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Bioorganic & Medicinal Chemistry 13 (2005) 2547–2559

Bioorganic & Medicinal Chemistry

QSAR study for a novel series of *ortho* monosubstituted phenoxy analogues of α_1-adrenoceptor antagonist WB4101

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Received 5 October 2004; revised 14 January 2005; accepted 21 January 2005

Abstract—A number of (*S*)- and (*R*)-2-[(2-phenoxyethyl)aminomethyl]-1,4-benzodioxanes unsubstituted or *ortho* monosubstituted at the phenoxy moiety were synthesized and tested in binding assays on the α_{1a}-AR, α_{1b}-AR, α_{1d}-AR and the 5-HT$_{1A}$ receptor. The affinity values of the new compounds **1**–**16** were compared with those of the enantiomers of the 2,6-dimethoxyphenoxy analogue, the well-known α_1 antagonist WB4101, finding that the unsubstituted derivative (*S*)-**1** and the *o*-methyl, the *o*-*t*-butyl, the *o*-fluoro and the *o*-methoxy derivatives, (*S*)-**2**, (*S*)-**4**, (*S*)-**8** and (*S*)-**16**, respectively, display a significantly specific 5-HT$_{1A}$ affinity, very close, with the exception of (*S*)-**4**, to the almost nanomolar one of (*S*)-WB4101. Otherwise, sensible affinity decreases were recorded for the three α_1-AR subtypes. A classical quantitative structure–activity relationship (Hansch) analysis was successfully applied to compounds (*S*)-**1** to (*S*)-**16** and (*S*)-WB4101 to rationalize such binding data.

1. Introduction

The existence of multiple α_1-adrenoceptor (α_1-AR) subtypes, realized within recent years,[1,2] has renewed the interest of medicinal chemists in ligands for such receptors and, more specifically, in subtype selective α_1 antagonists. In fact, current evidences, based on the distribution of α_1-ARs, indicate that subtype selective α_1 antagonists could be very useful in the treatment of lower urinary tract symptoms.[3]

One of the oldest and best-known α_1 antagonists is represented by WB4101, a 2-aminomethyl-1,4-benzodioxane derivative first described in 1965 by Green and co-workers[4] which exhibits about nanomolar affinity towards the three different α_1-ARs (α_{1a}, α_{1b} and α_{1d}) and the 5-HT$_{1A}$ serotoninergic receptor with a slight selectivity for α_{1A}- and, to a minor degree, for α_{1D}-ARs with respect to α_{1B}-AR and 5-HT$_{1A}$ receptor.[5,6] Such a lead molecule has been the subject of intense investigations aimed at identifying its pharmacophoric features and improving its affinity and selectivity.[6–20] As a result, a variety of analogues have been studied over the past decades and, more recently, some of them have been characterized for their α_1 subtype and 5-HT$_{1A}$ affinity. Already in the seventies, Kapur et al. pointed out the contribution of the 2,6-dimethoxyphenyl residue to the antagonistic activity of WB4101 suggesting 'a possible aromatic subsite through which the dimethoxy benzene may interact'.[21] In particular, they hypothesized 'that the 2,6-dimethoxy substituents play an important role in the drug–receptor interaction either indirectly through a conformational influence (on the phenyl ring orientation) or directly through a receptor subsite interaction'.[21] This would be demonstrated by the fact that the antagonistic potency is adversely affected by the removal of one or both *ortho*-methoxy substituents as well as by their repositioning around the phenyl ring or their replacement by methyl groups. More recently (1998), the same authors carried out a QSAR study on WB4101 derivatives enclosing methoxy- and methylphenyl

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Keywords: α_1-Antagonist; WB4101; WB4101 Analogues; α_1-Adrenergic receptor subtypes; 5-HT$_{1A}$ Serotoninergic receptor; Binding affinity; QSAR analysis.

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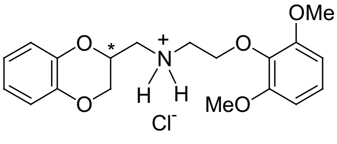
doi:10.1016/j.bmc.2005.01.034

analogues.[22] The results of their investigations confirmed the importance of the ring substitution though 'it was not possible to elucidate the mechanism by which substituents on that ring exert their effect' due to the fact that only methoxy and methyl substituents had been considered.[22]



