Conformationally Constrained Butyrophenones with Affinity for Dopamine (D_1, D_2, D_4) and Serotonin (5-HT_{2A}, 5-HT_{2B}, 5-HT_{2C}) Receptors: Synthesis of Aminomethylbenzo[*b*]furanones and Their Evaluation as Antipsychotics§

Enrique Raviña,*,† Isabel Casariego,† Christian F. Masaguer,† José A. Fontenla,‡ Gisela Y. Montenegro,‡ María E. Rivas,‡ M. Isabel Loza,‡ Maria J. Enguix,‡ Maria Villazon,‡ M. Isabel Cadavid,‡ and Gian C. Demontis||

Departamento de Química Orgánica, Laboratorio de Química Farmacéutica, and Departamento de Farmacología, Facultad de Farmacia, Universidad de Santiago, E-15706 Santiago de Compostela, Spain, and Dipartimento di Psichiatria, Neurobiologia, Farmacologia e Biotecnologia, Laboratorio di Neurobiologia, Universita di Pisa, Via Bonanno Pisano 6, I-56126 Pisa, Italy

Received May 16, 2000

A series of novel conformationally restricted butyrophenones (6-aminomethyl-4,5,6,7-tetrahydrobenzo[*b*]furan-4-ones bearing 4-(6-fluorobenzisoxazolyl)piperidine, 4-(*p*-fluorobenzoyl)piperidine, 4-(*o*-methoxyphenyl)piperazine, 4-(2-pyridyl)piperazine, 4-(2-pyrimidinyl)piperazine, or linear butyro(or valero)phenone fragments) were prepared and evaluated as antipsychotic agents by in vitro assays for affinity for dopamine receptors (D_1, D_2, D_4) and serotonin receptors (5-HT_{2A}, 5-HT_{2B}, 5-HT_{2C}), by neurochemical studies, and by in vivo assays for antipsychotic potential and the risk of inducing extrapyramidal side effects. Potency and selectivity depended mainly on the amine fragment connected to the cyclohexanone structure. Compounds **20b**, with a benzoylpiperidine moiety, and **20c**, with a benzisoxazolyl fragment, were selective for 5-HT_{2A} receptors. The in vitro and in vivo pharmacological profiles of *N*-[(4-oxo-4,5,6,7-tetrahydrobenzo-[*b*]furan-6-yl)methyl]-4-(*p*-fluorobenzoyl)piperidine (**20b**, QF1003B) and *N*-[(4-oxo-4,5,6,7-tetrahydrobenzo[*b*]furan-6-yl)methyl]-4-(6-fluorobenzisoxazol-3-yl)piperidine (**20c**, QF1004B) suggest that they may be effective as antipsychotic (neuroleptic) drugs.

Introduction

Schizophrenia is a complex disorder affecting approximately 1% of the population.[1] Classical neuroleptics used in its treatment, such as haloperidol (**1**, Chart 1), are poorly effective against its negative symptoms (apathy, motor retardation, flat affectivity, poor speech, etc.). Furthermore, the use of these classical antipsychotics is associated with severe side effects, notably acute extrapyramidal symptoms (EPS),[2] that appear to be an unavoidable consequence of their mechanism of action. Indeed, both their induction of EPS and their antipsychotic efficacy originate from blockade of dopamine (DA) receptors: striatal receptors in the case of EPS induction[3−5] and mesolimbic receptors in the case of antipsychotic activity.[6−8]

Clozapine (**2**) was the first of a new group of "atypical" or nonclassical antipsychotics that cause no EPS and are effective against negative symptoms.[9−12] Since clozapine blocks not only DA receptors but also subtype 5-HT_{2A} serotonin receptors, it is thought that its atypical activity profile may be due to its effects on interaction between the serotonin and DA systems;[13−15] more specifically, it is believed that blockade of 5-HT_{2A} receptors in the limbic and cortical areas favors efficacy against negative symptoms and that their blockade in the striatal area reduces EPS, in both cases due to pharmacological deconnection of serotoninergic projections from the midline nuclei.[16] Hence Meltzer et al.[17,18] suggested that the profiles of clozapine and other atypical antipsychotics, such as risperidone (**3**), olanzapine (**4**), and quetiapine (**5**), depend largely on their relative affinities for D_2 and 5-HT_{2A} receptors.[19] They proposed that the ratio pK_i 5-HT_{2A}/D_2 (between pK_i for 5-HT_{2A} and pK_i for D_2) may be used to discriminate atypical antipsychotics, having a ratio > 1.12, from classical antipsychotics, which have a ratio < 1.09. Note that this latter finding raises the question of whether there is a D_2 affinity threshold, below which there is no antipsychotic activity even if the pK_i 5-HT_{2A}/D_2 ratio is high: clozapine has only moderate affinity for D_2, and the 5-HT_{2A}-selective serotonin antagonist MDL 100907 (**6**), which has no specific affinity for DA receptors, has shown antipsychotic potential in experiments with neurochemical, electrophysiological, and behavioral models.[20−22] Nevertheless, the "5-HT_{2A}/D_2 concept" has contributed to the development not only of atypical antipsychotics with a close chemical similarity to clozapine, such as olanzapine (**4**), quetiapine (**5**), and zotepine (**9**), but also of others of an apparently different chemical nature, such as risperidone (**3**), ocaperidone (**7**), and sertindole (**8**).

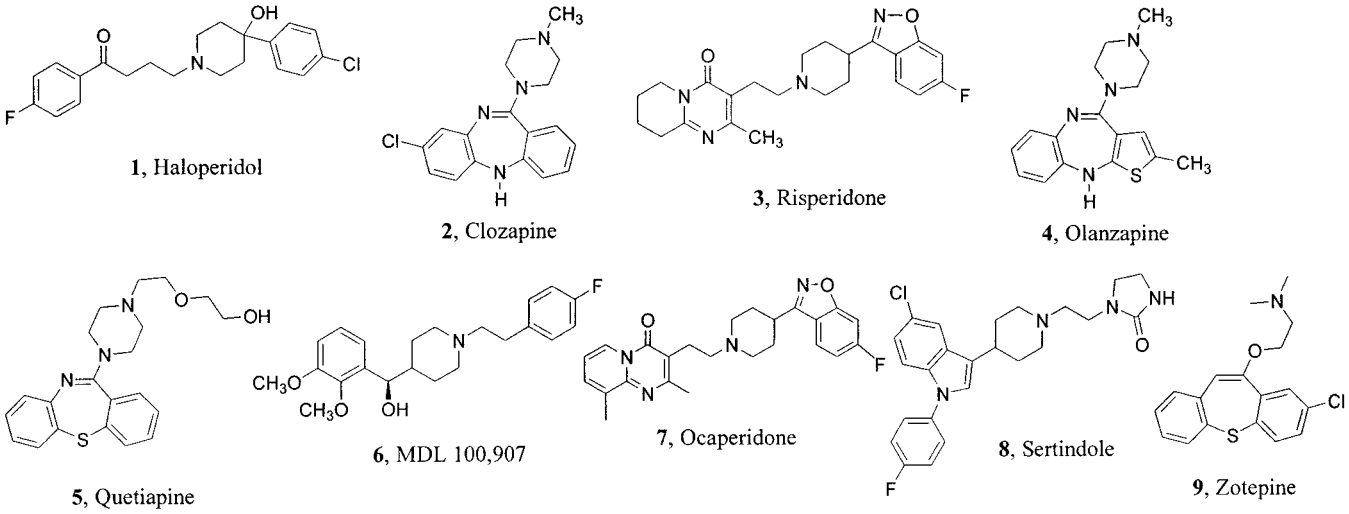
As well as to 5-HT_{2A}, atypical antipsychotics also bind to other subtype 2 serotonin receptors (5-HT_{2B} and 5-HT_{2C}),[23] and they are implicated not only in schizophrenia but also in anxiety, depression, anorexia nervosa, migraine, and cardiovascular disorders, including hypertension.[24] The subtypes of this family (5-HT_{2A}, 5-HT_{2B}, and 5-HT_{2C}) are defined on the basis of primary structure, secondary messengers, and operational pro-

§ This is the 22nd paper in the series "Synthesis and CNS Activity of Conformationally Restricted Butyrophenones"; for preceding papers, see ref 47.

* Correspondence and reprints: Prof. Enrique Raviña. Phone: +34-981-594628. Fax: +34-981-594912. E-mail: qofara@usc.es.
† Departamento de Química Orgánica, Universidad de Santiago.
‡ Departamento de Farmacología, Universidad de Santiago.
|| Universita di Pisa.

10.1021/jm0009890 CCC: $19.00
Published on Web 11/10/2000

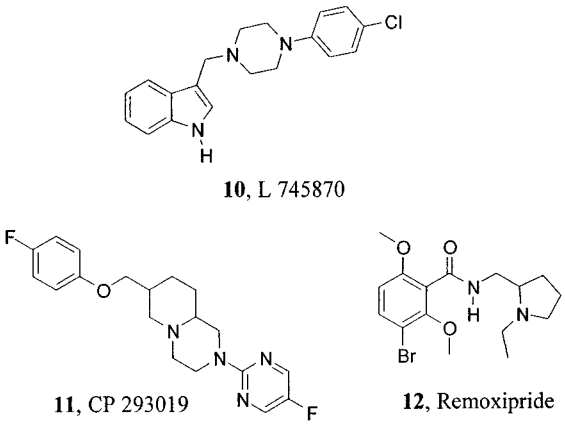
Chart 1



file[24,25] and have different distributions in the body: 5-HT_{2A} is found both in the brain and in peripheral tissues,[26] 5-HT_{2B} is mainly peripheral,[27] and 5-HT_{2C} has only been found in the central nervous system (CNS) (mainly in the choroid plexus, limbic structures, and basal ganglia, a distribution different from that of 5-HT_{2A}).[28] It is now thought that some pathologies or pharmacological effects previously attributed to dysfunction or action at 5-HT_{2A} may in fact be due to dysfunction or action at other subtype 2 receptors. In particular, it has been suggested that 5-HT_{2C} plays a role in anxiety, depression, and affective disorders,[29,30] and more recently the noninduction of EPS by atypical antipsychotics has also been attributed to blockade of 5-HT_{2C}.[31] Unfortunately, elucidation of these possibilities has been hindered by the lack of any 5-HT_2-blocking compounds with high specificity for one or another subtype, especially as regards differentiation of 5-HT_{2B} and 5-HT_{2C}.

An alternative approach to the development of improved antipsychotics originated from the discovery and characterization of new types of DA receptors. Specifically, it has been reported that schizophrenia is associated with high levels of D_4 receptor,[32] leading to the hypothesis that it is this receptor rather than D_2 that should be blocked by an antipsychotic.[33] Furthermore, the affinities of clozapine and some other atypical antipsychotics for D_4 are several times their affinities for D_2, raising the possibility that their superiority to classical antipsychotics is due to more precise targeting of the relevant DA receptor, rather than or in addition to their action at the 5-HT_{2A} receptor. However, compounds such as L 745870[34] (**10**, Chart 2) or CP 293019[35] (**11**), which block D_4 almost exclusively, have proved ineffective in clinical trials (despite excellent performance in preclinical trials, including experiments on live animal models);[36] the D_2 antagonist remoxipride (**12**), which has little affinity for D_4, behaves like the least EPS-inducing classical antipsychotics, and Meltzer et al.[17,18] found that 13 atypical antipsychotics were distinguished from 12 classical antipsychotics by pK_i 5-HT_{2A}/D_2 ratios but not by pK_i 5-HT_{2A}/D_4 or pK_i D_2/D_4 ratios. In short, more work must be done to clarify the role of D_4 in cerebral activity.

Chart 2

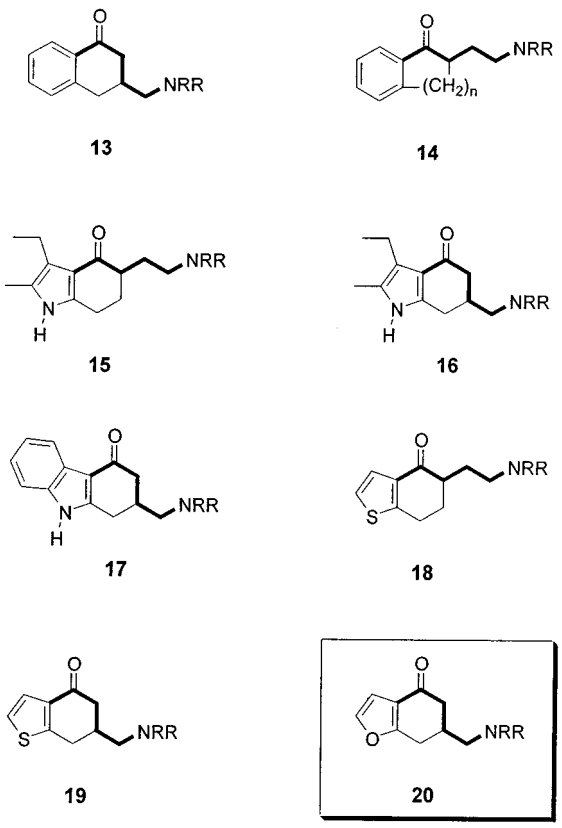


Despite the uncertainty about its mechanism of action, clozapine is still the most widely used atypical antipsychotic, and no other currently available agent appears to have an equal or superior spectrum of efficiency. However, its therapeutic use is severely limited by its being associated with increased risk of agranulocytosis and seizures;[37] likewise several cases of myocarditis and cardiomyopathy associated with clozapine treatment have been recently reported.[38] Accordingly, development of new antipsychotic agents remains a great challenge today.

In previous papers we have described the synthesis and atypical antipsychotic activities of 3-aminomethyltetralones (**13**, Chart 3) and 2-aminoethylbenzocycloalkanones (**14**),[39−42] which are conformationally restricted butyrophenone analogues of haloperidol; 5-aminoethyl- and 6-aminomethyl-4,5,6,7-tetrahydroindol-4-ones (**15**, **16**),[43−45] the former of which are butyrophenone homologues of the antipsychotic molindone; and 2-aminomethyl-1,2,3,9-tetrahydro-4*H*-carbazol-4-ones (**17**).[46,47] In all these series, the conformation of the butyrophenone butyl chain is restricted by its partial incorporation in a cycloalkanone ring fused to a benzene ring or a heterocycle. We have also recently described the synthesis, pharmacology, 3D QSARs, and molecular modeling of some aminoalkylthienocycloalkanones (**18**, **19**).[48]

In continuation of this work, we have now synthesized and pharmacologically evaluated some 6-aminomethyl-

Chart 3



4,5,6,7-tetrahydrobenzo[*b*]furan-4-ones (**20**; see Scheme 3 and Table 1). One of these compounds (**20b**) has two butyrophenone pharmacophores: the semirigid aminoalkyl furanone moiety and a 4-(*p*-fluorobenzoyl)piperidine fragment. The latter has been described as an antipsychotic pharmacophore with potency similar to that of linear butyrophenones,[49] and a SAR study of ketanserin analogues has suggested that the benzoyl carbonyl (present in **20b**,**h**,**i**) may be important for anchorage or orientation at the 5-HT_{2A} receptor.[50] Compound **20c** has the 4-(6-fluorobenzisoxazol-3-yl)-piperidine moiety of risperidone; 1,2-benzisoxazole is bioisosteric to benzoyl,[51] and 3-(4-piperazinyl)-6-hydroxybenzisoxazoles have also recently been reported to have potential as atypical antipsychotics.[52] The new compounds were all characterized pharmacologically by determination of their affinities for D_2, 5-HT_{2A}, and 5-HT_{2C} (in radioligand binding studies) and for 5-HT_{2B} (in functional studies); the most active were further characterized regarding their affinities for D_1 and D_4 and their effects on the behavior of experimental animals and on the levels of DA and its major metabolites in striatal tissue.

