlinear least-squares curve-fitting program Inplot 4, graphpad; IC_{50} values were calculated using the Cheng−Prusoff equation.[31]

$5HT_{2A}$ Receptor. Binding was determined using membranes prepared from bovine frontal cortex. The receptors was labeled with 0.8 nM [^{3}H]ketanserin by incubation for 30 min at 37 °C. Nonspecific binding was determined using 10^{-5} M spiperone.[33] Competition experiments were analyzed using the iterative nonlinear least-squares curve-fitting program Inplot 4, graphpad; IC_{50} values were calculated using the Cheng−Prusoff equation.[31]

$5HT_{2C}$ Receptor. Binding was determined using membranes prepared from pig choroid plexus. The receptor was labeled with 1.2 nM [^{3}H]-*N*-methylmesulergine by incubation at 25 °C for 30 min. Nonspecific binding was determined using 10^{-5} M mianserine.[34] Competition experiments were analyzed using the iterative nonlinear least-squares curve-fitting program Inplot 4, graphpad; IC_{50} values were calculated using the Cheng−Prusoff equation.[31]

D_2 Receptor. Binding was determined using membranes prepared from bovine striatum. The receptor was labeled with 1.2 nM [^{3}H]raclopride by incubation at 25 °C for 30 min with 11 concentrations of the test compounds (10^{-9}−10^{-4} M). Nonspecific binding was determined using 10^{-5} M spiperone.[35,36] Competition experiments were analyzed using the iterative nonlinear least-squares curve-fitting program Inplot 4, graphpad; IC_{50} values were calculated using the Cheng−Prusoff equation.[31]

α_1 Receptor. Binding was determined using membranes prepared from bovine frontal cortex. The receptor was labeled with 0.5 nM [^{3}H]prazosin by incubation at 25 °C for 40 min with 11 concentrations of the test compounds (10^{-9}−10^{-4} M). Nonspecific binding was determined using 10^{-5} M phentolamine.[37] Competition experiments were analyzed using the iterative nonlinear least-squares curve-fitting program Inplot 4, graphpad; IC_{50} values were calculated using the Cheng−Prusoff equation.[31]

Nature of the Interaction with Receptors. In Vivo 5HT Behavorial Syndrome in Rats. Wistar rats (n = 6) are injected with the test compound immediately before test and are scored for the intensity of forepaw treading on a scale; The intensity of forepaw treading is expressed as percentage of the maximal possible score. The $5HT_{1A}$ agonist 8-OH-DPAT induces forepaw treading and is used as a reference compound.[37]

Antagonism of head twitches induced by L-5-hydroxytryptophane (5HTP) in mice indicates classical antiserotonin-

ergic activities.[38] Swiss mice were injected with the test compound before an injection of 5HTP (400 mg/kg ip). The number of head twiches occurring in a 10 min period starting 10 min after the injection of 5HTP is counted. Cyproheptadine was used as reference compound.

Antagonism of a low dose of apomorphine in mice indicates a classical antidopaminergic activity.[39] The intensity of apomorphine (1 mg/kg sc) induced stereotypies, climbing, and the rectal temperature were measured 30 min after apomorphine. Haloperidol was used as a reference compound.

The interaction with α_1 adrenoceptors was assessed in vitro on rabbit thoracic aorta.[40,41] Compounds were tested for their ability to antagonize the contractile effect of phenylephrine (antagonistic effect) or for their ability to induce a contractile effect (agonistic effect) which can be blocked with prazosin.

Antagonism of *d*-Amphetamine-Induced Hyperactivity in Mice and Rats. Locomotor activity was measured in Plexiglass chambers. Swiss mice or Wistar rats were pretreated with *d*-amphetamine (4 mg/kg ip) followed by vehicle or test compounds 30 min after. Data were recorded for 30 min.[42] Treatment group means were compared with a Student's *t* test.

Interaction with Sleep Induced by Barbital. Swiss mice were injected with barbital (400 mg/kg ip), and the duration of sleep was recorded.[43] The data were calculated as the percent of potentiation of the sleep duration produced by barbital.

Mescaline-Induced Scratch Paroxysms in Mice. This is a modification of Deegan and Cook procedure.[44] Swiss mice were injected with mescalin·HCl at 25 mg/kg ip and placed into square cages. The number of scratching paroxysms was counting for 5 min, beginning 30 min after treatment. A scratching paroxysm was defined as a brief burst of scratching of the head or ear with the hind foot. The data were calculated as the percent antagonism of the scratching produced by mescaline at 25 mg/kg ip.

The tail suspension test is a new variant of the behavorial despair test which allows identification of different classes of psychotropic activities.[45] Neuroleptics increase the duration of immobility while they have little effects on the power of the movements. Swiss mice were suspended by the tail for 6 min. The behavior of the animals was recorded automatically using a special computerized apparatus (Itematic-TST).

Conditionned Avoidance Behavior in Rats. In this test, Lister Hooded rats have to press a lever in a Skinner Box in order to avoid an electric shock.[46] Antipsychotics block this behavior. Pretrained animals were conditioned to press the lever at regular intervals to avoid electric shocks. Every time the lever was pressed, the next shock was postponed by 20 s. When the animals failed to press, the shock lasted 5 s and returned 5 s later. Each animal was its own control. The number of avoidances and escapes were recorded. The data were calculated as the percent antagonism of avoidance and escape responding.

SKF 10047-Induced Hyperactivity in Rats. (+)-SKF 10047 (*N*-allylnormetazocine) induces psychotic behavior in humans and is the prototype agonist for σ sites. (+)-SKF 10047 (50 mg/kg sc) induces marked hyperactivity in rats which can be reduced by atypical antipsychotics acting at the sigma sites.[47] Groups of 12 Wistar rats received (+)-SKF 10047 and the test compounds 30 min before the test. They were then placed in an activity meter for a 30 min test. The data were expressed as the percent antagonism of the hyperactivity.

Catalepsy in Rats. Wistar rats were administered test compounds or vehicle and tested for catalepsy at 30 min intervals for 90 min. Catalepsy was assessed by three procedures: imposed crossing of the ipsilateral fore- and hindlimbs, placing in the Buddha position, and the tilting board, an automatic device which displaced the rat from a horizontal to vertical position and back while it clung to a wire gird with its front paw.[48]

Stereotypies Induced by *d*-Amphetamine in Mice and Rats. Swiss mice were pretreated with *d*-amphetamine (4 mg/kg ip) and were scored for the intensity of stereotypies on a 4-point scale 30 min later; Wistar rats receive the same dose of *d*-amphetamine but observations were carried out every 10 min until the disappearance of the stereotypies.

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