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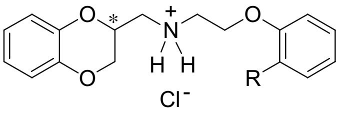
The hypothesized involvement of an aromatic subsite in the α_1 antagonist interaction was successively confirmed by Perez mutagenesis studies,[23,24] which demonstrated that two phenylalanine residues (Phe-312 and Phe-308) in transmembrane domain 7 (TM7) of the α_1-ARs are major sites of α_1-AR antagonist binding. Consistently with these results, our recent researches,[25] based on the automated docking of WB4101 into the α_{1a}-AR built by comparative modeling, showed that the phenoxy moiety of the ligand is inserted in an aromatic pocket lined by Phe-288, Phe-289 (TM6), Phe-308, Phe-312 and Tyr-316 (TM7). In addition, our interaction model supported the original thesis of WB4101 devisers highlighting a hydrogen bond between a glutamine residue (Gln-177) in extracellular loop 2 (EL2) and one ligand methoxy group, which mimics the norepinephrine α-hydroxyl. Furthermore, it is interesting to observe that our docked WB4101 conformer displays, relatively to the 2-phenoxyethyhlamino framework, a pronounced analogy to the two candidate bioactive forms of the ligand we had previously identified on the basis of a thorough study of the conformational properties of WB4101 and of its fitting with the more constrained structure of corynanthine.[19] In fact, these forms show, like the docked conformer, the phenyl ring perpendicular, or nearly, to the oxyethyl plane and a gauche disposition of the nitrogen with respect to the phenoxy oxygen giving rise to an intramolecular hydrogen bond between this latter and one of the ammonium protons. Therefore, a double role would be played by the 2,6-dimethoxy substituents: (a) a specific interaction with an aminoacidic residue of the receptor and (b) a determinant influence on the phenyl orientation and the nitrogen–phenoxy oxygen reciprocal disposition.

In this context, where the phenoxy group and its substitution pattern are generally recognized as key structural determinants for the WB4101 affinity, it is rather surprising that no ring substituents alternative to methoxyl and methyl have been ever considered. Therefore, it seemed worthwhile to investigate, in the first instance, a wide series of WB4101 analogues differently *ortho* monosubstituted at the phenoxy residue in order to get a better insight into the nature of the receptor subsite interaction of this molecular portion and, hopefully, to modulate the affinity profile of the lead compound. To this end, both the enantiomers of compounds **1**–**16** were synthesized and tested in binding assays on the α_{1a}-AR, α_{1b}-AR, α_{1d}-AR and the 5-HT$_{1A}$ receptor. Finally, a classical quantitative structure–activity relationship (Hansch) study was applied, in particular, to the *S* isomers of **1**–**16** in the expectation of finding significant correlations of each of the four biological affinities with properly chosen physicochemical parameters of the aromatic substituents.

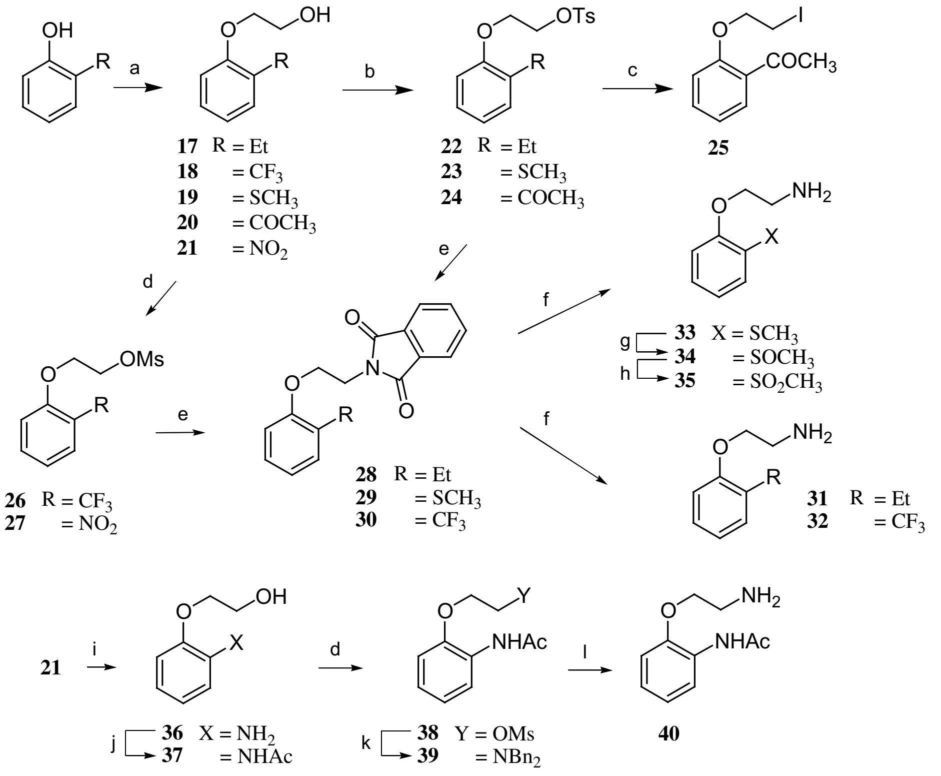
2. Chemistry

The enantiomeric pairs of compounds **1**–**16** were prepared by alkylation of *ortho* substituted 2-phenoxyethylamines with the enantiomers of 2-iodomethyl- or 2-mesyloxymethyl-1,4-benzodioxane or, vice versa, by alkylation of the enantiomers of 2-aminomethyl-1,4-benzodioxane with *ortho* substituted 2-phenoxyethyl halides or mesylates.