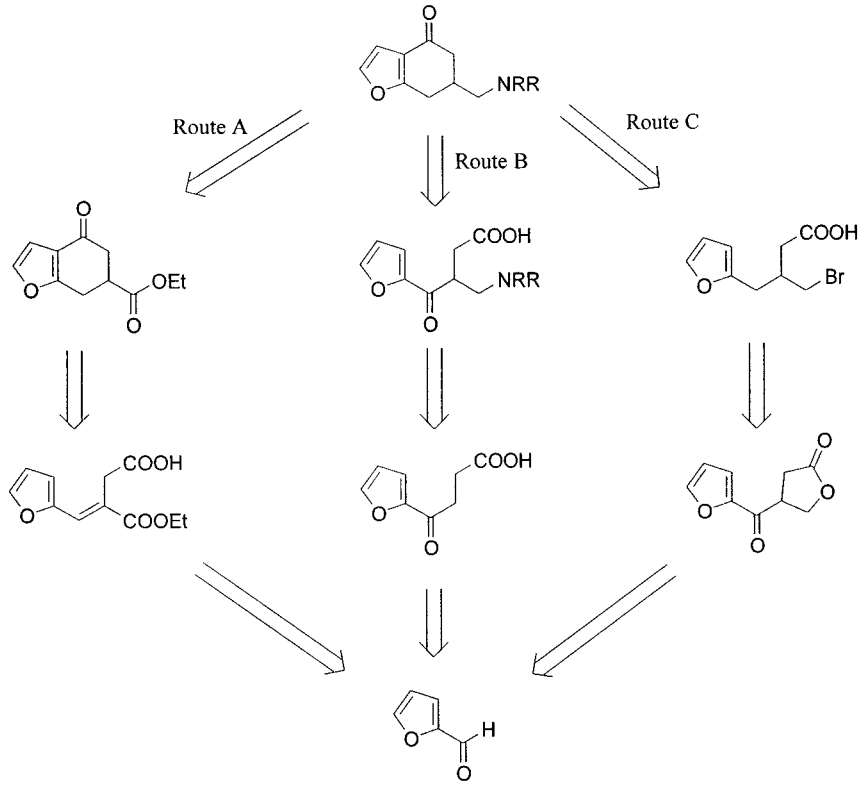
Chemistry

For preparation of the 6-aminomethyl-4-oxo-4,5,6,7-tetrahydrobenzo[*b*]furans **20a**−**i** we initially considered the routes shown in Schemes 1 and 2. Those shown in Scheme 1 all start from furfural: Route A involves Stobbe condensation with diethyl succinate, reduction of the double bond, cyclization, formation of an amide with the relevant −NRR moiety (Table 1), protection of the ketone carbonyl, and reduction of the amide carbonyl. Route B consists of conversion of furfural to *β*-(2-furoyl)propionic acid, formation of the Mannich base corresponding to the relevant −NRR, and final cyclization. Route C involves condensation of *β*-(2-furoyl)-propionic acid with formaldehyde, reduction of the resulting *β*-(2-furoyl)-*γ*-butyrolactone, opening of the lactone ring to obtain *γ*-bromo-*β*-furfurylbutyric acid, nucleophilic substitution of the bromine for the relevant −NRR moiety, and final cyclization. In the route shown in Scheme 2, the furan ring is constructed on a substituted cyclohexanone obtained by Birch reduction of 3,5-dimethoxybenzoic acid, subsequent reduction of the carboxyl group with a hydride, and acid hydrolysis of the resulting 1,4-dihydro-3,5-dimethoxybenzoic alcohol; condensation of the cyclohexanone with chloroacetaldehyde then affords a 6-substituted benzofuranone that can be transformed into amino ketones **20** by tosylation and nucleophilic substitution.

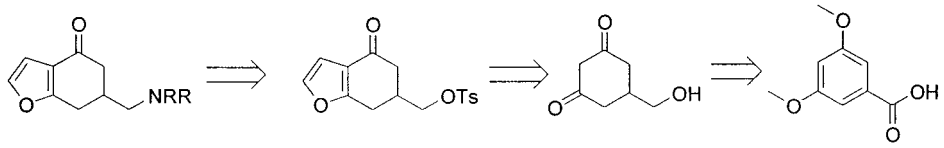
Route A was ruled out chiefly because of the long reaction times that in our experience have been needed for protection of the carbonyl in similar compounds, and routes B and C because of the difficulty in obtaining the common intermediate *β*-(2-furoyl)propionic acid. This latter cannot be prepared by straightforward succinoylation of furan or by reaction of diethyl malonate with 2-bromoacetylfuran because these procedures afford intractable resins. Although it can be obtained by hydrolysis of the corresponding nitrile or ester, which according to the literature (and in accordance with Scheme 1) is afforded by addition of acrylonitrile or methyl acrylate to furfural in the presence of sodium or potassium cyanide[53,54] or thiazolium salts,[55] the cyanide method has been reported to have moderate yields at best,[54] and our attempts at using 3-benzyl-5-(2-hydroxyethyl)-4-methylthiazolium as per Stetter and Kuhlmann[54] have provided very low yields of the target compound, the major product being furoin.

We accordingly decided to pursue the route shown in Scheme 2, which has the further advantage that 1,4-dihydro-3,5-dimethoxybenzoic alcohol (**21**)[56] is also a key intermediate in the preparation of a series of heterocyclic butyrophenones based on indole[45] and carbazole.[46,47] Alcohol **21** was hydrolyzed with 1 N HCl in tetrahydrofuran, the resulting cyclohexanone (**22**) was condensed with chloroacetaldehyde in the presence of $NaHCO_3$, and subsequent acid dehydration afforded the key intermediate 6-hydroxymethyl-4-oxo-4,5,6,7-tetrahydrobenzo[*b*]furan (**23**) in 60% yield from **21** (Scheme 3). Compound **23** was tosylated in 60% yield with *p*-toluenesulfonyl chloride in anhydrous pyridine, and reaction of the tosylate **24** with the amines of Table 1 in *N*-methyl-2-pyrrolidone (NMP) afforded 70−85% yields of compounds **20a** [HNRR = morpholine], **20b** [HNRR = 4-(*p*-fluorobenzoyl)piperidine], **20c** [HNNR = 4-(6-fluorobenzisoxazol-3-yl)piperidine], **20d** [HNNR = *N*-phenylpiperazine], **20e** [HNNR = *N*-(*o*-methoxyphenyl)piperazine], **20f** [HNNR = *N*-(2-pyridyl)piperazine], **20g** [HNNR = *N*-(2-pyrimidinyl)piperazine], and **25** [HNNR = *N*-*tert*-butoxycarbonylpiperazine] (Scheme 3). Cleavage of the *tert*-butoxycarbonyl group of **25** with trifluoroacetic acid in anhydrous dichloromethane then afforded piperazine **26** in 75% yield, and alkylation of **26** with 4-chloro-1,1-ethylenedioxy-1-(*p*-fluorophenyl)-butane in methyl isobutyl ketone containing potassium carbonate and catalytic amounts of potassium iodide,

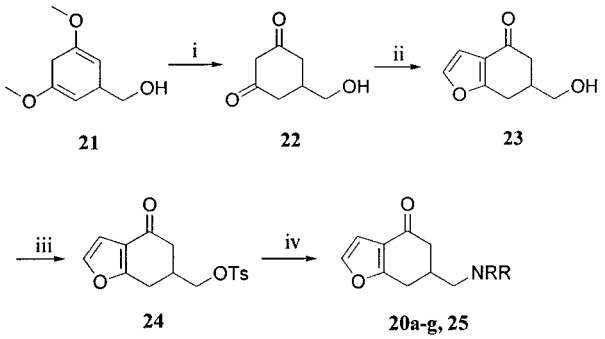
Scheme 1



Scheme 2



Scheme 3[a]



[a] Reagents: (i) HCl; (ii) $ClCH_2CHO$, $NaHCO_3$; (iii) Ts-Cl, Py; (iv) HNRR.

followed by acid hydrolysis of the ketal, afforded **20h** in 50% yield from **26**. Similarly, alkylation of **26** with 5-bromo-1-(*p*-fluorophenyl)-1-pentanone gave, via the same procedure, valerophenone **20i**, again in 50% yield.

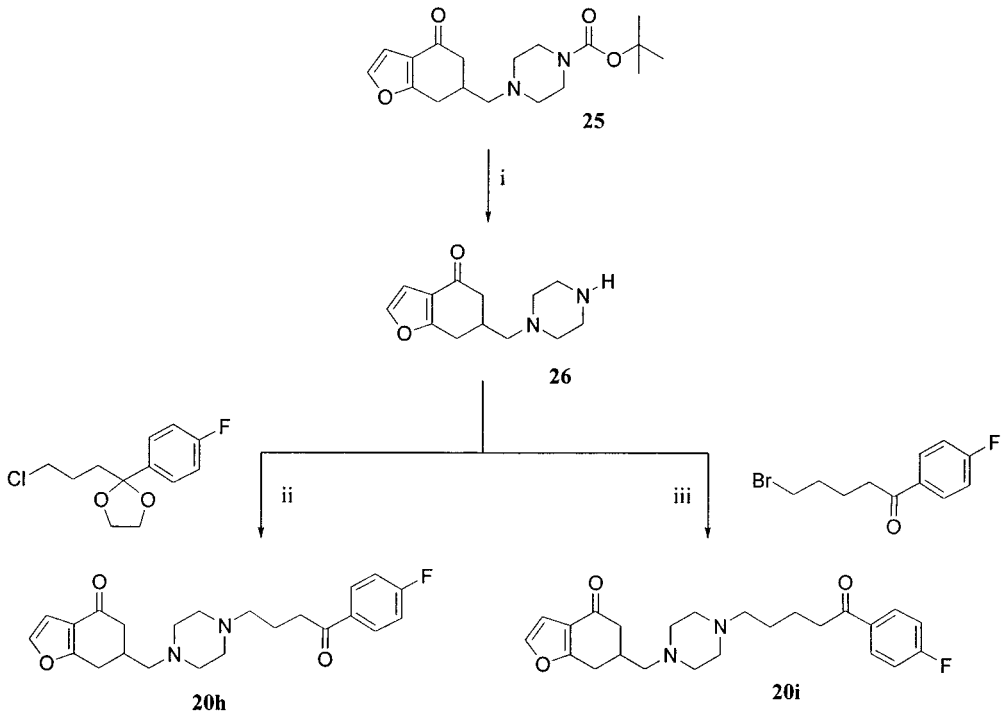
Results and Discussion

The results of the in vitro experiments are summarized in Table 2 and illustrated in Figures 1 and 2. The salient finding is that, regarding their interactions with serotonin receptors, both **20c**,**d** were selective for 5-HT_{2A}: the affinity of **20c** for 5-HT_{2A} was >23 times its affinity for 5-HT_{2B} and >1000 times its affinity for 5-HT_{2C} and **20d** had an affinity for 5-HT_{2A} >150 times its affinity for 5-HT_{2C} and was inactive for 5-HT_{2B} (pA_2 < 4). In addition, the affinities of **20i** for 5-HT_{2A} and 5-HT_{2B} were higher than for 5-HT_{2C}.

20c was the new compound with greatest affinity for D_1, D_2, 5-HT_{2A}, and 5-HT_{2B} (in this last case, jointly with **20b**), while the greatest affinity for 5-HT_{2C} was shown by the pyridylpiperazine **20f**. Affinity for D_1, D_2, and 5-HT_{2A} was reduced by replacing the benzisoxazolyl moiety of **20c** with the fluorobenzoyl group of **20b** and for D_2 and 5-HT_{2A} was reduced further by interpolating a flexible carbon chain between the nitrogenated ring and the benzoyl group as in **20h**, which also showed slightly reduced affinity for 5-HT_{2B}. The affinities for 5-HT_{2A}, D_2, and in most cases 5-HT_{2B} fell further still upon lengthening the benzoyl-bearing chain, as in **20i**, or on replacing the benzoylalkyl moiety with pyridyl, pyrimidinyl, or *o*-methoxyphenyl groups, as in **20e**−**g**, respectively (in particular, **20g** was inactive at 5-HT_{2A}, pK_i < 4). It appears that affinity for the D_2 subtype correlates with the presence of a piperidine moiety (compounds **20b**,**c**), while introduction of a piperazine (compounds **20d**−**i**) is associated with loss of affinity.

The affinity of **20c** for D_2 ($pK_i = 8.02$) was similar to that of risperidone (8.09), greater than that of clozapine (6.58), and less than that of haloperidol (8.48); see Figure 2. Its affinity for D_4 ($pK_i = 7.68$) was similar to that of clozapine (7.42), slightly smaller than that of haloperidol (8.20), and slightly greater than that of **20b**,

Scheme 4[a]



[a] Reagents: (i) TFA; (ii) (1) Na_2CO_3, KI, (2) HCl; (iii) Na_2CO_3, KI.

(7.04) (affinity for D_4 was only assayed for the new compounds with greatest affinity for D_2 and 5-HT_{2A}). Neither **20c** nor **20b** were as selective as haloperidol for D_2 versus D_1.

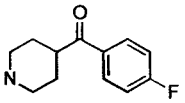
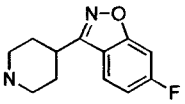
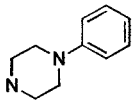
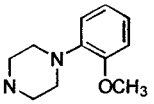
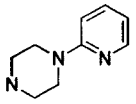
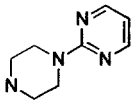
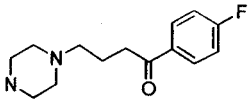
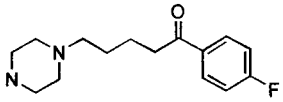
None of the new compounds had a pK_i 5-HT_{2A}/D_2 ratio greater than the Meltzer et al.[14,17] threshold for atypical antipsychotic activity.[1,12] It is also noteworthy that, like haloperidol, they all had low affinity for 5-HT_{2C}, for which the atypical antipsychotics clozapine and risperidone have high affinity. This result is in keeping with recent suggestions that it is 5-HT_{2C} blockade rather than 5-HT_{2A} blockade that can prevent the EPS induced by haloperidol (see Introduction).[31]

Experiments with rats or mice were carried out to evaluate the new compounds with greatest affinity for DA receptors (**20b**,**c**) for possible antipsychotic activity and induction of EPS. Antipsychotic potential was assayed in terms of reduction of spontaneous motor activity, *d*-amphetamine-induced hypermotility, and apomorphine-induced climbing behavior. Spontaneous activity was reduced, as expected,[57] by reference drugs haloperidol (2 mg/kg), clozapine (5 mg/kg), and risperidone (0.16−5.0 mg/kg) (Figure 3A) and also, dose-dependently, by both **20b,c** (ED_{50} = 1.21 and 0.48 mg/kg, respectively; see Figure 4A,B) which like risperidone generally exerted maximum effect within 20−30 min of administration (Figure 3B,C). The reference drugs and new compounds **20b**,**c** also slowed movement and inhibited rearing (data not shown).

The behavior of **20b**,**c** in the spontaneous activity tests correlates with their affinities for D_1, D_2, and D_4 but without further evidence is not necessarily attributable to action at these receptors; although D_1 and D_2 are known to be involved in the rapidity of initiation of movement and speed,[58] a reduction in spontaneous motor activity can be caused by many substances that are not DA antagonists including, for example, H_1-blockers, α_1-blockers, and reserpine (by induction of DA depletion), and is therefore by itself indicative only of general CNS depression. However, haloperidol (2 mg/kg), clozapine (5 mg/kg), risperidone (5.0 mg/kg), **20b** (4 mg/kg), and **20c** (0.5 mg/kg) also caused statistically significant reductions in amphetamine-induced hypermotility (Figure 5), speed, and rearing (data not shown). Amphetamine-induced hypermotility is attributed to the activation of D_2 receptors in the nucleus accumbens by DA released due to amphetamine-induced reversal of the DA re-uptake system.[59,60] However, effects on behavior are dose-dependent: while doses in the range 4.5−5 mg/kg induced maximum increase in hypermotility,[61] higher doses cause a predominance of stereotyped activity by inducing the release of DA in other pathways.[62] Results obtained with the *d*-amphetamine dosage used in this work (5 mg/kg) is a more specific sign of in vivo D_2-blocking activity, and although the hypermotility induced by low amphetamine doses (0.5 mg/kg) can be inhibited by both D_2 antagonist and 5-HT_{2A} antagonist,[63,64] which appear to prevent the 5-HT_{2A}-mediated activation of DA synthesis,[65−68] 5-HT_{2A} blockers are ineffective against amphetamine dosages > 2 mg/kg.[64]

Further corroboration of the in vivo DA antagonism of **20b**,**c** was provided by experiments in which they inhibited the effects of the direct D_1 and D_2 agonist apomorphine, which at low dosages causes hypomotility and yawning (due to activation of autoreceptors[69,70]) and at the high dosages used in this work (2 mg/kg) induces climbing by simultaneous activation of postsynaptic D_1 and D_2 receptors.[71−80] In keeping with the in vitro binding results on affinity for D_2 (Table 2), **20b**,**c** inhibited apomorphine-induced climbing more efficiently than clozapine but less efficiently than haloperidol or risperidone, and **20c** was more potent than **20b** (see Figure 6 and Table 3). However, the involvement

Table 1. 6-Aminomethyl-4,5,6,7-tetrahydrobenzo[*b*]furan-4-ones

compd	-NRR	mp (°C) base[a]	mp (°C) hydrochlor salt[b]	formula
20a		133-134	138-139	$C_{13}H_{17}NO_3$
20b		113-114	244-245	$C_{21}H_{22}FNO_3$
20c		131-132	271-272	$C_{21}H_{21}FN_2O_3$
20d		121-122	198-199	$C_{19}H_{22}N_2O_2$
20e		135-136	201-203	$C_{20}H_{24}N_2O_3$
20f		108-109	128-130	$C_{18}H_{21}N_3O_2$
20g		112-113	241-243	$C_{17}H_{20}N_4O_2$
20h		112-114	202-204	$C_{23}H_{27}FN_2O_3$
20i		92-94	235-240	$C_{24}H_{29}FN_2O_3$

[a] Recrystallized from *i*-PrOH. [b] Recrystallized from AcOEt.