1	R = H	**9**	R = Cl
2	= Me	**10**	= SMe
3	= Et	**11**	= SOMe
4	= *t*-Bu	**12**	= SO$_2$Me
5	= CN	**13**	= NO$_2$
6	= CF$_3$	**14**	= NHCOMe
7	= COMe	**15**	= NH$_2$
8	= F	**16**	= OMe

As outlined in Scheme 1, the syntheses of various 2-phenoxyethylamines and 2-phenoxyethyl halides and sulfonates invariably started from the corresponding *ortho* substituted phenols. The synthetic pathways consisted in the following steps: (a) 2-hydroxyethylation of the phenolic moiety with ethylene carbonate; (b) tosylation of the *o*-ethyl, *o*-thiomethyl and *o*-acetyl phenoxyethyl alcohols **17**, **19** and **20**; (c) conversion of this latter into 2-(2-iodoethoxy)acetophenone (**25**); (d) mesylation of the *o*-trifluoromethyl and *o*-nitro intermediates **18** and **21**; (e) N-alkylation of phthalimide with the tosyl esters **22** and **23** and the mesyl ester **26**; (f) hydrazinolysis of the *N*-alkylphthalimides **28**–**30** to give the target 2-phenoxyethylamines **31**–**33**; (g) oxidation of the thioamine **33** to the corresponding sulfoxide **34**; (h) oxidation of this latter to the sulfone **35**. As shown in the same scheme, 2-(2-nitrophenoxy)ethanol (**21**) was also used to synthesize 2-(2-acetamidophenoxy)ethylamine (**40**) by the following steps: (i) reduction to the aniline **36**; (j) N-acetylation; (d) O-mesylation; (k) aminolysis with dibenzylamine; (l) hydrogenolytic debenzylation to yield **40**. 2-Phenoxyethylamine, the other *ortho* substituted 2-phenoxyethylamines (*o*-CN, *o*-F, *o*-Cl, *o*-MeO, *o*-*t*-BuO) and 2-(2-chloroethoxy)anisole were prepared according to the literature methods, while the enantiomers of 2-iodomethyl-, 2-mesyloxymethyl- and 2-aminomethyl-1,4- benzodioxane were obtained practicing our previously reported synthetic methods.[6,19,26]

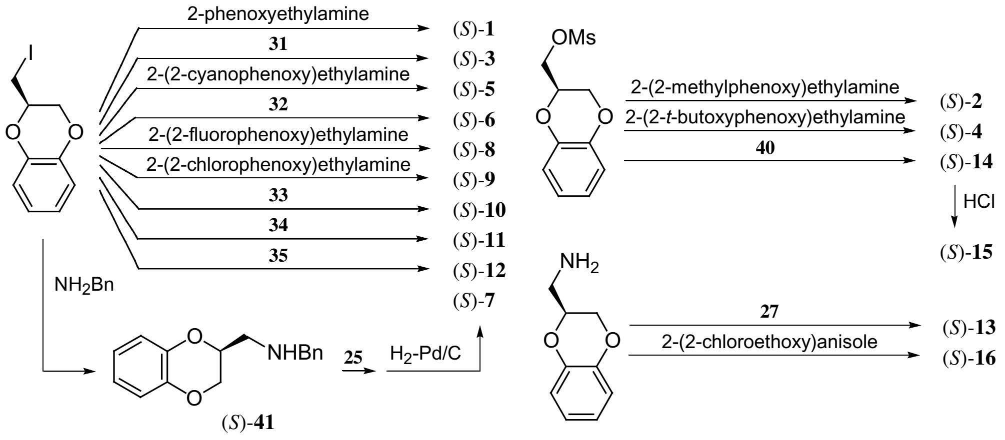


Scheme 1. Reagents: (a) ethylene carbonate, K_2CO_3, DMF or toluene or xylene; (b) TsCl, Py. (c) NaI, acetone; (d) MsCl, TEA, DCM; (e) potassium phthalimide, DMF; (f) hydrazine, 2-methoxyethanol; (g) 30% H_2O_2; (h) *m*-CPBA, DCM; (i) H_2–Pd/C, MeOH; (j) CH_3COCl, TEA, DCM; (k) $NHBn_2$, 2-propanol; (l) H_2–Pd/C, 2.5/1 EtOH–EtOAc.

The final nucleophilic substitutions leading to the *S* isomers of **1**–**16** are illustrated in Scheme 2. The *R* isomers of **1**–**16** were synthesized by the same reactions as their antipodes, but using the 2-substituted 1,4-benzodioxane intermediates with inverted configuration. In particular, compounds **1**–**6**, **8**–**12**, **14** and **15** were prepared by N-alkylation of *ortho* substituted 2-phenoxyethylamines, whereas compounds **7**, **13** and **16** by aminolysis of *ortho* substituted 2-phenoxyethyl halides or mesylates. In the case of **7**, the enantiomers of 2-*N*-benzylaminomethyl-1,4-benzodioxane (**41**) were alkylated with **25** and the resultant tertiary amines submitted to hydrogenolysis, while the enantiomeric pairs of **13** and **16** were directly prepared by reaction of (*S*)- and (*R*)-2-aminomethyl-1,4-benzodioxane with **27** and 2-(2-chloroethoxy)anisole, respectively.

3. Results and discussion

Table 1 reports the affinities, expressed as pK_i values, at the three cloned human α_1-AR subtypes and 5-HT_{1A}



Scheme 2. Synthesis of the *S* isomers of compounds **1**–**16**.

Table 1. Experimental and predicted affinity constants, expressed as pK_i (−logK_iM), of the enantiomers of compounds **1**–**16** and of WB4101 for cloned human α_1-adrenoceptor subtypes and 5-HT$_{1A}$ receptor

Phenoxy *o*-substituent	Compd	α–**1a**		α–**1b**		α–**1d**		5-HT$_{1A}$	
		p$K_{\text{i exptl}}$ ± SE	p$K_{\text{i pred}}$	p$K_{\text{i exptl}}$ ± SE	p$K_{\text{i pred}}$	p$K_{\text{i exptl}}$ ± SE	p$K_{\text{i pred}}$	p$K_{\text{i exptl}}$ ± SE	p$K_{\text{i pred}}$
H	(*S*)-**1**	7.34 (±0.05)	7.30	7.02 (±0.03)	7.01	7.57 (±0.08)	7.31	8.57 (±0.07)	8.40
	(*R*)-**1**	7.07 (±0.08)		6.03 (±0.12)		7.26 (±0.12)		7.36 (±0.09)	
Me	(*S*)-**2**	7.51 (±0.04)	7.17	7.13 (±0.05)	7.03	7.10 (±0.05)	7.31	8.55 (±0.04)	8.21
	(*R*)-**2**	7.16 (±0.07)		6.73 (±0.03)		6.79 (±0.08)		7.66 (±0.08)	
Et	(*S*)-**3**	7.58 (±0.14)	6.80	7.06 (±0.07)	6.90	8.01 (±0.17)	7.65	7.26 (±0.04)	7.43
	(*R*)-**3**	7.40 (±0.07)		6.91 (±0.05)		8.06 (±0.30)		6.36 (±0.11)	
t-Bu	(*S*)-**4**	6.22 (±0.04)	6.20	6.29 (±0.08)	6.72	6.12 (±0.04)	6.20	7.59 (±0.03)	7.49
	(*R*)-**4**	6.22 (±0.06)		5.78 (±0.05)		5.37 (±0.09)		6.34 (±0.11)	
CN	(*S*)-**5**	7.06 (±0.09)	6.92	6.05 (±0.08)	6.80	8.17 (±0.22)	8.47	7.15 (±0.06)	6.92
	(*R*)-**5**	7.22 (±0.07)		6.72 (±0.06)		7.31 (±0.19)		7.49 (±0.04)	
CF_3	(*S*)-**6**	7.16 (±0.04)	6.57	6.99 (±0.08)	6.67	7.77 (±0.18)	7.17	6.75 (±0.05)	7.34
	(*R*)-**6**	7.05 (±0.28)		6.72 (±0.08)		7.73 (±0.13)		6.18 (±0.14)	
COMe	(*S*)-**7**	7.26 (±0.04)	6.81	6.73 (±0.05)	6.77	7.54 (±0.19)	7.50	7.17 (±0.08)	7.21
	(*R*)-**7**	7.63 (±0.06)		7.28 (±0.10)		7.91 (±0.27)		7.57 (±0.07)	
F	(*S*)-**8**	7.47 (±0.10)	7.79	7.27 (±0.04)	7.35	7.16 (±0.05)	7.76	8.52 (±0.02)	8.81
	(*R*)-**8**	7.15 (±0.05)		6.57 (±0.06)		7.72 (±0.24)		7.21 (±0.07)	
Cl	(*S*)-**9**	7.62 (±0.07)	7.27	7.24 (±0.29)	7.08	8.08 (±0.32)	8.00	7.96 (±0.04)	7.88
	(*R*)-**9**	7.27 (±0.07)		6.73 (±0.09)		7.80 (±0.23)		6.87 (±0.05)	
SMe	(*S*)-**10**	7.72 (±0.10)	7.08	7.46 (±0.26)	7.04	8.34 (±0.17)	7.75	7.62 (±0.08)	7.65
	(*R*)-**10**	7.58 (±0.14)		7.06 (±0.07)		8.01 (±0.17)		7.35 (±0.06)	
SOMe	(*S*)-**11**	7.17 (±0.07)	6.75	6.90 (±0.04)	6.75	7.11 (±0.11)	7.26	7.33 (±0.04)	7.23
	(*R*)-**11**	n.d.		n.d.		n.d.		n.d.	
SO_2Me	(*S*)-**12**	6.98 (±0.15)	6.62	7.00 (±0.04)	6.75	6.69 (±0.06)	7.03	6.96 (±0.04)	7.31
	(*R*)-**12**	n.d.		n.d.		n.d.		n.d.	
NO_2	(*S*)-**13**	7.31 (±0.27)	6.94	7.24 (±0.12)	6.82	7.96 (±0.13)	7.62	7.44 (±0.06)	7.48
	(*R*)-**13**	7.07 (±0.11)		6.41 (±0.14)		8.09 (±0.29)		6.95 (±0.06)	
NHCOMe	(*S*)-**14**	7.46 (±0.19)	7.44	6.81 (±0.14)	7.23	7.73 (±0.03)	8.00	8.16 (±0.43)	7.73
	(*R*)-**14**	7.26 (±0.25)		6.23 (±0.13)		7.28 (±0.16)		7.09 (±0.03)	
NH_2	(*S*)-**15**	7.26 (±0.11)	7.52	6.88 (±0.10)	7.19	7.88 (±0.17)	7.41	8.06 (±0.08)	8.54
	(*R*)-**15**	6.72 (±0.13)		5.91 (±0.17)		7.51 (±0.16)		6.58 (±0.13)	
OMe	(*S*)-**16**	7.90 (±0.09)	7.91	7.37 (±0.14)	7.50	7.52 (±0.04)	8.06	8.67 (±0.02)	8.62
	(*R*)-**16**	7.61 (±0.09)		6.94 (±0.04)		6.95 (±0.07)		8.37 (±0.05)	
	(*S*)-WB4101	9.39 (±0.06)	8.51	8.24 (±0.04)	7.99	9.29 (±0.11)	8.82	8.61 (±0.04)	8.84
	(*R*)-WB4101	7.95 (±0.04)		7.14 (±0.06)		7.98 (±0.08)		7.39 (±0.03)	