Table 2. In Vitro Assays

compd	pK_i 5-HT_{2A}	pK_i 5-HT_{2C}	pK_i D_1	pK_i D_2	pK_i D_4	pK_i ratio 5-HT_{2A}/D_2	pA_2 5-HT_{2B}
20b (QF 1003B)	7.29 ± 0.20	33.3% (10^{-6} M)	6.71 ± 0.11	7.02 ± 0.14	7.04 ± 0.03	1.03	6.71 ± 0.57
20c (QF 1004B)	7.97 ± 0.03	4.74 ± 0.15	7.54 ± 0.06	8.02 ± 0.11	7.68 ± 0.19	0.99	6.61 ± 0.38
20d (QF 1007B)	7.66 ± 0.05	5.47 ± 0.08		<5			<4
20e (QF 1006B)	5.64 ± 0.11	4.89 ± 0.04		<5			6.19 ± 0.20
20f (QF 1008B)	5.08 ± 0.10	6.64 ± 0.02		<5			5.46 ± 0.03
20g (QF 1010B)	<4	<4		5.14 ± 0.25			5.74 ± 0.24
20h (QF 1005B)	6.02 ± 0.03	3.7% (10^{-6} M)		6.39 ± 0.12		0.94	6.36 ± 0.12
20i (QF 1011B)	5.90 ± 0.04	44.3% (10^{-6} M)		<5			6.23 ± 0.08
haloperidol	7.28 ± 0.03	5.34 ± 0.03	6.70 ± 0.08	8.48 ± 0.12	8.20 ± 0.03	0.93	
clozapine	7.56 ± 0.07	8.30 ± 0.02	6.77 ± 0.13	6.58 ± 0.05	7.42 ± 0.07	1.19	6.90 ± 0.2
risperidone	9.51 ± 0.03	7.38 ± 0.03		8.09 ± 0.29		1.20	

of factor(s) other than D_1 and D_2 blockade is suggested by the finding that risperidone was more potent than haloperidol despite having less affinity for both D_1 and D_2, and furthermore, risperidone was 4 times more potent than **20c** despite having quite similar affinity for D_2 and less affinity for D_1.[81] The putative additional factor is unlikely to be 5-HT_{2A} blockade, because although risperidone has at least 30 times more affinity for 5-HT_{2A} than any of the other drugs used, the selective 5-HT_{2A} blocker MDL 100907 has no effect on

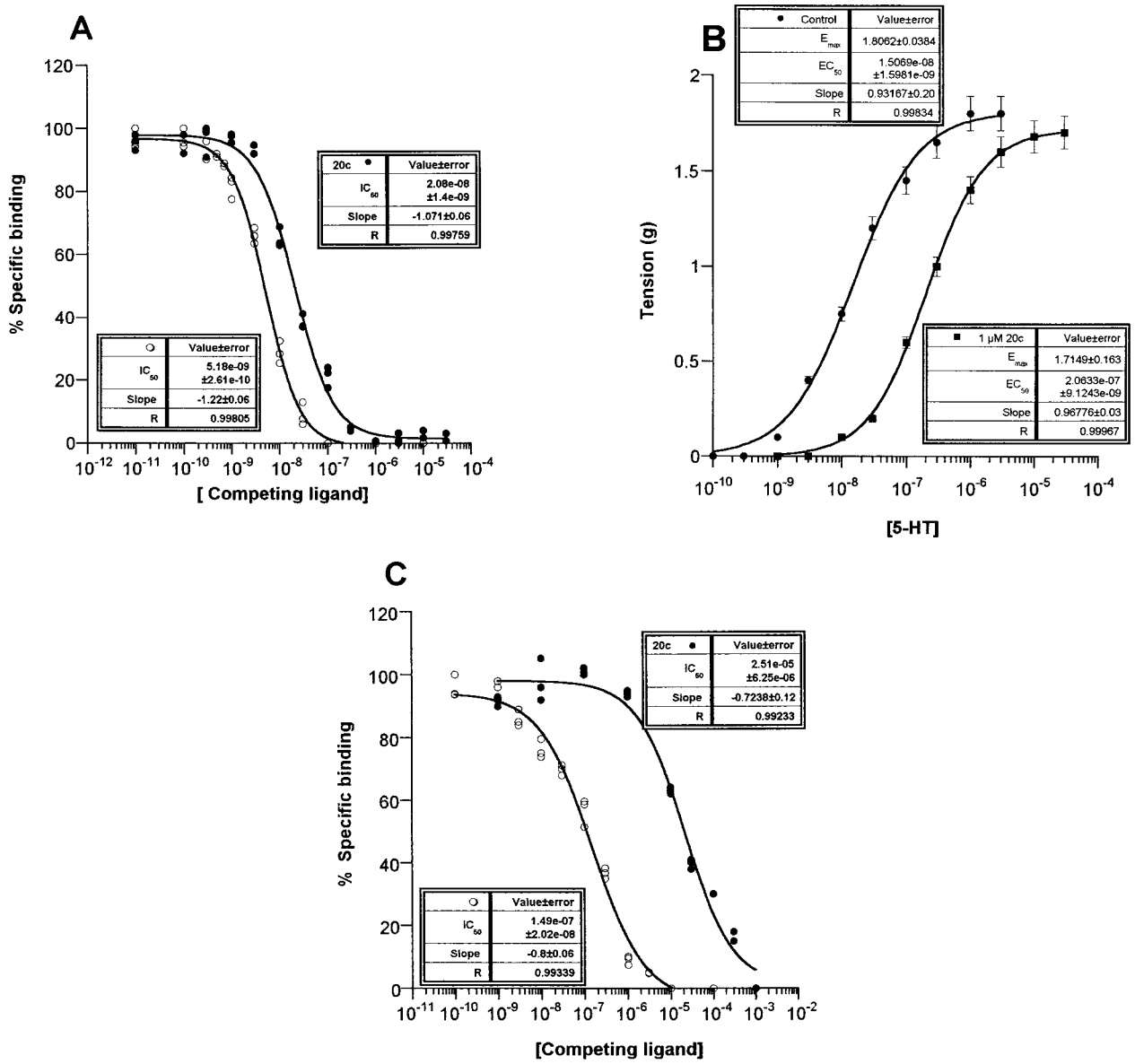


Figure 1. (A) Inhibition by **20c** (●) and risperidone (○) of [^{3}H]ketanserin binding by rat frontal cortex membrane preparations. (B) Inhibition by **20c** of serotonin-induced contrations of rat stomach fundus (curves from a single representative of 3−4 replicate experiments; vertical bars indicate SEM). (C) Inhibition by **20c** (●) and risperidone (○) of [^{3}H]mesulergine binding by bovine choroid plexus membrane preparations. Graphs A and C show triplicate points from a single experiment; a total of two replicate experiments were performed in each case.

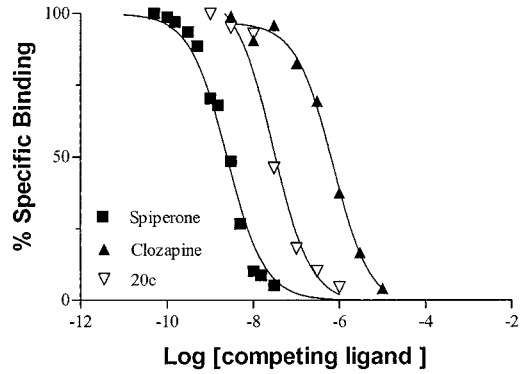


Figure 2. Inhibition by reference drugs and new compound **20c** of [^{3}H]spiperone binding by striatal D_2.

apomorphine-induced climbing at dosages at which it inhibits amphetamine-induced hypermotility,[82] whereas the ED_{50} value of **20c** for apomorphine-induced climbing, 0.34 mg/kg, was lower than the dose assayed in amphetamine-induced hypermotility (0.5 mg/kg). Additionally, clozapine, with affinity for 5-HT_{2A} similar to the new compounds, was less potent that **20c**.

If it is accepted that apomorphine-induced climbing is mediated chiefly by activation of mesolimbic DA receptors[83,84] and that blocking these receptors has antipsychotic effects because it blocks the mesolimbic dopaminergic system, which is functionally altered in schizophrenics, then the above results suggest that **20b**,**c** may have antipsychotic activity. To evaluate the risk of any such activity being accompanied by EPS, we assayed their ability to induce catalepsy in mice, catalepsy being attributed, like EPS, to blockade of striatal DA receptors.[85−87] Neither **20b** nor **20c** induced catalepsy at dosages lower than 2 mg/kg, but when higher dosages were assayed they exhibited ED_{50} values of respectively 3.44 mg/kg (95% CL 2.78−4.25) and 3.84

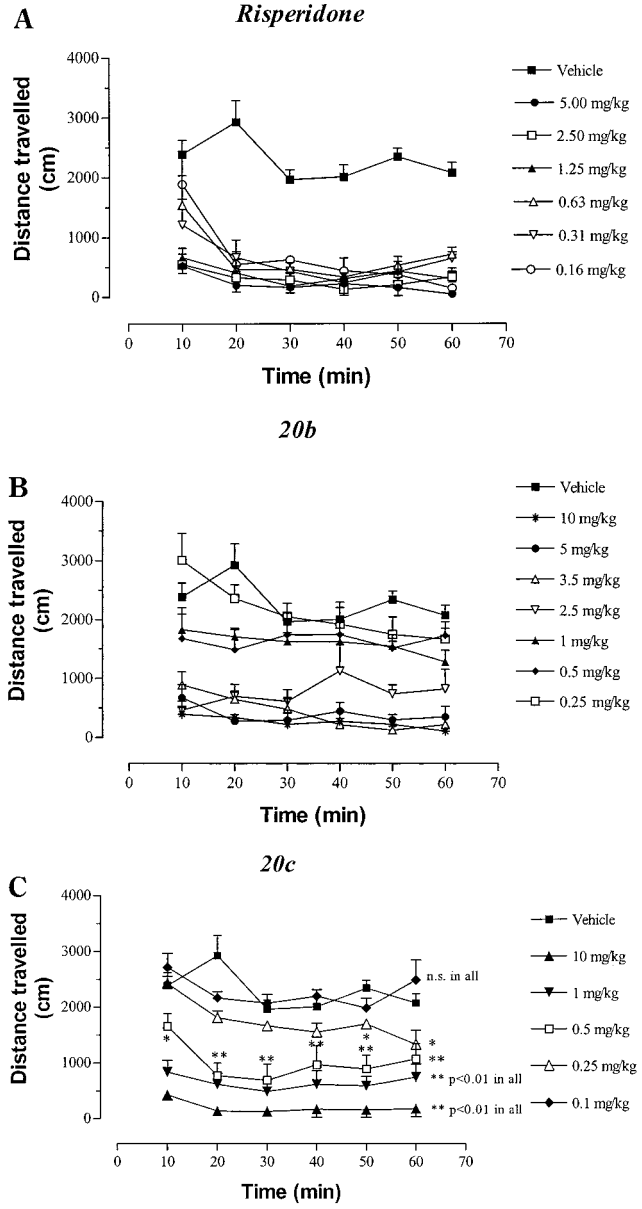


Figure 3. Time dependence of the effects of various drug dosages on the spontaneous activity of mice (means ± SEM): (A) risperidone, $p < 0.01$ with respect to vehicle at all dosages and times except after 10 min with 0.63 and 0.16 mg/kg, for which the difference was not significant; (B) **20b**, $p < 0.01$ with respect to vehicle with 10, 5, 3.5, and 2.5 mg/kg (except after 40 min with 2.5 mg/kg, not significant), not significant with 1 mg/kg except after 50 min ($p < 0.01$) and 60 min ($p < 0.05$), not significant with 0.5 mg/kg except after 20 and 50 min ($p < 0.05$ in both cases), not significant with 0.25 mg/kg; (C) **20c**, n.s. indicates no statistical significance with respect to vehicle, $*p < 0.05$, $**p < 0.01$.

mg/kg (95% CL 2.78−5.30) (Figure 7); these values are respectively 3.6 and 11.3 times the corresponding ED_{50} for inhibition of apomorphine-induced climbing (Table 3). The ED_{50} catalepsy/ED_{50} climbing ratio for haloperidol was 4.8, and the atypical antipsychotics used as reference drugs, clozapine and risperidone, induced no catalepsy even at dosages 20 times their ED_{50} values for climbing inhibition.

The action of **20c** in the striatum was confirmed by determination of the concentrations of DA and of its two major metabolites, DOPAC and HVA, in striatal tissue 2 h after administration. DA levels were not significantly affected by the reference drugs haloperidol and risperidone or the new compound **20c**. Clozapine, at the highest dosage used, increased the DA level. Metabolites, DOPAC and HVA, were significantly increased by all four compounds, risperidone especially (see Figure 8). These changes are thought to be due to the blockade of DA receptors giving rise to a compensatory increase in DA turnover that is reflected in the raised levels of the metabolites.[88−91]

Although **20c** fails to meet the Meltzer et al. criterion for atypical antipsychotic activity (pK_i 5-$HT_{2A}/D_2 > 1.12$), it nevertheless has a relative high ED_{50} catalepsy/ED_{50} climbing ratio. The cause may be its poor selectivity for D_2 versus D_1 (see above), since cataleptogenic potency may be favored by a high D_2 selectivity like that of haloperidol.[92] Likewise, the similarity between the D_2:D_1 selectivities of **20b**,**c** may explain their having very similar ED_{50} values for induction of catalepsy. Since neither **20b** nor **20c** have high affinities for 5-HT_{2C} or high pK_i 5-HT_{2A}/D_2 ratios, their being less cataleptogenic than haloperidol is thus attributable to a combination of lower affinity and lower selectivity for D_2, while their being more cataleptogenic than clozapine or risperidone may be attributed to their low affinity for 5-HT_{2C}.

In conclusion, the new compounds **20b**,**c** have moderate-to-high affinity for 5-HT_{2A}, D_1, D_2, and D_4 (**20c** especially), moderate affinity for 5-HT_{2B}, and low affinity for 5-HT_{2C}. In behavioral experiments with rats and mice they caused effects compatible with blockade of DA receptors in the limbic region (reduction of *d*-amphetamine-induced hypermotility and apomorphine-induced climbing) and the striatum (induction of catalepsy), and the striatal activity of **20c** was confirmed by determination of its effect on striatal levels of DA and its major metabolites. These results are in keeping with their pK_i 5-HT_{2A}/D_2 ratios, which are in the range of that for typical or classical antipsychotics. However, their induction of catalepsy less potently than haloperidol, which is also in keeping with their pK_i 5-HT_{2A}/D_2 ratios (1.03 and 0.99) and with their relatively poor selectivity for D_2 versus D_1, suggests relatively low capacity to induce EPS. The high selectivity of **20b**,**c** for 5-HT_{2A} and 5-HT_{2B} versus 5-HT_{2C} and of **20d** for 5-HT_{2A} and 5-HT_{2C} versus 5-HT_{2B} may make these compounds useful for researching the physiological and pharmacological roles of these receptors.