serotoninergic receptor of the enantiomeric pairs of compounds **1**–**16** and, as comparison terms, of (*S*)- and (*R*)-WB4101.

Considering the binding data, it is possible to observe that the affinities of the *S* enantiomers are generally higher than those of their antipodes, the only significant exceptions being represented by the cyano and the acetyl substituted analogues, **5** and **7**, respectively, at the α_{1a} and α_{1b} AR subtypes and the 5-HT$_{1A}$ receptor. Anyhow, the eudismic indexes are relatively small, exceeding the unity in the only cases of the 5-HT$_{1A}$ affinity displayed by the enantiomeric pairs of **1**, **4**, **8** and **14**.

Compared to the respective homochiral enantiomers of WB4101, the *ortho* monosubstituted phenoxy derivatives **1**–**16** are, for the most part, less potent. In particular, all the *S* isomers, excepting the monomethoxy derivative (*S*)-**16** at the 5-HT$_{1A}$ receptor, display lower affinities than (*S*)-WB4101 and the gap is more pronounced in the case of the three α_1 subtypes. Otherwise, a few exceptions can be counted among the *R* enantiomers, especially at the 5-HT$_{1A}$ receptor. Indeed, the 5-HT$_{1A}$ affinities of (*R*)-**2**, (*R*)-**5**, (*R*)-**7** and, above all, of (*R*)-**16** exceed that of the WB4101 *R* enantiomer.

A little more detailed analysis of the binding data for each of the four receptors reveals quite different affinity decreases with respect to the 2,6-dimethoxy substituted lead compound. In the case of the α_{1a}-AR, a generalized and considerable drop was observed from the subnanomolar affinity of (*S*)-WB4101 to the values around 0.1 μM. More moderate decreases were instead registered for the α_{1b} affinity, but analogously resulting in nearly 0.1 μM K_i values due to the lower affinity of the lead compound towards this AR subtype than for the α_{1a}-AR. For the third subtype, the α_{1d}-AR, a large minority of compounds, including *S* and *R* isomers, showed K_i around 10 nM, thus approximating the potencies of (*S*)- and (*R*)-WB4101. Finally, more than one of the tested compounds displayed almost nanomolar 5-HT$_{1A}$ affinity, sometimes surpassing the corresponding homochiral enantiomers of WB4101. Furthermore, in the case of (*S*)-**1**, (*S*)-**2**, (*S*)-**4**, (*S*)-**8** and (*S*)-**16**, the very high or remarkable affinity for the serotoninergic receptor was associated with a significant selectivity with respect to the three α_1-AR subtypes.

While such a bird's-eye view of the values listed in Table 1 makes evident some significantly distinctive trends of the four groups of binding data, a qualitative SAR analysis of these latter appears problematic and little constructive considering that the large variety in substituent properties does not always match with a very wide range of affinity values. In fact, excluding the binding data of (*S*)-**4** and (*R*)-**4**, which frequently behave as modestly potent outliers, the pK_i range keeps almost unaltered for both the *R* and *S* isomers only at the 5-HT$_{1A}$ receptor ($\approx$2) and the α_{1b}-AR ($\approx$1.4), whereas it sensibly decreases at the other two ARs, even dropping to 0.9 in the case of the α_{1a} subtype. This notwithstanding, the mass of data was judged suitable material for a QSAR investigation, which could elicit latent correlations between the physicochemical parameters of the aromatic substituents and the biological data and sort out some determinants for the selectivity modulation. Obviously, a broad range of affinities facilitates the recognition of QSAR relationships. Therefore, in order to expand the affinity scale, the enantiomers of WB4101, in addition to those of **4**, were included in the series and the best linear correlations were searched between each of the four affinities and the physicochemical parameter values resulting from the sum of the contributions of the two *ortho* substituents, namely two methoxy groups in WB4101, a hydrogen atom and another substituent in compounds **2**–**16** and two hydrogen atoms in compound **1**. Thus, the pK_i range was enlarged over two or three units also for the α_1 affinities, in particular, of the *S* isomers, whose QSAR investigation is reported here in detail.