Experimental Section

Chemistry. Melting points were determined with a Kofler hot stage instrument or a Gallenkamp capillary melting point apparatus and are uncorrected. Infrared spectra were recorded with a Perkin-Elmer 1600 FTIR spectrophotometer; the main bands are given, in cm^{-1}. ^1H and ^{13}C NMR spectra were recorded with a Bruker WM AMX apparatus (300 MHz); chemical shifts are given in parts per million (δ) downfield from tetramethylsilane, and *J* values are given in hertz (Hz). Mass spectra were recorded on Kratos MS-50 or Varian Mat-711 mass spectrometer in fast atom bombardment (FAB) mode (with 2-hydroxyethyl disulfide as matrix) or by electron impact (EI). Optical rotations at the sodium D-line were determined in MeOH solutions of the indicated concentrations using a Perkin-Elmer 241 polarimeter. Flash column chromatography was performed using Kieselgel 60 (60−200 mesh; E. Merck AG, Darmstadt, Germany). Reactions were monitored by thin-layer chromatography (TLC) on Merck 60 GF_{254} chromatogram sheets using iodine vapor and/or UV light for detection; unless

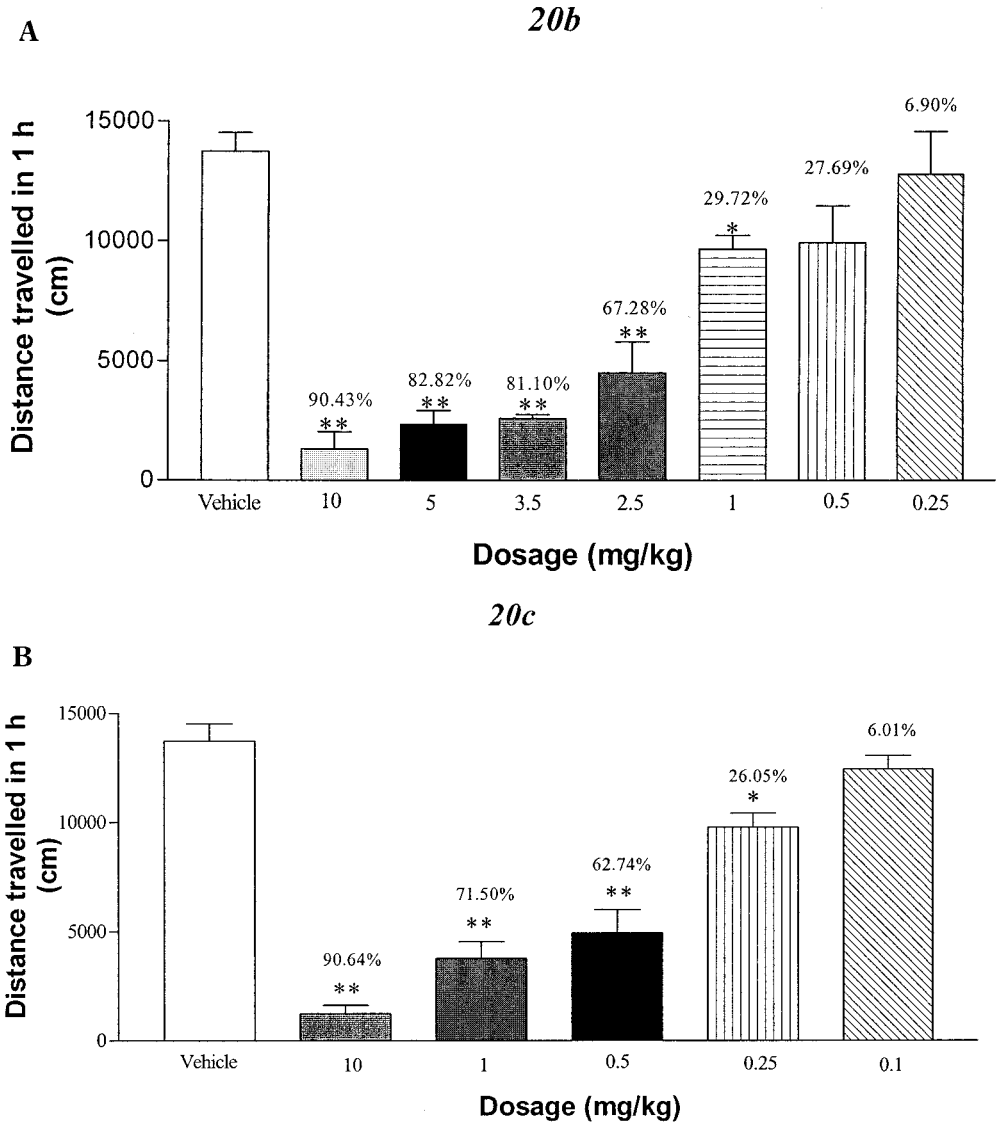


Figure 4. Total distance travelled in 1 h by mice treated with **20b** (A) or **20c** (B) and corresponding percent reduction in spontaneous locomotor activity. The data shown are means ± SEM. Significant differences with respect to controls are indicated by $*p < 0.05$ or $**p < 0.01$.

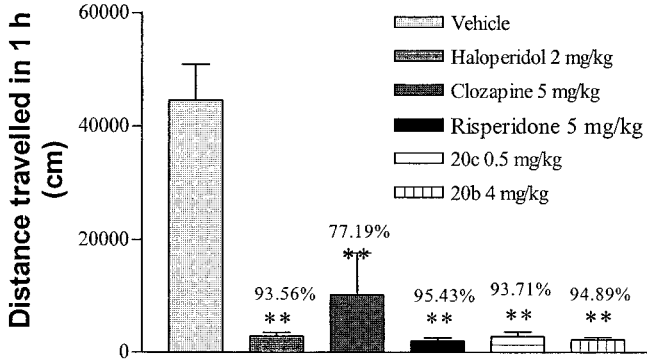


Figure 5. Inhibition of *d*-amphetamine-induced hypermotility in mice by haloperidol, clozapine, risperidone, **20b**, and **20c**: distances travelled in 1 h (means ± Sem) and percent reductions with respect to controls. All differences with respect to controls are statistically significant ($p < 0.01$).

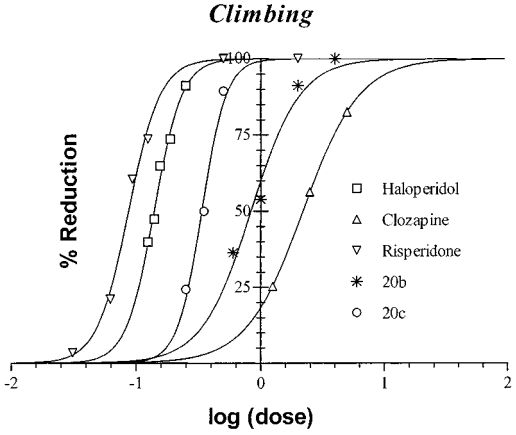


Figure 6. Dose−effect curves for inhibition of apomorphine-induced climbing by reference drugs and selected new compounds.

otherwise stated the purified compounds each showed a single spot. Elemental combustion analyses were performed on a Perkin-Elmer 240B apparatus at the Microanalysis Service of the University of Santiago de Compostela; unless otherwise stated all reported values are within ±0.4% of the theoretical compositions. Solvents were purified as per Vogel[93] by distillation over the drying agent under an argon atmosphere and were used immediately. The drying agents used were Na/benzophenone for THF, ether and toluene; P_2O_5 for CH_2Cl_2; K_2CO_3 for acetone and ethyl acetate; KOH for pyridine and

Table 3. In Vivo Assays

	ED_{50} (mg/kg)		rel potency (ED_{50} haloperidol/ED_{50} drug)		
compd	climbing (95% CL)	catalepsy	climbing	catalepsy	ED_{50} catalepsy/ED_{50} climbing
20b (QF 1003B)	0.95 (0.80−1.14)	3.44	0.15	0.19	3.62
20c (QF 1004B)	0.34 (0.18−0.61)	3.84	0.41	0.17	11.29
haloperidol	0.14 (0.13−0.15)	0.67	1.00	1.00	4.79
clozapine	2.21 (2.03−2.41)	>50.0	0.06		
risperidone	0.09 (0.08−0.09)	>2.0	1.56		

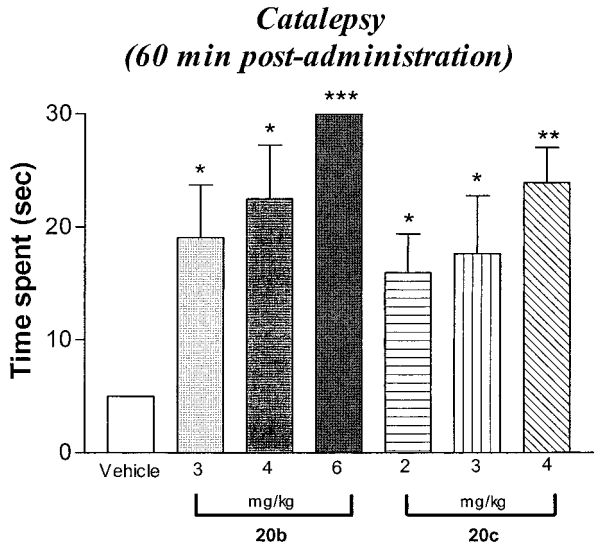


Figure 7. Results of catalepsy test, showing the time for which initial posture was maintained by mice treated with vehicle or new compounds (means ± SEM). Significant differences with respect to controls are indicated by *$p < 0.05$, **$p < 0.01$, or ***$p < 0.001$.

TEA; and $CaSO_4$/4 Å molecular sieves for DMF. Hydrochlorides were prepared by dropwise addition, with cooling, of a saturated solution of HCl in anhydrous ether to a solution of the amine in anhydrous ether or absolute ethanol/ether.

6-Hydroxymethyl-4,5,6,7-tetrahydrobenzo[*b*]furan-4-one (23). A solution of alcohol **21**[56] (1 g, 6.84 mmol) in a mixture of THF (30 mL) and 1 N HCl (6 mL) was stirred at room temperature for 2 h and then concentrated in vacuo, giving 0.9 g of a yellow oil that was identified as alcohol **22** and was used without further purification in the next step, as follows. A solution of **22** (0.9 g, 6.33 mmol) in 6 mL of water was added at a rate of 0.4 mL/min, with stirring, to a mixture of 50% chloroacetaldehyde (1.01 mL), $NaHCO_3$ (0.63 g, 7.53 mmol) and water (5 mL) maintained at 0−5 °C. Stirring was continued at room temperature for 12 h, AcOEt (5 mL) was added, followed by enough 10% HCl to bring the mixture to pH 1, and after stirring for a further 1 h the organic layer was separated, washed with 10% K_2CO_3, dried (Na_2SO_4) and concentrated to dryness, affording 0.73 g of an oil that upon column chromatography with 4:1 AcOEt/hexane as eluant gave 0.61 g of an oil identified as the hydroxy ketone **23**: yield 60%. IR: 3394, 2935, 1668, 1598. ^{1}H NMR ($CDCl_3$): δ 2.37−2.43 (m, 1H, $1H_5$); 2.50−2.60 (m, 2H, $1H_5$, $1H_7$); 2.75 (dd, 1H, J = 17.2, 9.4, $1H_7$); 3.01 (dd, J = 17.2, 4.8, H_6); 3.66−3.70 (m, 2H, CH_2-OH); 6.63 (d, 1H, J = 1.9, H_2); 7.31 (d, 1H, J = 1.8, H_3). ^{13}C NMR ($CDCl_3$): δ 26.6 (C_7); 38.4 (C_6); 40.9 (C_5); 65.8 (-*C*H_2-OH); 106.7 (C_3); 121.2 (C_{3a}); 143.6 (C_2); 167.3 (C_{7a}); 194.8 (-CO-). MS (FAB, *m*/*z*): 167 (MH^+). Anal. ($C_9H_{10}O_3$) C, H.

6-(*p*-Toluenesulfonyl)oxymethyl-4,5,6,7-tetrahydrobenzo[*b*]furan-4-one (24). *p*-Toluenesulfonyl chloride (2.3 g, 12 mmol) was added to a solution of alcohol **23** (1.6 g, 9.64 mmol) in dry pyridine (30 mL) at 0 °C, and the mixture was stirred at this temperature for 24 h. After addition of water (20 mL) and extraction with CH_2Cl_2, the organic layer was washed with water, dried over Na_2SO_4 and concentrated to dryness, affording 2 g of a brown dark oil that upon column chromatography with 1:1 AcOEt/hexane as eluant gave 1.84 g of a white solid identified as the tosylate **24**: yield 60%, mp 104−105 °C (*i*-PrOH). IR: 2957, 1679, 1593. ^{1}H NMR ($CDCl_3$): δ 2.27−3.24 (m, 1H, $1H_5$); 2.47 (s, 3H, -CH_3); 2.50−2.52 (m, 1H, $1H_5$); 2.69−2.78 (m, 2H, $1H_7$, H_6); 3.01 (dd, 1H, J = 15.6, 3.7, $1H_7$); 3.99−4.10 (m, 2H, -C*H*$_2$-OTs); 6.62 (d, 1H, J = 2 Hz, H_2); 7.32 (d, 1H, J = 1.9, H_3); 7.35 (d, 2H, J = 8.22, H_3, H_5 Ph), 7.78 (d, 2H, J = 8.3, H_2, H_6 Ph). ^{13}C NMR ($CDCl_3$): δ 22.1 (-CH_3); 26.4 (C_7); 35.5 (C_6); 40.4 (C_5); 72.1 (-CH_2-OTs); 106.8 (C_3); 121.4 (C_{3a}); 128.3 (C_3 Ph); 130.4 (C_2 Ph); 132.9 (C_1 Ph); 143.7 (C_2); 145.6 (C_4 Ph); 165.5 (C_{7a}); 192.06 (-CO-). MS (FAB, *m*/*z*): 321 (MH^+). Anal. ($C_{16}H_{16}O_5S$) C, H, S.

6-Morpholinylmethyl-4,5,6,7-tetrahydrobenzo[*b*]furan-4-one (20a). A solution of tosylate **24** (0.6 g, 1.87 mmol) and morpholine (0.33 g, 3.75 mmol) was stirred in NMP (15 mL) for 16 h at 85 °C. The solvent was removed in vacuo, and the residue was dissolved in CH_2Cl_2. This solution was washed twice with water, dried (Na_2SO_4), and the solvent was removed in vacuo, affording an orange solid that upon column chromatography with AcOEt as eluant gave **20a** (0.33 g, 75%) as a white crystalline solid of mp 133−134 °C (*i*-PrOH). IR: 2958, 1673. ^{1}H NMR ($CDCl_3$): δ 2.21−2.30 (m, 1H, $1H_5$); 2.36−2.51 (m, 6H, $1H_5$, $1H_7$, -N(C*H*$_2$-CH_2)$_2$O); 2.52−2.68 (m, 3H, H6, $1H_7$, -HC*H*-N<); 3.03−3.08 (m, 1H, -*H*CH-N<); 3.69 (t, 4H, J = 4.6, -N(CH_2-C*H*$_2$)$_2$O); 6.65 (d, 1H, J = 1.7, H_2); 7.30 (d, 1H, J = 1.7, H_3). ^{13}C NMR ($CDCl_3$): δ 28.4 (C_7); 33.3 (C_6); 42.9 (C_5); 54.4 (-N(*C*H_2-CH_2)$_2$O); 63.6 (-CH_2-N<); 67.4 (-N(CH_2-*C*H_2)$_2$O); 106.8 (C_3); 121.4 (C_{3a}); 143.3 (C_2); 166.9 (C_{7a}); 194.1 (-CO-). MS (FAB, *m*/*z*): 236 (MH^+). Anal. ($C_{13}H_{17}NO_3$) C, H, N. Hydrochloride: mp 138−139 °C (AcOEt).

Compounds **20b**−**g** and **25** were prepared similarly, with the following results.

6-[4-(*p*-Fluorobenzoyl)piperidin-1-ylmethyl]-4,5,6,7-tetrahydrobenzo[*b*]furan-4-one (20b): yield 75%, mp 113−114 °C (*i*-PrOH). IR: 2949, 2768, 1671, 1595. ^{1}H NMR ($CDCl_3$): δ 1.81−1.83 (m, 4H, -N(CH_2-C*H*$_2$)$_2$C-); 2.05−2.46 (m, 5H, $1H_7$, -C*H*$_2$-N<, -N(*H*CH-CH_2)$_2$CH-); 2.51−2.99 (m, 5H, $1H_5$, $1H_6$, $1H_7$, -N(HC*H*-CH_2)$_2$CH-); 3.04−3.21 (m, 2H, H_5, -N(HCH-CH_2)$_2$C*H*-); 6.4 (d, 1H, J = 2.02, H_2); 7.13 (t, 2H, J = 6.6, *o*-F-Ph); 7.3 (d, 1H, J = 1.9, H_3); 7.96 (dd, 2H, J = 5.3, 2, *m*-F-Ph). ^{13}C NMR ($CDCl_3$): δ 28.4 (C_7); 29.1 (-N(CH_2-*C*H_2)$_2$CH-); 29.2 (-N(CH_2-*C*H_2)$_2$CH-); 33.9 (C_6); 43.1 (C_5); 43.9 (-N(CH_2-*C*H_2)$_2$CH-); 53.4 (-N(*C*H_2-CH_2)$_2$CH-); 54.6 (-N(*C*H_2-CH_2)$_2$CH-); 63.4 (-CH_2-N<); 106.8 (C_3); 116.2 (d, 2C, J = 21.8, *o*-F-Ph); 121.4 (C_{3a}); 131.2 (d, 2C, J = 9.3, *m*-F-Ph); 132.8 (*p*-F-Ph); 143.3 (C_2); 167.2 (C_{7a}); 166.0 (d, 1C, J = 250.7, C-F); 194.2 (-CO-); 201.4 (-CO-). MS (FAB, *m*/*z*): 356 (MH^+). Anal. ($C_{21}H_{22}FNO_3$) C, H, N. Hydrochloride: mp 244−245 °C (AcOEt).

6-[4-(6-Fluorobenzisoxazol-3-yl)piperidin-1-ylmethyl]-4,5,6,7-tetrahydrobenzo[*b*]furan-4-one (20c): yield 85%, mp 131−132 °C (*i*-PrOH). IR: 2927, 2739, 1665, 1610. ^{1}H NMR ($CDCl_3$): δ 2.03−2.38 (m, 8H, -C*H*$_2$-N(CH_2-C*H*$_2$)$_2$CH-, $1H_7$, H_6); 2.40−2.59 (m, 1H, $1H_7$); 2.61−2.99 (m, 3H, $1H_5$, -N(*H*CH-CH_2)$_2$CH-); 3.04−3.51 (m, 4H, $1H_5$, -N(HC*H*-CH_2)$_2$C*H*-); 6.67 (d, 1H, J_{C-F} = 1.9, H_2); 7.11 (dt, 1H, J_{C-F} = 8.8, 2.5, $H_{5'}$); 7.23−7.26 (m, 1H, $1H_7$); 7.33 (d, 1H, J = 1.9, H_3); 7.69 (dd, 1H, J_{C-F} = 8.7, 5.1, $H_{4'}$). ^{13}C NMR ($CDCl_3$): δ 28.5 (C_7), 30.9 (-N(CH_2-*C*H_2)$_2$CH-); 31.4 (-N(CH_2-*C*H_2)$_2$CH-); 33.9 (C_6); 34.9 (-N(CH_2-CH_2)$_2$CH-); 43.0 (C_5); 53.9 (-N(*C*H_2-CH_2)$_2$CH-); 54.7 (-N(CH_2-CH_2)$_2$CH-); 63.5 (-CH_2-N<); 97.8 (d, J_{C-F} = 26.8, $C_{7'}$); 106.8 (C_3); 112.7 (d, J_{C-F} = 25.3, $C_{5'}$); 117.7 ($C_{3a'}$); 121.5 (C_{3a}); 122.9 (d, J_{C-F} = 11.1, $C_{4'}$); 143.3 (C_2); 161.5 ($C_{3'}$); 164.5 (d, J = 250.7, $C_{6'}$); 164.3 (d, J = 13.8, $C_{7a'}$); 167.2 (C_{7a}); 194.3 (-CO-). MS (FAB,

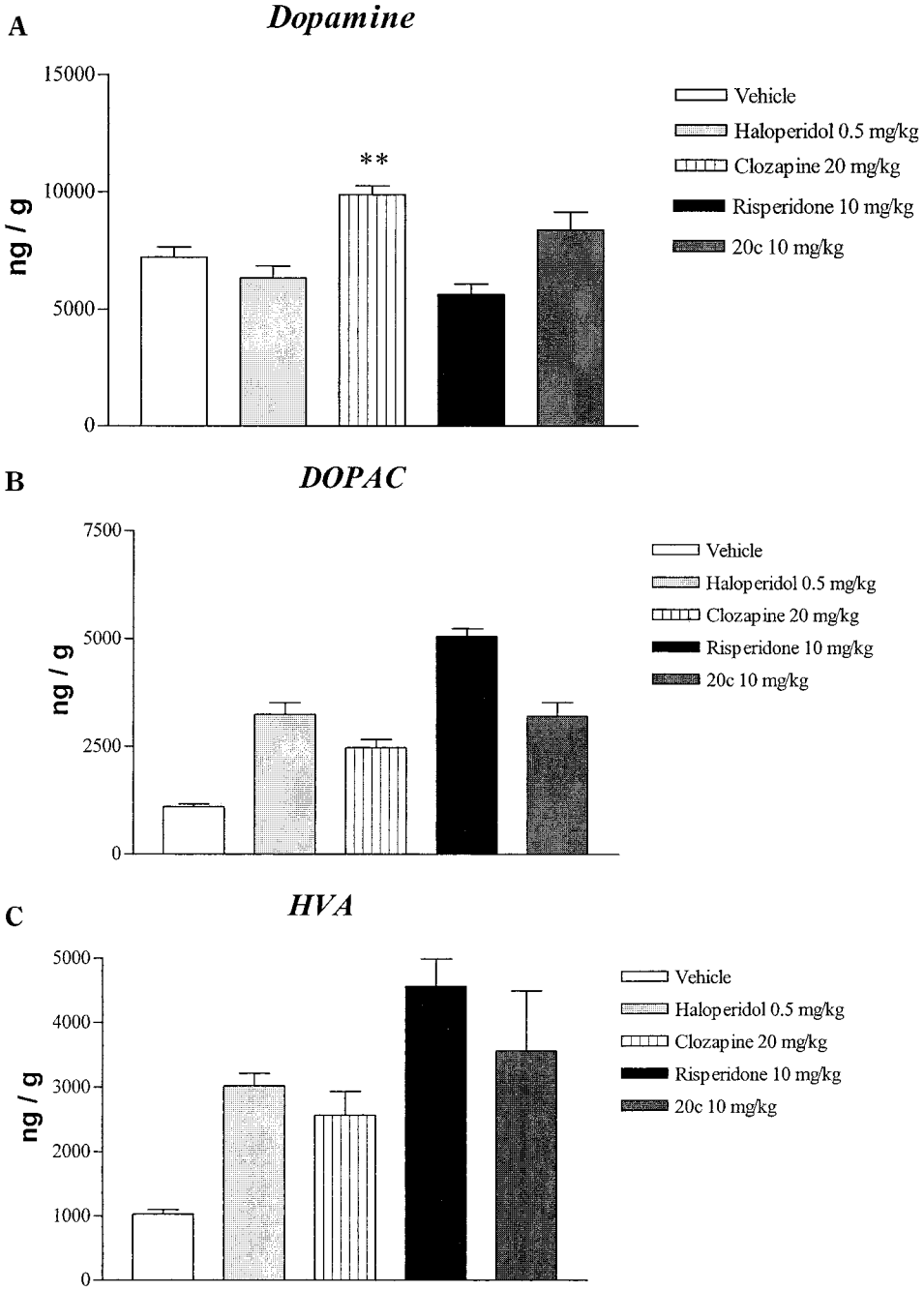


Figure 8. Effects of reference drugs and **20c** on striatal DA, DOPAC, and HVA levels (means ± SEM): (A) DA, only for clozapine (20 mg/kg) is the difference with respect to vehicle statistically significant ($p < 0.01$); (B) DOPAC, $p < 0.01$ with respect to vehicle for all drugs; (C) HVA, $p < 0.01$ with respect to vehicle for all drugs.

m/*z*): 369 (MH^+). Anal. ($C_{21}H_{21}FN_2O_3$) C, H, N. Hydrochloride: mp 271−272 °C (AcOEt). Anal. ($C_{21}H_{21}FN_2O_3$·HCl) C, H, N.

6-(4-Phenylpiperazin-1-ylmethyl)-4,5,6,7-tetrahydrobenzo[*b*]furan-4-one (20d): yield 75%, mp 121−122 °C (*i*-PrOH). IR: 3852, 1673. ^{1}H NMR ($CDCl_3$): δ 2.24−2.62 (m, 9H, $2H_5$, $1H_7$, -N(CH_2-CH_2)$_2$N-, -CH_2-N<); 2.68 (dd, 1H, J = 15, 3.6, $1H_7$); 3.08−3.15 (m, 1H, $1H_6$); 3.19 (t, 4H, J = 5, -N(CH_2-CH_2)$_2$N-); 6.67 (d, 1H, J = 2, H_2); 6.83−6.94 (m, 3H, *o*-Ph, *p*-Ph); 7.23−7.29 (m, 2H, *m*-Ph); 7.3 (d, 1H, J = 2, H_3). ^{13}C NMR ($CDCl_3$): δ 28.5 (C_7); 33.7 (C_6); 43.0 (C_5); 49.6 (-N(CH_2-CH_2)$_2$N-); 53.9 (-N(CH_2-CH_2)$_2$N-); 63.2 (-CH_2-N<); 106.8 (C_3); 116.8 ($C_{4'}$); 120.1 ($C_{3'}$); 121.5 (C_{3a}); 129.5 ($C_{2'}$); 143.3 (C_2); 151.6 (C_{7a}); 167.1 ($C_{1'}$); 194.2 (-CO-). MS (FAB, *m*/*z*): 311 (MH^+). Anal. ($C_{19}H_{22}N_2O_2$): C, H, N. Hydrochloride: mp 198−199 °C (AcOEt). Anal. ($C_{19}H_{22}N_2O_2$·2HCl·H_2O) C, H, N.

6-[4-(*o*-Methoxyphenyl)piperazin-1-ylmethyl]-4,5,6,7-tetrahydrobenzo[*b*]furan-4-one (20e): yield 70%, mp 135−136 °C (*i*-PrOH). IR: 2934, 2811, 1656, 1591. ^{1}H NMR ($CDCl_3$): δ 2.27−2.33 (m, 1H, $1H_5$); 2.43−2.47 (m, 2H, $1H_5$, $1H_7$); 2.58−2.70 (m, 8H, $1H_7$, $1H_6$, -CH_2-N(CH_2-CH_2)$_2$N-); 3.85 (s, 4H, -N(CH_2-CH_2)$_2$N-); 3.85 (s, 3H, -OCH_3); 6.66 (d, 1H, J = 2.01, H_2); 6.91−6.99 (m, 4H, Ph); 7.32 (d, 1H, J = 1.9, H_3). ^{13}C NMR ($CDCl_3$): δ 28.5 (C_7); 33.7 (C_6); 43.0 (C_5); 51.1 (-N(CH_2-CH_2)$_2$N-); 54.2 (N(CH_2-CH_2)$_2$N-); 55.7 (-OCH_3); 63.3 (-CH_2-N<); 106.8 (C_3); 111.6 ($C_{6'}$); 118.6 ($C_{3'}$); 121.4 ($C_{5'}$); 121.5 (C_{3a}); 123.3 ($C_{4'}$); 141.7 ($C_{1'}$); 143.2 (C_2); 152.7 ($C_{2'}$); 167.2 (C_{7a}); 194.2 (-CO-). MS (FAB, *m*/*z*): 341 (MH^+). Anal. ($C_{20}H_{24}N_2O_3$) C, H, N. Hydrochloride: mp 201−203 °C (AcOEt). Anal. ($C_{20}H_{24}N_2O_3$·2HCl) C, H, N.

6-[4-(2-Pyridyl)piperazin-1-ylmethyl]-4,5,6,7-tetrahydrobenzo[*b*]furan-4-one (20f): yield 80%, mp 108−109 °C (*i*-PrOH). IR: 2826, 1677, 1592. ^{1}H NMR ($CDCl_3$): δ 2.24−2.64 (m, 9H, $2H_5$, $1H_7$, -CH_2-N(CH_2-CH_2)$_2$N-); 2.67 (dd, 1H, J = 13.4, 4.5, $1H_7$); 3.04−3.15 (m, 1H, H_6), 3.51 (t, 4H, J = 5.1, -N(CH_2-CH_2)$_2$N-); 6.61 (d, 1H, J = 2.2, H_2); 6.63−6.66 (m, 2H,

H_3, $H_{5'}$); 7.32 (d, 1H, $J = 2.0$, H_3); 7.46 (dd, 1H, $J = 8.5, 5.1$, $H_{4'}$); 8.17 (dd, 1H, $J = 4.8, 1.2$, $H_{6'}$). ^{13}C NMR ($CDCl_3$): δ 28.5 (C_7); 33.6 (C_6); 42.9 (C_5); 45.6 (-N(CH_2-CH_2)$_2$N-); 53.7 (-N(CH_2-CH_2)$_2$N-); 63.3 (-CH_2-N<); 106.8 (C_3); 107.4 ($C_{3'}$); 113.7 ($C_{5'}$); 121.5 (C_{3a}); 137.8 ($C_{4'}$); 143.3 (C_2); 148.34 ($C_{6'}$); 159.9 ($C_{2'}$); 167.1 (C_{7a}); 194.1 (-CO-). MS (FAB, *m/z*): 312 (MH^+). Anal. ($C_{18}H_{21}N_3O_2$) C, H, N. Hydrochloride: mp 128−130 °C (AcOEt).

6-[4-(2-Pyrimidinyl)piperazin-1-ylmethyl]-4,5,6,7-tetrahydrobenzo[*b*]furan-4-one (20g): yield 70%, mp 112−113 °C (*i*-PrOH). IR: 2826, 1677, 1592. ^{1}H NMR ($CDCl_3$): δ 2.23−2.69 (m, 10H, $2H_5$, $2H_7$, -CH_2-N(CH_2-CH_2)$_2$N-); 3.04−3.14 (m, 1H, H_6), 3.80 (t, 4H, $J = 5.1$, -N(CH_2-CH_2) $_2$N-); 6.46 (t, 1H, $J = 4.7$, $H_{5'}$); 6.65 (d, 1H, $J = 2.0$, H_2); 7.32 (d, 1H, $J = 2.0$, H_3); 8.28 (d, 2H, $J = 4.7$, $H_{6'}$, $H_{4'}$). ^{13}C NMR ($CDCl_3$): δ 28.5 (C_7); 33.7 (C_6); 43.0 (C_5); 44.1 (-N(CH_2-CH_2)$_2$N-); 53.8 (-N(CH_2-CH_2)$_2$N-); 63.3 (-CH_2-N<); 106.8 (C_3); 110.2 ($C_{5'}$); 121.5 (C_{3a}); 143.3 (C_2); 158.1 ($C_{4'}$, $C_{6'}$); 162.0 ($C_{2'}$); 167.0 (C_{7a}); 194.1 (-CO-). MS (FAB, *m/z*): 313 (MH^+). Anal. ($C_{17}H_{20}N_4O_2$) C, H, N. Hydrochloride: mp 241−243 °C (AcOEt). Anal. ($C_{17}H_{20}N_4O_2$·2HCl) C, H, N.

6-[4-(*tert*-Butoxycarbonyl)piperazin-1-ylmethyl]-4,5,6,7-tetrahydrobenzo[*b*]furan-4-one (25): yield 70%, mp 86−87 °C (*i*-PrOH). IR: 2918, 1693, 1681. ^{1}H NMR ($CDCl_3$): δ 1.45 (s, 9H, 3x -CH_3); 2.30−2.38 (m, 7H, -N(CH_2-CH_2)$_2$N-, $2H_5$, $1H_7$); 2.40−2.67 (m, 3H, -CH_2N<, $1H_7$); 3.03−3.08 (m, 1H, H_6); 3.40 (t, 4H, $J = 5$,.N(CH_2-CH_2)$_2$N-); 6.65 (d, 1H, $J = 2$, H_2); 7.31 (d, 1H, $J = 2$, H_3). ^{13}C NMR ($CDCl_3$): δ 28.4 (C_7); 28.8 (3x -CH_3); 33.6 (C_6); 42.9 (C_5); 44.2 (-N(CH_2-CH_2)$_2$N-); 53.7 (-N(CH_2-CH_2)$_2$N-); 63.2 (-CH_2N<); 80.1 (-C(CH_3)$_3$); 106.8 (C_3); 121.5 (C_{3a}); 143.3 (C_2); 155.1 (-COO-); 166.9 (C_{7a}); 194.1 (-CO-). MS (FAB, *m/z*): 335 (MH^+). Anal. ($C_{18}H_{26}N_2O_4$) C, H, N.

6-Piperazin-1-ylmethyl-4,5,6,7-tetrahydrobenzo[*b*]furan-4-one (26). Trifluoroacetic acid (2.87 mL, 37.4 mmol) was added to a solution of amine **25** (1.25 g, 3.74 mmol) in dry CH_2Cl_2 (10 mL), the mixture was stirred at room temperature for 20 min, and the solvent was then removed under reduced pressure. The residue was dissolved in CH_2Cl_2, and this solution was washed with 10% $NaHCO_3$ (2 × 20 mL). The organic phase was dried over Na_2SO_4, and the solvent was removed in vacuo, affording **26** (0.65 g, 75%) as a white crystalline solid: mp 128−130 °C (*i*-PrOH). IR: 3342, 2938, 1681, 1596. ^{1}H NMR ($CDCl_3$): δ 2.25−2.53 (m, 10H, $2H_7$, $2H_5$, -CH_2-N(CH_2-CH_2)$_2$NH); 2.75 (t, 4H, $J = 4.9$, -N(CH_2-CH_2)$_2$NH); 2.93 (m, 1H, H_6); 6.4 (d, 1H, $J = 2$, H_2); 7.3 (d, 1H, $J = 2$, H_3); 8.25 (s, 1H, -NH). ^{13}C NMR ($CDCl_3$): δ 28.4 (C_7); 33.3 (C_6); 42.9 (C_5); 43.8 (-N(CH_2-CH_2)$_2$NH); 54.0 (-N(CH_2-CH_2)$_2$NH); 63.7 (-CH_2-N<); 106.8 (C_3); 121.4 (C_{3a}); 142.3 (C_2); 165.7 (C_{7a}); 198 2 (-CO-). MS (FAB, *m/z*): 235 (MH^+). Anal. ($C_{13}H_{18}N_2O_2$) C, H, N. Hydrochloride: mp 213−214 °C (AcOEt).