A classical Hansch multivariate regression analysis using the least-squares method was carried out to derive QSAR equations for our data sets. For each of the four receptors, the equation adopted as the final model is given in Table 2 and the resultant predicted pK_i values for all the *S* isomers are listed in Table 1, where they can be compared with the corresponding experimental ones. The level of significance of the results was judged by the squared correlation coefficient (r^2), the squared cross-validated correlation coefficient (q^2), the standard error of the estimates (SE) and the Fisher significance ratio (*F*). The statistical relevance was also assured by a >5 ratio of compounds ($n = 17$) to variables (**3**) and a reasonably extended pK_i range. The significance of each regression coefficient was expressed by its standard error. The substituent effects were described by four physicochemical parameters, suitably chosen out of 27. As electronic parameter, the resonance constant of Swain and Lupton ($\mathscr{R}$)[27] was used, while the fragmental volume of aromatic substituents (V)[28] and the length parameter of Verloop (L)[29] were employed as steric descriptors. The ability of accepting H bonds was parameterized by the HBβ[30] value derived from the fragmental system Systahl 2.0. The values of the input parameters are shown in Table 3.

For the α_{1a}-AR, Eq. 1 (Table 2) shows that the substituent volume exerts an adverse effect, while the negative regression coefficient of the resonance parameter indicates that the affinity values benefit from electron donor substituents. Furthermore, the positive sign of HBβ is consistent with the stabilizing role of hydrogen bond for the α_{1a}-AR interaction, already postulated for the reference ligand WB4101. The same parameters with the same coefficient sign are found in the equation for

Table 3. Values of the physicochemical parameters[a] used in the QSAR study

Compd	Subst	$\mathscr{R}$[b]	V[c]	L[d]	HBβ[e]
(*S*)-**1**	H	0	9.24	4.12	0
(*S*)-**2**	Me	−0.13	22.7	4.93	0
(*S*)-**3**	Et	−0.1	36.16	6.17	0
(*S*)-**4**	$C(CH_3)_3$	−0.13	63.08	6.17	0
(*S*)-**5**	CN	0.19	22	6.29	0.23
(*S*)-**6**	CF_3	0.19	31.12	5.36	0.09
(*S*)-**7**	Ac	0.2	37.16	6.12	0.51
(*S*)-**8**	F	−0.34	11.41	4.71	0.03
(*S*)-**9**	Cl	−0.15	20.32	5.58	0.01
(*S*)-**10**	SMe	−0.18	37.7	6.36	0.2
(*S*)-**11**	SOMe	0.22	42.50	6.17	0.62
(*S*)-**12**	SO_2Me	0.1	47.04	6.17	0.40
(*S*)-**13**	NO_2	0.16	25.08	5.5	0.28
(*S*)-**14**	NHAc	−0.26	46.03	7.15	0.67
(*S*)-**15**	NH_2	−0.18	19.53	4.84	0.21
(*S*)-**16**	OMe	−0.51	27.76	6.04	0.3
(*S*)-WB4101	2,6-OMe	−1.02	46.28	7.96	0.6

[a] For each parameter, the reported value is the sum of the contributions of the two substituents in 2 and in 6. For compounds **2**–**16**, one of them is invariably a hydrogen atom. For compound **1**, both substituents are a hydrogen atom.

[b] Data taken from Ref. 27.

[c] Data taken from Ref. 28.

[d] Data taken from Ref. 29.

[e] Data taken from Ref. 30.

Table 2. Equations for QSAR of the α_1-ARs and the 5-HT$_{1A}$ receptor

Receptor	Equations	*n*	r^2	q^2	SE	*F*
α_{1a}	(1) $pK_i = 7.52(\pm 0.19) + 0.94(\pm 0.22)HB\beta - 1.51(\pm 0.28)\mathscr{R} - 0.024(\pm 0.007)V$	17	0.78	0.74	0.34	15.50
α_{1b}	(2) $pK_i = 7.08(\pm 0.21) + 0.37(\pm 0.11)HB\beta - 1.03(\pm 0.18)\mathscr{R} - 0.0079(\pm 0.0073)V$	17	0.53	0.45	0.36	4.96
α_{1d}	(3) $pK_i = 4.22(\pm 0.27) - 0.17(\pm 0.06)\mathscr{R} + 0.87(\pm 0.19)L - 0.054(\pm 0.012)V$	17	0.71	0.68	0.41	11.07
5-HT$_{1A}$	(4) $pK_i = 10.79(\pm 0.54) + 0.79(0.13)HB\beta - 0.58(\pm 0.11)L - 2.15(\pm 0.16)\mathscr{R}$	17	0.85	0.81	0.27	24.31

α_{1b}-AR affinity (Eq. 2, Table 2). This means that the physicochemical properties favouring α_{1a} and α_{1b} affinities are quite constant. Conversely, the statistical goodness of Eq. 2 is significantly minor than that for α_{1a}-AR and this may suggest that the role of the phenoxy moiety is not so important for the α_{1b} as for the α_{1a} interaction. For the α_{1d}-AR, the best equation (Eq. 3, Table 2) lacks the HBβ parameter while including an additional steric descriptor (L). The positive sign of this latter, combined with the invariably negative one of V, would mean that the phenoxy residue is inserted in a cavity, which is size-limited in all three AR subtypes, but can accept, in the case of the α_{1d}-AR, substituents of rather elongated than spherical shape.