6-{4-[4-(4-Fluorophenyl)-4-oxobutyl]piperazin-1-ylmethyl}-4,5,6,7-tetrahydrobenzo[*b*]furan-4-one (20h). A solution of 4-chloro-1,1-ethylenedioxy-1-(4-fluorophenyl)butane (0.55 g, 2.23 mmol) in methyl isobutyl ketone (15 mL) was added under argon, with stirring, to a solution of the amine **26** (0.5 g, 2.13 mmol), anhydrous Na_2CO_3 (0.65 g) and KI (0.17 g) in the same solvent (45 mL), and the mixture was refluxed for 36 h. The precipitate was filtered out, the solvent was removed from the filtrate under reduced pressure, and a solution of the resulting residue in 10% HCl was refluxed for 1 h (100 °C). Once cool, this solution was brought to basic pH with 10% NaOH and extracted with AcOEt (3 × 50 mL). The organic phase was dried over Na_2SO_4, and removal of the solvent under reduced pressure left 0.88 g of a dark brown oil that upon column chromatography with 1:1 AcOEt/hexane as eluant afforded **20h** (0.35 g, 42%) as a white solid: mp 112−114 °C (*i*-PrOH). IR: 2810, 1682, 1669. ^{1}H NMR ($CDCl_3$): δ 1.85−1.92 (m, 2H, -CH_2-CH_2-CH_2-CO-); 2.34−2.52 (m, 11H, H_5, $2H_7$, -CH_2-N(CH_2-CH_2)$_2$N-CH_2-); 2.83−2.98 (m, 8H, $1H_6$, -N(CH_2-CH_2)$_2$N-, -CH_2-CO-Ph, H_5); 6.96−7.02 (m, 1H, H_2); 7.29−7.34 (m, 1H, $1H_3$); 7.56−7.63 (m, 2H, H_3 Ph); 7.84−7.89 (m, 2H, H_2 del Ph). ^{13}C NMR ($CDCl_3$): δ 22.6 (-CH_2-CH_2-CH_2-CO-); 28.4 (C_7); 36.2 (C_6); 38.6 (-CH_2-CO-Ph); 46.2 (C_5); 46.5 (2C, -N(CH_2-CH_2)$_2$N-); 52.4 (2C,-N(CH_2-CH_2)$_2$N-); 57.6 (RRN-CH_2-); 63.2 (-CH_2-NRR); 106.7 (C_3); 115.8 (d, 2C, $J_{C-F} = 21.8$, *m*-F-Ph); 121.4 (C_{3a}); 129.9 (*p*-F-Ph); 130.8 (d, 2C, $J_{C-F} = 9.3$, *o*-F-Ph); 143.2 (C_2); 165.9 (d, $J_{C-F} = 251$, C-F); 167.1 (C_{7a}); 194.2 (-CO-); 198.9 (-CO-Ph). Anal. ($C_{23}H_{27}FN_2O_3$) C, H, N. Hydrochloride: mp 202−204 °C (AcOEt).

6-{4-[5-(4-Fluorophenyl)-5-oxopentyl]piperazin-1-ylmethyl}-4,5,6,7-tetrahydrobenzo[*b*]furan-4-one (20i). A solution of 5-bromo-1-(*p*-fluorophenyl)-1-pentanone[94] (0.35 g, 1.37 mmol) in methyl isobutyl ketone (15 mL) was added under argon, with stirring, to a mixture of piperazine **26** (0.32 g, 1.37 mmol), anhydrous Na_2CO_3 (0.43 g) and KI (0.1 g) in 15 mL of the same solvent, and the mixture was refluxed for 24 h. The precipitate was filtered out, and removal of the solvent from the filtrate under reduced pressure left 0.56 g of a dark brown oil that upon column chromatography with 19:1 CH_2Cl_2/CH_3OH as eluent afforded amino ketone **20i** (0.28 g, 50%) as a white solid: mp 92−94 °C (*i*-PrOH). IR: 2810, 1682, 1669. ^{1}H NMR ($CDCl_3$): δ 1.56−1.63 (m, 2H, -CH_2-CH_2-CH_2-CO-); 1.70−1.80 (m, 2H, -CH_2-CH_2-CH_2-CO-); 2.20−2.67 (m, 16H, H_5, $1H_6$, $2H_7$, -CH_2-N(CH_2-CH_2)$_2$N-CH_2-); 2.96 (t, $J = 7.2$, 2H, -CH_2-CO-Ph); 3.01−3.08 (m, 1H, $1H_5$); 6.65 (d, $J = 2.0$, 1H, H_2); 7.12 (t, $J = 8.6$, 2H, H_3 Ph); 7.32 (d, 1H, H_3); 7.98 (dd, $J = 8.9, 5.4$, 2H, H_2 Ph). ^{13}C NMR ($CDCl_3$): δ 22.7 (-CH_2-CH_2-CH_2-CO-); 26.8 (-CH_2-CH_2-CH_2-CO-); 28.5 (C_7); 33.7 (C_6); 38.6 (-CH_2-CO-Ph); 43.0 (C_5); 53.6 (2C, -N(CH_2-CH_2)$_2$N-); 53.8 (2C,-N(CH_2-CH_2)$_2$N-); 58.6 (RRN-CH_2-); 63.2 (-CH_2-NRR); 106.8 (C_3); 116.0 (d, 2C, $J_{C-F} = 21.8$, *o*-F-Ph); 121.5 (C_{3a}); 131.0 (d, 2C, $J_{C-F} = 9.4$, *m*-F-Ph); 133.8 (d, 2C, $J_{C-F} = 2.9$, *p*-F-Ph); 143.2 (C_2); 165.9 (d, $J_{C-F} = 251$, C-F); 167.1 (C_{7a}); 194.2 (-CO-); 198.9 (-CO-Ph). Anal. ($C_{24}H_{29}FN_2O_3$) C, H, N. Hydrochloride: mp 235−240 °C (AcOEt). Anal. ($C_{24}H_{29}FN_2O_3$·HCl·H_2O) C, H, N.

Pharmacological Test Methods. D_1 and D_2 Binding Assays. Male Sprague−Dawley rats were killed by decapitation and their brains were rapidly removed and dissected on an ice-cold plate. Striatal membrane preparations were obtained by homogenization (Polytron homogenizer, setting 6, 10 s) in 50 mM Tris-HCl (pH 7.7 at 25 °C; about 100 μL/mg of tissue) containing 5 mM EDTA; the homogenates were centrifuged (49000*g* for 15 min at 4 °C; Sorvall RC-26 plus), resuspended in 50 mM Tris-HCl buffer (pH 7.4 at 25 °C) and centrifuged again (same conditions), and the final pellets were stored at −80 °C pending use. All drugs were stored in 1 mM solutions at −20 °C and diluted to the required concentrations on ice immediately before binding assays. Just before binding assays, the striatal membrane pellets were resuspended (1.25 mg original wet weight/750 μL for D_2 assays, 1.00 mg/750 μL for D_1) in 50 mM Tris-HCl buffer (pH 7.4 at 25 °C) containing 120 mM NaCl, 5 mM KCl, 2 mM $CaCl_2$ and 1 mM $MgCl_2$. For D_2 binding assays, 750-μL aliquots of striatal membrane preparation were added to ice-cold tubes containing (a) 100 μL of [^{3}H]spiperone, (b) 50 μL of ketanserin (final concentration 50 nM) to block 5-HT_{2A} receptors, and (c) either 100 μL of buffer (for total binding assay) or 100 μL of sulpiride (final concentration 10 μM) to allow quantification of unspecific binding of [^{3}H]spiperone or 100 μL of the compounds to be tested. For D_1 binding assays, the same procedure was followed except that [^{3}H]spiperone was replaced by [^{3}H]SCH23390, ketanserin by buffer, and sulpiride by nonradiolabeled SCH23390 (final concentration 1 μM) to allow quantification of nonspecific binding by [^{3}H]SCH23390. The final assay volume was thus 1 mL in all cases. All assays were performed in duplicate. Incubations (15 min at 37 °C) were stopped by rapid vacuum filtration through GF-52 glass fiber filters (Schleicher and Schuell) in a Brandel M-30 cell harvester. The filters were rinsed three times with 3 mL of ice-cold 50 mM Tris-HCl buffer (pH 7.4), and radioactivity was determined by liquid scintillation counting in a Beckman LS-6000LL apparatus.

D_4 Binding Assays. Radioreceptor binding studies with [^{3}H]YM-09-151-2 (emonapride, a D_2-like receptor antagonist, 81−87 Ci/mmol) were performed in membrane preparations from bovine retina. Retinae were homogenized in a saline solution (1:20 w/v) containing in mM: Tris, 50; EDTA, 1; $CaCl_2$, 1.5; $MgCl_2$, 4; KCl, 5; NaCl, 120; pH 7.4. The homogenate was centrifuged at 48000*g* for 20 min at 4 °C, resuspended 1:20,

recentrifuged and resuspended at a final dilution of about 4 mg original wet tissue/mL of saline (1:250 w/v) for use in the binding assay. For competition experiments, membranes were incubated at 30 °C in the presence of 0.1 nM [^{3}H]YM-09-151-2 for 60 min. For [^{3}H]YM-09-151-2 binding, duplicate test tubes contained in a final volume of 1.5 mL: 0.5 mL membranes, 0.15 tracer, and drug. The affinity of [^{3}H]YM-09-151-2 was measured by saturation experiments in similar conditions and was 0.4 nM. Nonspecific binding was assessed by 2 mM DA. At the end of the incubation period, the radioactivity bound to the receptor was separated from the free ligand by rapid vacuum filtration through glass fiber filters (GF/C) in a Brandel 30-well filtration apparatus. Filters were counted by liquid scintillation spectroscopy in a Packard LS-1600 apparatus and converted from cpm to dpm.

5-HT$_{2A}$ Binding Assays. Male 200−250-g Sprague−Dawley rats were asphyxiated with CO_2 and decapitated. The frontal cortex, containing 5-HT$_{2A}$ receptors,[95,96] was dissected free on ice, frozen on dry ice and stored at −70 °C until use (generally less than 1 week later). All membrane preparation procedures were carried out at 4 °C. The tissue was thawed on ice and homogenized with 10 volumes of 0.32 M sucrose in a Polytron homogenizer (Brinkmann Instruments, Westbury, NY). The homogenate was centrifuged twice at 4 °C (900*g* for 10 min followed by 40000*g* for 30 min), the supernatant was discarded, and the pellet was resuspended in Tris HCl-buffer (pH 8.07) in a Teflon/glass homogenizer (10 strokes by hand). This suspension was incubated at 37 °C for 15 min to remove endogenous 5-HT and centrifuged for 30 min at 40000*g*, and the final pellet was resuspended in Tris-HCl buffer of pH 8.07 containing 4 mM $CaCl_2$ and 0.1% ascorbic acid. Competition at [^{3}H]ketanserin binding sites was assayed in triplicate in assay mixtures consisting of 750 μL of membrane homogenate, 50 μL of [^{3}H]ketanserin, 50 μL of either buffer or the compound under test, 50 μL of masking ligand solution (1 μM methysergide) as required, and buffer to a final volume of 1 mL. Mixtures were incubated for 30 min at 37 °C. The assay was terminated by rapid filtration through Whatman GF/C filter strips (presoaked in 3% polyethylenimine) in a Brandel cell harvester (Gaithersburg, MD) followed by washing with ice-cold Tris-HCl buffer (pH 6.6) to remove unbound radioligand. The radioactivity retained on filters was determined by liquid scintillation counting in a beta counter (Beckman, LS-6000LL).

5-HT$_{2C}$ Binding Assays. Bovine choroid plexus containing 5-HT$_{2C}$ receptors[97] was treated as described in the 5-HT$_{2A}$ binding assay. A suspension of the resulting pellet in the same buffer was stored on ice while not being manipulated. Competition at [^{3}H]mesulergine binding sites was determined by a protocol analogous to that described above for the 5-HT$_{2A}$ binding assay, using a final [^{3}H]mesulergine concentration of 2 nM and 1 mM mianserine as 5-HT$_{2A}$ masking ligand. The mixtures were incubated for 1 h at room temperature. Membranes were harvested on Whatman GF/B filters.

Functional Experiments. Male Sprague−Dawley rats (250−300 g) were asphyxiated with CO_2 and decapitated. The stomach was dissected free from the abdomen and immersed in modified Krebs solution of the following composition (mM): NaCl, 119; KCl, 4.7; $MgSO_4 \cdot 7H_2O$, 1.2; $CaCl_2 \cdot 2H_2O$, 2.5; KH_2PO_4, 1.18; $NaHCO_3$, 25; glucose, 11. Strips of stomach fundus were prepared by Vane's method[98] and mounted in organ baths containing 10 mL of the same Krebs solution as above, maintained at 37 °C and aerated with carbogen (95% O_2, 5% CO_2). Before addition of drugs, the tissue strips were equilibrated for 1 h under a 1-g load. Isometric contractions were recorded during cumulative addition of serotonin using a Grass FTO3C transducer and a Grass 7D polygraph.

Concentration−response curves for serotonin were constructed as per Van Rossum.[99] In the initial control runs, stable contractions were achieved over the concentration range 0.01 nM−10 μM. Following the initial control run, each tissue strip was run alternately with and without antagonist. Between runs, the tissues were washed and allowed to rest for 60 min. Antagonist potency was measured as per Mackay[100] in terms of pA_2 (−log concentration of antagonist required to maintain a constant response when the agonist concentration is doubled).

Behavioral Assays. Mice weighing 25 ± 5 g and rats weighing 250 ± 50 g were housed in groups of 12 (mice) or 4 (rats) under regulated conditions (light/dark cycle between 8.00 and 20.00 h at 21 ± 2 °C) in standard Makrolon cages (215 × 465 × 145 mm). The animals received standard laboratory chow and tap water ad libitum until the beginning of the experiments.

Locomotor Activity. The locomotor activity of animals in square arenas (50 × 50 × 30 cm) under a video camera suspended from the ceiling was recorded over a 1-h period between 10.00 and 14.00 h and analyzed, using a computerized animal observation system (EthoVision V. 1.90, Noldus Information Technology, Wageningen, The Netherlands) located in a separated room. The variables considered were distance travelled (in cm), rearing (reduction of vertical projection area by 15% or more), and velocity.

Amphetamine-Induced Hypermotility. Animals were treated with 5 mg/kg dextroamphetamine sulfate 30 min after pretreatment with vehicle, haloperidol, clozapine, risperidone, **20b** or **20c**, and each animal was then immediately placed in its experimental arena.

Apomorphine-Induced Climbing. The method of Protais et al.[101] was used, with a minor modification. Vehicle, haloperidol, clozapine, risperidone, **20c** (0.25−0.5 mg/kg) or **20b** (0.6−4 mg/kg) was administered to mice (6 for drug and dosage, 12 for vehicle) that 30 min later were treated subcutaneously with 2 mg/kg apomorphine. The mice were then placed in individual cylindrical wire cages (diameter, 12 cm; height, 14 cm), and over the next 30 min their climbing activity was recorded every 10 min using the following scale: 0, four paws on the floor; 1, one or two paws against the cage wall; 2, three or four paws used to cling to the grid wall. The scores recorded 20 and 30 min post-apomorphine were added for use in further calculations.

Catalepsy. Thirty minutes after administration of vehicle, haloperidol, clozapine, risperidone, **20c** (0.5−8 mg/kg) or **20b** (0.5−6 mg/kg), mice (n = 6−12) were placed with their forepaws on one horizontal wire and their hindpaws on another 6 cm away and 2 cm lower. The time during which the mouse maintained this position was recorded; maintenance for more than 30 s was considered to indicate catalepsy.

Neurochemical Assays. Sprague−Dawley rats were killed by decapitation 120 min after injection of vehicle, reference drugs haloperidol (0.5 mg/kg), clozapine (20 mg/kg), risperidone (10 mg/kg) or **20c** (10 mg/kg). Within 1 min of decapitation the brain was removed and the striatum dissected free on an ice-cold plate as described by Glowinski and Iverssen.[102] Samples of striatal tissue were weighed in 1.5-mL conical test tubes and 20 μL/mg of PCA solution (0.1 M perchloric acid and 4 × 10^{-5} M sodium bisulfite) was then added. The mixture was homogenized (Ultra-turrax T8, position 5, 10 s), and the homogenate was centrifuged twice for 15 min at 13 000 rpm (Heraeus Biofugue-13). The supernatant was filtered (Millipore HV 0.45 μm) and stored at −80 °C prior to HPLC, which was performed using 20-μL samples, a Tracer RP-18 column (15 × 0.4 cm, 5 μm Spherisorb), a Waters pump 616 pump with a 600S controller, a Waters 717 plus autosampler refrigerated at 4 °C and a Coulochem 5100 A detector with a 5021 conditioning cell and a 5011 analytical cell. The mobile phase (an 88:12 mixture of citrate buffer (pH 4.0) and methanol containing 0.83 mM sodium octanesulfonate) was filtered (0.22-μm pore size) and degassed with ultrasound before use. The flow rate was 1 mL/min and ambient temperature was maintained. HPLC operations were controlled and the detection data processed using Waters Millenium software (V 2.10).

Drugs and Chemicals. [^{3}H]Spiperone (104 Ci/mmol), [^{3}H]-SCH23390 (81 Ci/mmol) and [^{3}H]mesulergine (76 Ci/mmol) were obtained from Amersham International (England), [^{3}H]-ketanserin (60.08 Ci/mmol) from DuPont NEN (Boston, MA), [^{3}H]YM-09-151-2 from New England Nuclear (Boston, MA), unlabeled (*R*)-(+)-SCH23390·HCl, mianserine and methysergide from Research Biochemicals Inc., and unlabeled spiper-

one, sulpiride·HCl and 5-hydroxytryptamine·HCl from Sigma (St. Louis, MO). In behavioral and neurochemical experiments, all compounds were administered in 0.01 mL/g injections (apomorphine subcutaneously and all others intraperitoneally). Haloperidol (Sigma) was dissolved in 1% lactic acid in water. Apomorphine hydrochloride (R.B.I.), dextroamphetamine sulfate (Sigma), risperidone (Janssen), clozapine (Sandoz) and the new drugs were prepared in saline (apomorphine with 1% ascorbic acid (w/v) to prevent oxidation). Methanol, citric acid monohydrate, perchloric acid and sodium chloride were all of reagent grade. DA·HCl, 3,4-dihydroxyphenylacetic acid (DOPAC), 3-methoxytyramine·HCl (3-MT), homovanillic acid (HVA) and 1-octanesulfonic acid (sodium salt) were purchased from Sigma. All other reagents were of analytical grade and supplied by Sigma or Merck (Darmstadt, Germany).

Calculations and Statistical Analysis. The nonlinear curve-fitting programs Kaleidagraph (Synergy Software, Reading, PA) and Graphpad Prism version 2.1 (GraphPad, San Diego, CA) were used to fit the equation: $E = E_{max} - [E_{max} - E_{min}]/[1 + (IC_{50}/C)^n]$, where E_{max} and E_{min} are dpm at the beginning and end of the competition experiment, respectively, C is the concentration of the inhibitor, IC_{50} is the inhibitor concentration required to inhibit binding by 50%, and n is the slope of the decay. Nonspecific binding was determined independently in the presence of an unlabeled masking ligand. In D_4 binding assays, specific binding was obtained by subtracting nonspecific binding from totals and normalized to specific binding in the absence of drugs. In preliminary experiments, normalized data from 10−12 concentrations were fitted by either a single-site model (eq 1) or a two-site model (eq 2) by nonparametric fitting using a modified Lowenberg−Marquardt algorithm implemented in the data analysis program Microcal Origin version 3.5 (Microcal Software, Inc., Northampton, MA). pK_i values were calculated from IC_{50} values using the equation $K_i = IC_{50}/(1 + D/K_d)$, where K_d is the equilibrium dissociation constant of the radioligand (determined by saturation binding studies) and D is the concentration of radioligand used. Other pharmacological calculations were performed using the Pharmacologic Calculation System[103] or Prism version 2.1 (GraphPad, San Diego, CA). The statistical significance of differences between means was determined by Student's test for unpaired data, and differences with probabilities lower than 0.05 were considered statistically significant. Percentage change in locomotor activity (%LC) was calculated as %LC = 100 × [(mean cm/h control animals) − (mean cm/h treated animals)]/(mean cm/h control animals). ED_{50} values for inhibition of apomorphine-induced climbing were calculated by nonlinear regression, and ED_{50} values for induction of catalepsy were calculated by Litchfield and Wilcoxon's method.

Acknowledgment. This research was supported by the Spanish Comisión Interministerial de Ciencia y Tecnología (CICYT) under Grant SAF 95-1081 and by the Xunta de Galicia Grants XUGA20308B94, XUGA20308B96, and XUGA20319B97. M.E.R. was aided by a predoctoral grant from Xunta de Galicia and G.Y.M. with a predoctoral grant from the Agencia Española de Cooperación Iberoamericana (AECI). We also thank Sandoz and Janssen Laboratories for the generous gifts of clozapine and risperidone, respectively.

References

(1) Reynolds, G. P. Developments in the drug treatment of schizophrenia. *Trends Pharmacol. Sci.* **1992**, *13*, 116−121.
(2) Martin, A. R. Antipsychotic Agents. In *Burger's Medicinal Chemistry*, 5th ed.; Wolf, M. E., Ed.; Wiley-Interscience: New York, 1997; Vol. 5, p 195.
(3) Sanberg, P. R. Haloperidol-induced catalepsy is mediated by postsynaptic dopamine receptors. *Nature (London)* **1980**, *284*, 472−473.
(4) Seeman, P.; Bzowej, N. H.; Guan, H. C.; Bergeron, C.; Becker, L. E.; Reynols G. P.; Bird, E. D.; Riedeger, P.; Jellinger, K.; Watanabe, S.; Tourtellote, W. W. Human Brain Dopamine Receptors in Children and Aging Adults. *Synapse* **1987**, *1*, 399−404.
(5) Nowak, K.; Welsch-Kunze, S.; Kuschinsky, K. Conditioned tolerance to haloperidol- and droperidol-induced catalepsy. *Naunyn-Schmiedeberg's Arch. Pharmacol.* **1988**, *337*, 385−391.
(6) Seeman, P.; Chou-Wong, M.; Tadesco, J.; Wong, K. Antipsychotic drug doses and neuroleptic/dopamine receptors. *Nature* **1976**, *261*, 717−719.
(7) Seeman, P.; Lee, T. Antipsychotic drugs: direct correlation between clinical potency and presynaptic action on dopamine neurons. *Science* **1975**, *188*, 1217−1219.
(8) Peroutka, S. J.; Synder, S. H. Relationship of neuroleptic drug effects at brain dopamine, serotonin, alpha-adrenergic, and histamine receptors to clinical potency. *Am. J. Psychiatry* **1980**, *137*, 1518−1522.
(9) Filton, A.; Heel, R. C. Clozapine. A review of its pharmacological properties and therapeutic use in schizophrenia. *Drugs* **1990**, *40*, 722−747.
(10) Meltzer, H. Y. An overview of the mechanism of action of clozapine. *J. Clin. Psychiatry* **1994**, *55*, 45−52.
(11) Schwarz, J. T.; Brotman, A. W. A clinical guide to antipsychotic drugs. *Drugs* **1992**, *44*, 981−992.
(12) Rosenheck, R.; Cramer, J.; Xu, W.; Henderson, W.; Frismann, L. A comparison of clozapine and haloperidol in hospitalized patients. *N. Engl. J. Med.* **1997**, *337*, 809−815.
(13) Sanders-Bush, E.; Mayer, S. E. 5-Hydroxytryptamine (Serotonin) Receptor Agonists and Antagonists. In *The pharmacological basis of therapeutics*, 9th ed.; Hardman, J. G., Limbird, L. E., Molinoff, P. B., Ruddon, R. W., Goodman, A., Eds.; McGraw-Hill: New York, 1996; pp 249−263.
(14) Meltzer, H. Y.; Matsubara, S.; Lee, J. C. Classification of typical and atypical antipsychotic drugs on the basis of dopamine D-1, D-2 and serotonin-2 p*Ki* values. *J. Pharmacol. Exp. Ther.* **1989**, *251*, 238−246.
(15) Roth, B. L.; Meltzer, H. Y. The role of Serotonin in Schizophenia. In *Psychopharmacology: The Fourth Generation of Progress*; Bloom, F. E., Kupfer, D. J., Eds.; Raven Press: Ltd.: New York, 1995; pp 1215−1227.
(16) Leysen, J. E.; Janssen, P. M.; Megens, A. A.; Schotte, A. Risperidone: a novel antipsychotic with balanced serotonin-dopamine antagonism, receptor occupancy profile, and pharmacologic activity. *J. Clin. Psychiatry* **1994**, *55*, 5−12.
(17) Meltzer, H. Y.; Matsubara, S.; Lee, J. C. The ratios of serotonin-2 and dopamine-2 affinities differentiate atypical and typical antipsychotic drugs. *Psychopharmacol. Bull.* **1989**, *25*, 390−392.
(18) Roth, B. L.; Tandra, S.; Burgess, L. H.; Sibley, D. R.; Meltzer, H. Y. D_4 receptor binding affinity does not distinguish between typical and atypical antipsychotic drugs. *Psychopharmacology* **1995**, *120*, 365−368.
(19) Lowe, J. A., III Atypical Antipsychotics based on the D_2/5-HT_2 Ratio Hypothesis. *Curr. Med. Chem.* **1994**, *1*, 50.
(20) Martin, P.; Waters, N.; Carlsson, A.; Carlsson, M. L. The apparent antipsychotic action of the 5-HT2A receptor antagonist MDL 100907 in a mouse model of schizophrenia is counteracted by ritanserin. *J. Neural. Transm.* **1997**, *104*, 561−564.
(21) Padich, R. A.; McCloskey T. C.; Kehne, J. H. 5-HT modulation of auditory and visual sensorimotor gating: II. Effects of the 5-HT2A antagonist MDL 100,907 on disruption of sound and light prepulse inhibition produced by 5-HT agonists in Wistar rats. *Psychopharmacology* **1996**, *124*, 107−116.
(22) Schmidt, C. J. Development of a selective 5-HT_{2A} receptor antagonist for the treatment of schizophrenia. IBS's International Conference on Serotonin Receptors. Central Nervous System. Targets for New Therapeutic Agents, Philadelphia, PA, 1996.
(23) Meltzer, H. Y. Atypical antipsychotic drugs: which receptors are relevant? IBS's International Conference on Serotonin Receptors. Central Nervous System. Targets for New Therapeutic Agents, Philadelphia, PA, 1996.
(24) Baxter, G.; Kennett, G. A.; Blaney, F.; Blackburn, T. 5-HT_2 Receptor Subtypes: A Family Re-united? *Trends Pharmacol. Sci.* **1995**, *16*, 105−110.
(25) (a) Humphrey, P. P. A.; Hartig, P.; Hoyer, D. A. Proposed New Nomenclature of 5-HT Receptors. *Trends Pharmacol. Sci.* **1993**, *14*, 233−236. (b) Nelson, D. L. The 5-HT_2 Subfamily of Receptors: Pharmacological Challenges. *Med. Chem. Res.* **1993**, *3*, 306−316.
(26) Hoyer, D.; Clarke, D. E.; Fozard, J. R.; Hartig, P. R.; Martin, G. R.; Mylecharane, E. J.; Saxena, P. R.; Humphrey, P. P. A. VII International Union of Pharmacology Classification of Receptors for 5-Hydroxytryptamine (Serotonin). *Pharmacol. Rev.* **1994**, *46*, 157−203.

(27) Duxon, M. S.; Flanigan, T. P.; Reavley, A. C.; Baxter, G. S.; Blackburn, T. P.; Fona, K. C. F. Evidence for the expresion of the 5-hydroxytriptamine-2B receptor protein in the rat central nervous system. *Neurosciences* **1997**, *76*, 323−329.

(28) (a) Pazos, A.; Probst, A.; Palacios, J. M. Serotonin receptors in the human brain-III. Autoradiographic mapping of serotonin-1 receptors. *Neuroscience* **1987**, *21*, 97−122. (b) Molineaux, S. M.; Jessell, T. M.; Axel, R.; Julius, D. 5-HT1c receptor is a prominent serotonin receptor subtype in the central nervous system. *Proc. Natl. Acad. Sci. U.S.A.* **1989**, *86*, 6793−6797.

(29) Westphal, R. S.; Sanders-Bush, E. Reciprocal binding properties of 5-hydroxytryptamine type 2C receptor agonists and inverse agonists *Mol. Pharmacol.* **1994**, *46*, 937−942.

(30) Barker, E. L.; Westphal, R. S.; Schmidt, D.; Sanders-Bush, E. Constitutively active 5-hydroxytryptamine$_{2C}$ receptors reveal novel inverse agonist activity of receptors ligands. *J. Biol. Chem.* **1994**, *269*, 11687−11690.

(31) Reavill, C.; Kettle, A.; Holland, V.; Riley, G.; Blackburn, T. P. Attenuation of haloperidol-induced catalepsy by a 5-HT$_{2C}$ receptor antagonist. *Br. J. Pharmacol.* **1999**, *126*, 572−574.

(32) Seeman, P.; Guan, H. C.; Van Tol, H. H. Dopamine D4 receptors elevated in schizophrenia. *Nature* **1993,** *365*, 441−445.

(33) Steiner, G.; Bach, A.; Bialojan, S.; Greger, G.; Hege, H.-G.; Höger, T.; Jochims, K.; Munschauer, R.; Neumann, B.; Teschendorf, H.-J.; Traut, Unger, L.; Gross, G. D$_4$/5-HT$_{2A}$ Receptor antagonists: LU-111995 and other potential new antipsychotics in development. *Drugs Future* **1998**, *23*, 191−204.

(34) Kulagowski, J.; Broughton, H. B.; Curtis, N. R.; Mawer, I. M.; Ridgill, M. P.; Baker, R.; Emms, F.; Marwood, R.; Patel, S.; Patel, S.; Ragan, C. I.; Leeson, P. A. 3-[[4-(4-Chlorophenyl)piperazin-1-yl]methyl]-1H-pyrrolo[2,3-b]pyridine: An Antagonist with High Affinity and Selectivity for the Human Dopamine D4 Receptor. *J. Med. Chem.* **1996**, *39*, 1941−1942.

(35) Sanner, M. A.; Chappie, T. A.; Dunaiskis, A. R.; Fliri, A. F.; Desai, K. A.; Zorn, S. H.; Jackson, E. R.; Johnson, C. G.; Morrone, J. M.; Seymour, P. A.; Majchrzak, M. J.; Faraci, W. S.; Collins, J. L.; Duignan, D. B.; Di Prete, C. C.; Lee, J. S.; Trozzi, A. Synthesis, SAR and pharmacology of CP-239, 019: a potent, selective dopamine D4 receptor antagonist. *Bioorg. Med. Chem. Lett.* **1998**, *8*, 725−730.

(36) Bristow, L. J.; Kramer, M. S.; Kulagowski, J.; Patel, S.; Ragan, C. I.; Seabrook, G. R. Schizophrenia and L-745,870, a novel dopamine D4 receptor antagonist. *Trends Pharmacol. Sci.* **1997**, *18*, 186−187.

(37) Lieberman, J. A.; Hohn, C. A.; Mikane, J.; Rai, K.; Pisciotta, A. V.; Salz, B. L.; Howard, A. Clozapine-induced agranulocytosis: non-crossreactivity with other antipsychotic drugs. *J. Clin. Psychiatry* **1988**, *49*, 271−277.

(38) Kilian, J. G.; Kerr, K.; Lawrence, C. Myocarditis and cardiomyopathy associated with clozapine. *Lancet* **1999**, *354*, 1851−1855.

(39) Cortizo, L.; Santana, L.; Raviña, E.; Orallo, F.; Fontenla, J. A.; Castro, E.; Calleja, J. M.; de Ceballos, M. L. Synthesis and antidopaminergic activity of some 3-(aminomethyl)tetralones as analogues of butyrophenone. *J. Med. Chem.* **1991**, *34*, 2242−2247.

(40) Fontenla, J. A.; Osuna, J.; Rosa, E.; Castro, E.; Ferreiro, T. G.; Loza García, I.; Calleja, J. M.; Sanz, F.; Rodriguez, J.; Raviña, E.; Fueyo, J.; Masaguer, C. F.; Vidal, A.; de Ceballos, M. L. Synthesis and atypical antipsychotic profile of some 2-(2-piperidinoethyl) benzocycloalkanones as analogues of butyrophenone. *J. Med. Chem.* **1994**, *37*, 2564−2573.

(41) Raviña, E.; Fueyo, J.; Masaguer, C. F.; Negreira, J.; Cid, J.; Loza, I.; Honrubia, A.; Tristan, H.; Ferreiro, T. G.; Fontenla, J. A.; Rosa, E., Calleja, J. M.; de Ceballos, M. L. Synthesis and affinities for dopamine (D$_2$) and 5-hydroxytryptamine (5-HT$_{2A}$) receptors of 1-(benzoylpropyl)-4-(1-oxocycloalkyl-2-ethyl)piperazines as cyclic butyrophenone derivatives. *Chem. Pharm. Bull.* **1996**, *44*, 534−541.

(42) Loza, M. I.; Verde, I.; Castro, E.; Orallo, F.; Fontenla, J. A.; Calleja, J. M.; Raviña, E.; Cortizo, L. 5-HT$_2$ antagonist activity of 3-aminomethyl tetralones. *Bioorg. Med. Chem. Lett.* **1991**, *1*, 717−720.

(43) Raviña, E.; Masaguer, C. F.; Cid, J.; Fontenla, J. A.; Ferreiro, T. G.; Cadavid, M. I.; Loza, I.; de Ceballos, M. L. Butyrophenone analogues: Synthesis of 2-methyl-3-ethyl-5-aminomethyl-4,5,6,7-tetrahydroindol-4-ones and their affinities for D$_1$, D$_2$ and 5-HT$_{2A}$ receptors. *Bioorg. Med. Chem. Lett.* **1995**, *5*, 579−584.

(44) Masaguer, C. F.; Raviña, E. A practical and efficient route for synthesis of 6-aminomethyl-4-oxo-4,5,6,7-tetrahydroindoles as new CNS precursors. *Tetrahedron Lett.* **1996**, *37*, 5171−5174.

(45) Masaguer, C. F.; Casariego, I.; Raviña, E. Conformationally restricted butyrophenones with mixed dopaminergic (D$_2$) and serotoninergic (5-HT$_{2A}$) affinities. Synthesis of 5-aminoethyl- and 6-aminomethyl-4-oxotetrahydroindoles as potential atypical antipsychotics. *Chem. Pharm. Bull.* **1999**, *47*, 621−632.

(46) Masaguer, C. F.; Formoso, E.; Raviña, E.; Tristán, H.; Loza, I.; Rivas, E.; Fontenla, J. A. Butyrophenone analogues in the carbazole series: synthesis and determination of affinities at D$_2$ and 5-HT$_{2A}$ receptors. *Bioorg. Med. Chem. Lett.* **1998**, *8*, 3571−3576.