Finally, the last equation (Eq. 4, Table 2) shows that electron-richness (see the negative coefficient of $\mathscr{R}$) and ability to accept H bonds (see the positive coefficient of HBβ) are beneficial properties also for the 5-HT$_{1A}$ receptor affinity, which is however considerably lowered by the substituent length. In particular, the negative coefficient of L indicates that substituents of spherical shape are better accepted, as in the case of (*S*)-**1**, (*S*)-**2**, (*S*)-**8**, which exhibit almost nanomolar 5-HT$_{1A}$ affinities and of (*S*)-**4**, whose remarkable affinity for the serotoninergic receptor (26 nM) is 20–30-fold more higher than for the α_1-ARs. Furthermore, differently from the QSARs for the α_1 receptors, this significant 5-HT$_{1A}$ affinity, in connection with such a bulky substituent, cancels the negative effects of the V parameter making it uninfluential in the present series of substituents as demonstrated by unsuccessful attempts to introduce this descriptor into Eq. 4.

The four equations were recalculated maintaining the same physicochemical parameters, but excluding (*S*)-WB4101, the only compound in the series without *ortho* hydrogens at the phenoxy residue. The r^2 values of the new relations were 0.83 (5-HT$_{1A}$ receptor), 0.64 (α_{1a}-AR), 0.63 (α_{1d}-AR) and 0.27 (α_{1b}-AR). Such results indicate that, apart from the QSAR for the α_{1b}-AR, these equations were still significant, retaining, in the case of the 5-HT$_{1A}$ receptor, a very high correlation. Furthermore, it is noteworthy that the equations recalculated for the α_{1a}, α_{1d} and 5-HT$_{1A}$ receptors without (*S*)-WB4101 maintained the original coefficient signs, excepting the $\mathscr{R}$ coefficient in the α_{1d}-AR, which changed from a slightly negative value (−0.17) to +0.36. This suggests that the inclusion of (*S*)-WB4101 in the present analysis does not subvert the QSARs of the *ortho* monosubstituted analogs, but somehow validates them. In fact, as expected on the basis of its very high affinities associated with the presence of two identical *ortho* substituents, (*S*)-WB4101 enhances the statistical significance of such relationships and makes their nature clearer.

4. Conclusions

The present QSAR study on a wide series of (*S*)-WB4101 *S* analogues differently *ortho* monosubstituted at the phenoxy residue indicates that the site interacting with this moiety is subjected to tight steric constraints in both α_1 and 5-HT$_{1A}$ receptors. In fact, steric descriptors are present in all the equations generally as the terms of great weight and, excepting L in the α_{1d} equation, their effect is always negative. Contrariwise, the electron-richness is invariably a beneficial property and the ability to accept H bonds increases the affinity for three receptors (α_{1a},α_{1b} and 5-HT$_{1A}$) of the four considered. However, such a similarity does not imply that the *ortho* monosubstitution at the phenoxy residue exerts univocal effects on the affinity for the α_1-ARs and the 5-HT$_{1A}$ receptor. Indeed, the different incidence of the steric and the electronic parameters results in variegated affinity profiles leading, in the case of (*S*)-**1**, (*S*)-**2**, (*S*)-**4**, (*S*)-**8** and (*S*)-**16**, to high 5-HT$_{1A}$ affinities combined with a specificity for this receptor experimentally even more significant than that predicted.

On the basis of these equations, it is difficult to rationalize the affinity shift of this series of *ortho* monosubstituted phenoxy derivatives towards the 5-HT$_{1A}$ receptor and, to a minor extent, the α_{1d}-AR. In this regard, though prediction is neither the proper faculty nor the main goal of QSARs, it is interesting to observe that the equations for the α_{1a} and α_{1b} receptors are so structured as to let no *ortho* monosubstituted derivative compete with the 2,6-dimethoxy substituted reference compound (*S*)-WB4101. This would be possible if the volumetric term had a more negative incidence. Such a condition is realized, in connection with a much lower influence of the $\mathscr{R}$ parameter, in the α_{1d} equation, but it is counteracted by the concomitant largely positive effect of the substituent length. Again, like in the two previous QSARs, the α_{1d} affinity of the monosubstituted derivatives, though generally higher than for the α_{1a} and α_{1b} ARs, seems to be inevitably handicapped in comparison with that of (*S*)-WB4101. Finally, the situation sensibly changes in the case of the 5-HT$_{1A}$ receptor as a proof of its different nature. Negative values of $\mathscr{R}$ exert the most beneficial effect and only the length of the substituents becomes detrimental while their volume is uninfluential. This results in many *ortho* monosubstituted derivatives, bearing an electron-donor and not too long substituent, which show calculated and experimental 5-HT$_{1A}$ affinities comparable with that of (*S*)-WB4101 and not far from those predicted for the corresponding *ortho* disubstituted analogues. It is significant that the exclusion of (*S*)-WB4101 from the QSAR analysis is uninfluential only in the case of the 5-HT$_{1A}$ receptor.