(47) Masaguer, C. F.; Raviña, E.; Fontenla, J. A.; Brea, J.; Tristán, H.; Loza, M. I.; Butyrophenone analogues in the carbazole series as potential atypical antipsychotics: synthesis and determination of affinities at D$_2$, 5-HT$_{2A}$, 5-HT$_{2B}$ and 5-HT$_{2C}$ receptors. *Eur. J. Med. Chem.* **2000**, *35*, 83−95.

(48) Raviña, E.; Negreira, J.; Cid, J.; Masaguer, C. F.; Rosa, E.; Rivas, M. E.; Fontenla, J. A.; Loza, M. I.; Tristán, H.; Cadavid, M. I.; Sanz, F.; Lozoya, E.; Carotti, A.; Carrieri, A. Conformationally Constrained Butyrophenones with Mixed Dopaminergic (D$_2$) and Serotoninergic (5-HT$_{2A}$, 5-HT$_{2C}$) Affinities: Synthesis, Pharmacology, 3D-QSAR and Molecular Modeling of (Aminoalkyl)benzo and -thienocycloalkanones as Putative Atypical Antipsychotics. *J. Med. Chem.* **1999**, *42*, 2774−2797.

(49) Duncan, R. L., Jr.; Helsey, G. C.; Welstead, W. J., Jr.; Da Vanzo, J. P.; Funderburk, W. H.; Lunsford, C. D. Aroylpiperidines and pyrrolidines. A new class of potent central nervous system depressants. *J. Med. Chem.* **1970**, *13*, 1−6.

(50) Ismaiel, A. M.; Arruda, K.; Teitler, M.; Glennon, R. A. Ketanserin analogues: the effect of structural modifications on 5-HT$_2$ serotonin receptor binding. *J. Med. Chem.* **1995**, *38*, 1196−1202.

(51) For benzisoxazolyl−benzoyl bioisosterism, see: (a) Shutske, G. M.; Setescak, L. L.; Allen, R. C.; Davis, L.; Effland, R. C.; Ranbom, K.; Kitzen, J. M.; Wilker, J. C.; Novick, W. J., Jr. [(3-Aryl-1,2-benzisoxazol-6-yl)oxy]acetic acids. A New Diuretic Series. *J. Med. Chem.* **1982**, *25*, 36−44. (b) Strupczewski, J. T.; Allen, R. C.; Gardner, B. A.; Schmid, B. L.; Stache, U.; Glamkowski, E. J.; Jones, M. C.; Ellis, D. B.; Huger, F. P.; Dunn, R. W. Synthesis and neuroleptic activity of 3-(1-substituted-4-piperidinyl)-1,2-benzisoxazoles. *J. Med. Chem.* **1985**, *28*, 761−769. (c) Mewshaw, R. E.; Silversman, L. S.; Mathew, R. M.; Kaiser, C.; Sherrill, R. G.; Cheng, M.; Tiffanny, C. W.; Karbon, E. W.; Bailey, M. A.; Borosky, S. A.; Ferkany, J. W.; Abreu, M. E. Bridged γ-carbolines and derivatives possessing selective and combined affinity for 5-HT$_{2A}$ and D$_2$ receptors. *J. Med. Chem.* **1993**, *36*, 1488−1495.

(52) Palermo, M. G.; Corbett, R.; Hartman, H. B.; Hrib, N. J.; Kafka, S.; Kongsamut, S.; Nmeoto, P. A.; Schilp, D. E.; Tseng, H.; Woods-Kettelberger, A.; Martin, L. L. Synthesis and evaluation of 3-(4-piperazinyl)-6-hydroxybenzisoxazoles and depot analogues as potential atypical neuroleptic compounds. Abstracts of Papers, 214 National Meeting of the American Chemical Society, Division of Medicinal Chemistry, Las Vegas, NV, 1997.

(53) Stetter, H.; Schreckenberg, M. Ein neue Methode zur Addition von an aktivierte Doppelbindungen, III. Aldehyden Addition von aromatischen und heterocyclischen Aldehyden an α,β-ungesättigte Nitrile. *Chem. Ber.* **1974**, *107*, 210−214.

(54) Stetter, H.; Kulmann, H. Addition von Aldehyden an aktivierte Doppelbindungen-XII. Ein neuer weg zu 4-oxooctandisäuren. *Tetrahedron* **1977**, *33*, 353−356.

(55) Stetter, H.; Kulmann, H. Addition von Aldehyden an aktivierte Doppelbindungen, X. Addition von aliphatischen, heterocyclischen und aromatischen Aldehyden an α,β-ungesättigte Ketone, Nitrileund Ester. *Chem. Ber.* **1976**, *109*, 2890−2896.

(56) Chapmann, O. L.; Fitton, P. A General Synthesis of the Troponoid System Based on Solvolysis of 1,4-Dihydrobenzyl Tosylates. *J. Am. Chem. Soc.* **1963**, *85*, 41−47.

(57) Casey, D. E. Behavioral effects of sertindole, risperidone, clozapine and haloperidol in Cebus monkeys. *Psychopharmacology* **1996**, *124*, 134−140.

(58) Hauber, W. Impairments of movement initiation and execution induced by a blockade of dopamine D1 or D2 receptors are reversed by a blockade of *N*-methyl-D-aspartate receptors. *Neuroscience* **1996**, *73*, 121−130.

(59) Sabol, K. E.; Seiden, L. S. Reserpine attenuates D-amphetamine and MDMA-induced transmitter release in vivo: a consideration of dose, core temperature and dopamine synthesis. *Brain Res.* **1998**, *806*, 69−78.

(60) Pugh, M. T.; O'Boyle, K. M.; Molloy, A. G.; Waddington, J. L. Effects of the putative D1 antagonist SCH23390 on stereotyped behaviour induced by the D2 agonist RU 24213. *Psychopharmacology* **1985**, *87*, 308−312.

(61) Busnell, P. J. Differential effects of amphetamine and related compounds on locomotor activity and metabolic rate in mice. *Pharmacol. Biochem. Behav.* **1986**, *25*, 161−170.

(62) Kolta, M. G.; Shreve, P.; Uretsky, N. J. Effect of pretreatment with amphetamine on the interaction between amphetamine and dopamine neurons in the nucleus accumbens. *Neuropharmacology* **1989**, *28*, 9−14.

(63) Moser, P. C.; Moran, P. M.; Frank, R. A.; Kehne, J. H. Reversal of amphetamine-induced behaviours by MDL 100,907, a selective 5-HT2A antagonist. *Behav. Brain Res.* **1996**, *73*, 163−167.

(64) Arnt, J. Differential effects of classical and newer antipsychotics on the hypermotility induced by two dose levels of D-amphetamine. *Eur. J. Pharmacol.* **1995**, *283*, 55−62.
(65) Pichler, L.; Pifl, C. Locomotor behaviour of selective agonist DA in mice: is endogenius dopamine the only catecholamine involved? *J. Pharm. Pharmacol.* **1989**, *41*, 690−693.
(66) Gustafsson, B.; Christensson, E. Amperozide, a new putatively antipsychotic drug with a limbic mode of action on dopamine mediated behaviour. *Pharmacol. Toxicol.* **1990,** *66*, 12−17.
(67) Schmidt, C. J.; Black, C. K.; Taylor, V. L.; Fadayel, G. M.; Humphreys, T. M.; Nieduzak, T. R.; Sorensen, S. M. The 5-HT2 receptor antagonist MDL 28,133A disrupts the serotonergic-dopaminergic interaction mediating the neurochemical effects of 3,4-methylenedioxymethamphetamine. *Eur. J. Pharmacol.* **1992**, *220*, 151−159.
(68) Schmidt, C. J.; Sorenssen, S. M.; Kehne, J. H.; Carr, A. A.; Palfreyman, M. G. The role of 5-HT2A receptors in antipsychotic activity. *Life Sci.* **1995**, *56*, 2220−2222.
(69) Di Chiara, G.; Porceddu, M. L.; Argiolas, A.; Gessa, G. L. Evidence for dopamine receptors mediating sedation in the mouse brain. *Nature* **1976**, *26*, 564−567.
(70) Ahlenius, S. Effects of the local application of 3-PPP and sulpiride enantiomer into the nucleus accumbens or into the ventral tegmental area on rat locomotor activity: evidence for the somatodendritic autorreceptors. *Naunyn-Schmiedeberg's Arch. Pharmacol.* **1992**, *314*, 516−522.
(71) Costall, B.; Naylor, R. J.; Nohria, V. Climbing behaviour induced by apomorphine in mice: A potential model for the detection of neuroleptic activity. *Eur. J. Pharmacol.* **1978**, *50*, 39−50.
(72) Boyce, S.; Kelly, E.; Davis, A.; Flemminger, S.; Jenner, P.; Marsden, C. D. SCH23390 may alter dopamine-mediated behaviour via striatal D1 receptors. *Biochem. Pharmacol.* **1985**, *34*, 1665−1669.
(73) Arnt, J.; Hyttel, J.; Perregaard, J. Dopamine D1 receptor agonists combined with the selective quinpirole facilitate the expression of oral stereotyped behaviour in rats. *Eur. J. Pharmacol.* **1987**, *133*, 137−145.
(74) Vaccheri, A.; Dall'Olio, R.; Gandolfi, O.; Roncada, P.; Montarano, N. Enhanced stereotyped response to apomorphine after chronic D1 blockade with SCH23390. *Psychopharmacology* **1987**, *91*, 394−396.
(75) Moore, N. A.; Axton, M. S. Production of climbing behaviour in mice requires both D1 and D2 receptor activation. *Psychopharmacology* **1988**, *94*, 263−266.
(76) Vasse, M.; Chagraoui, A.; Protais, P. Climbing and stereotyped behaviours in mice require the stimulation of the D1 dopamine receptors. *Eur. J. Pharmacol.* **1988**, *148*, 221−229.
(77) Davis, A.; Jenner, P.; Marsden, C. D. Differential ability of selective and nonselective dopamine agonists to induce climbing in the rat indicates the involvement of both D1 and D2 receptors in this behaviour. *Psychopharmacology* **1990**, *100*, 19−26.
(78) Schauss, J. M.; Titus, R. D.; Foreman, M. M.; Mason, N. R.; Truex, L. L. Aporphines as antagonists of dopamine D1 receptors. *J. Med. Chem.* **1990**, *33*, 600−607.
(79) Simon, P.; Charpentier, S.; Constentin, J. An automated method for the assessment of spontaneous and stereotyped climbing behavior in mice. Effects of the selective D1 and D2 dopamine receptor agonists SKF-38393 and RU-24926 and their association. *Methods Find. Exp. Clin. Pharmacol.* **1991**, *13*, 99−104.
(80) Skrinskaya, J. A.; Nikulina, E. M.; Popova, N. K. Role of genotype in brain dopamine metabolism and dopamine-dependent behaviour of mice. *Pharmacol. Biochem. Behav.* **1992**, *42*, 261−267.
(81) Megens, A. A.; Awouters, F. H.; Schotte, A.; Meert, T. F.; Dugovic, C.; Niemegeers, C. J.; Leysen, J. E. Survey on the pharmacodynamics of the new antipsychotic risperidone. *Psychopharmacology* (*Berlin*) **1994**, *114*, 9−23.
(82) Sorensen, S. M.; Kehne, J. H.; Fadayel, G. M.; Humphreys, T. M.; Ketteler, H. J.; Sullivan, C. K.; Taylor, V. L.; Schmidt, C. J. Characterization of the 5-HT2 receptor antagonist MDL 100907 as a putative atypical antipsychotic: behavioral, electrophysiological and neurochemical studies. *J. Pharmacol. Exp. Ther.* **1993**, *266*, 684−691.
(83) Costall, B.; Eniojukam, J. F.; Naylor, R. J. Spontaneous climbing behaviour of mice, its measurement and dopaminergic involvement. *Eur. J. Pharmacol.* **1982**, *85*, 125−132.
(84) Ferris, R. M.; Harfenist, M.; McKenzie, G. M.; Cooper, B.; Soroko, F. E.; Maxwell, R. A. BW234U (*cis*-9-[3-(3,5-dimethyl-1-piperazinyl)propyl]carbazole dihydrochloride): a novel antipsychotic agent. *J. Pharm. Pharmacol.* **1982**, *34*, 388−390.
(85) Morelli, M.; Di Chiara, G. Catalepsy induced by SCH23390 in rats. *Eur. J. Pharmacol.* **1985**, *117*, 179−185.
(86) Calderon, S. F.; Sanberg, P. R.; Norman, A. B. Quinolinic acid lesions of rat striatum abolish D1 and D2-dopamine receptor-mediated catalepsy. *Brain Res.* **1988**, *450*, 403−407.
(87) Novak, K.; Welsch-Kunzie, S.; Kuschinsky, K. Conditioned haloperidol and droperidol-induced catalepsy. *Naunyn-Schmiedeberg's Arch. Pharmacol.* **1988**, *337*, 385−391.
(88) Westerink, B. H. C. The effects of drugs on dopamine biosynthesis and metabolism in the rat brain. In *The Neurobiology of Dopamine*; Horn, A. S., Korf, J., Westerink, B. H. C., Eds.; Academic Press: London, 1979; pp 225−291.
(89) Anden, N. E. Regulation of monoamine synthesis and utilization by receptors. In *Handbook of Experimental Pharmacology*; Szekeres, L., Ed.; Springer: Berlin, 1980; Vol. 50, pp 429−462.
(90) Bunney, B. S.; Walters, J. R.; Roth, R. H.; Aghajanian, G. K. Dopaminergic neurons: effect of antipsychotic drugs and amphetamine on single cell activity. *J. Pharmacol. Exp. Ther.* **1973**, *185*, 560−571.
(91) Carlsson, A. Receptor-mediated control of dopamine metabolism. In *Pre and Postsynaptic Receptors*; Usdin, E., Bunney, W. E., Eds.; Marcel Dekker: New York, 1975; pp 49−65.
(92) Hyttel, J.; Arnt, J.; Berghe, M. V. D. Selective dopamine D_1 and D_2 receptor antagonists. In *Clinical Pharmacology in Psychiatry*; Dahl, S. G., Gram, L. F., Eds.; Springer-Verlag: Berlin, 1989; pp 109−122.
(93) Vogel, A. I. *Vogel's Textbook of Practical Organic Chemistry*, 5th ed.; Longman: Harlow, U.K., 1989.
(94) Mewshaw, R. E.; Silverman, L. S.; Mathew, R. M.; Kaiser, C.; Sherrill, R. G.; Cheng, M.; Tiffany, C. W.; Karbon, E. W.; Bailey, M. A.; Borosky, S. A.; Ferkany, J. W.; Abreu, M. E. Bridged γ-Carbolines and Derivatives Possessing Selective and Combined Affinity for 5-HT_2 and D_2 Receptors. *J. Med. Chem.* **1993**, *36*, 1488−1495.
(95) Bradley, P. B.; Engel, G.; Feniuk, W.; Fozard, J. R.; Humphrey, P. P. A.; Middlemiss, D. N.; Mylecharane, E. J.; Richardson, B. P.; Saxena, P. R. Proposals for the classification and nomenclature of functional receptors for 5-hydroxytryptamine. *Neuropharmacology* **1986**, *25*, 563−576.
(96) Pazos, A.; Cortes, R.; Palacios, J. M. Quantitative autoradiographic mapping of serotonin receptors in the rat brain II. Serotonin-2 receptors. *Brain Res.* **1985**, *346*, 231−245.
(97) Wong, D. T.; Threlkeld, P. G.; Robertson, D. W. Affinities of fluoxetine, its enantiomers, and other inhibitors of serotonin uptake for subtypes of serotonin receptors. *Neuropsychopharmacology* **1991**, *5*, 43−47.
(98) Vane, J. R.. A sensitive method for the assay of 5-hydroxytryptamine. *Br. J. Pharmacol.* **1957**, *12*, 344−349.
(99) Van Rossum, J. M. Cumulative dose−response curves II. Technique for the making of dose−response curves in isolated organs and the evaluation of drug parameters. *Arch. Int. Pharmacol.* **1963**, *143*, 299.
(100) Mackay, D. How should values of pA_2 and affinity constants for pharmacological competitive antagonists be estimated? *J. Phar. Pharmacol.* **1978**, *30*, 312−313.
(101) Protais, P.; Costentin, J.: Schwartz, J. C. Climbing behavior induced by apomorphine in mice: A simple test for the study of dopamine receptors in striatum. *Psychopharmacology* **1976**, *50*, 1−6.
(102) Glowinsky, J.; Iverssen, L. L. Regional studies of catecholamines in rat brain. The disposition of [3H]norepinephrine, [3H]dopamine and [3H]dopa in various regions of the brain. *J. Neurochem.* **1966**, *13*, 656−669.
(103) Tallarida, R. J.; Murray, R. B. *Manual of Pharmacologic Calculations with Computer Programs*, 2nd ed.; Springer-Verlag: New York, 1987; pp 153−158.

JM0009890