In summary, the present results demonstrate the already theorized importance of the 2,6-disubstitution, in particular with two methoxy groups, at the phenyl residue of 2-[(2-phenoxyethyl)aminomethyl]-1,4-benzodioxane for the interaction with the α_1-ARs and, especially, with the α_{1a}-AR subtype. Otherwise, the same results and their QSAR analysis clearly indicate that such a feature is not essential to the interaction with the 5-HT$_{1A}$ receptor as proved by the high and specific 5-HT$_{1A}$ affinity of the unsubstituted derivative (*S*)-**1** as well as of several WB4101 analogues suitably *ortho* monosubstituted at the phenyl ring.

5. Experimental

5.1. Chemistry

Melting points were measured on Buchi melting point apparatus and are uncorrected. ^{1}H NMR spectra were recorded operating at 60 MHz, 200 MHz or 300 MHz. Chemical shifts are reported in parts per million relative to residual solvent ($CHCl_3$ or DMSO) as internal standard. Signal multiplicity is designed according to the following abbreviations: s = singlet, d = doublet, dd = doublet of doublets, t = triplet, m = multiplet, br s = broad singlet, br t = broad triplet. Optical rotations were determined by a Perkin-Elmer 241 Polarimeter at 25 °C. Elemental analyses (CHN) of the new substances are within 0.40% of theoretical values. Purifications were performed by flash chromatography using silica gel (particle size 40–63 μm, Merck).

5.1.1. 2-(2-Ethylphenoxy)ethanol (17). A mixture of 2-ethylphenol (20 g, 164 mmol), ethylene carbonate (28.86 g, 328 mmol) and potassium carbonate (22.6 g, 164 mmol) in toluene (70 mL) was refluxed for 2 h. After cooling at room temperature, water was added. The aqueous layer was separated and extracted with toluene twice. The organic phase was treated with 10% NaOH (150 mL), washed with water twice (200 mL), dried and concentrated to give the crude product, which was purified by chromatography on silica gel. Elution with cyclohexane–ethyl acetate (90:10) yielded 21 g (77%) of **17** as an oil. ^{1}H NMR ($CDCl_3$) δ 1.28 (t, 3H), 2.45 (br s, 1H), 2.65 (q, 2H), 4.3–4.37 (m, 4H), 6.7–7.4 (m, 4H). Anal. Calcd for $C_{10}H_{14}O_2$ (166.22).

5.1.2. 2-(2-Trifluoromethylphenoxy)ethanol (18). Prepared from *ortho*-trifluoromethylphenol as described for **17** but replacing toluene with DMF. After chromatography on silica gel (cyclohexane–ethyl acetate 70:30) the product was isolated as a white solid (64%): mp 75.8 °C; ^{1}H NMR ($CDCl_3$) δ 2.20 (br s, 1H), 3.94 (m, 2H), 4.15 (m, 2H), 6.97–7.05 (m, 2H), 7.43–7.57 (m, 2H). Anal. Calcd for $C_9H_9F_3O_2$ (206.16).

5.1.3. 2-(2-Methylthiophenoxy)ethanol (19). Prepared from 2-methylthiophenol as described for **17**. Crystallization from diethyl ether yielded **19** as a white solid (84%): mp 40–42 °C; ^{1}H NMR ($CDCl_3$) δ 2.4 (s, 3H), 4.28 (t, 2H), 4.4–4.9 (br s, 3H), 6.7–7.5 (m, 4H). Anal. Calcd for $C_9H_{12}O_2S$ (184.25).

5.1.4. 2-(2-Acetylphenoxy)ethanol (20). Prepared from 2-hydroxyacetophenone as described for **17**. After chromatography on silica gel the product was isolated as a yellow oil (48%): ^{1}H NMR ($CDCl_3$) δ 1.60 (br s, 1H), 2.60 (s, 3H), 4.05 (m, 2H), 4.25 (m, 2H), 6.60–7.79 (m, 4H). Anal. Calcd for $C_{10}H_{12}O_3$ (180.203).

5.1.5. 2-(2-Nitrophenoxy)ethanol (21). Prepared as described for **17** but replacing toluene with DMF. The resulting crude product, isolated as a yellow oil (57%), was used for the subsequent step without further purification: ^{1}H NMR ($CDCl_3$) 3.42 (s, 1H), 3.84 (t, 2H), 4.09 (t, 2H), 6.89 (t, 1H), 6.99 (d, 1H), 7.39 (dt, 1H), 7.79 (dd, 1H). Anal. Calcd for $C_8H_9NO_4$ (183.164).

5.1.6. 2-(2-Ethylphenoxy)-1-tosyloxyethane (22). Tosyl chloride (22.8 g, 120 mmol) was added in small portions to a stirred solution of **17** (20 g, 120 mmol) in pyridine (70 mL) at 0 °C. The resulting mixture was stirred at room temperature overnight, diluted with diethyl ether (100 mL) and washed with 10% HCl. The aqueous layer was separated and extracted with diethyl ether twice. The organic phases were combined, dried and concentrated to give 25.5 g (66%) of **22** as a white solid: mp 79–80 °C; ^{1}H NMR ($CDCl_3$) δ 1.1 (t, 3H), 2.45 (s, 3H), 2.55 (q, 2H), 4.2 (t, 2H), 4.4 (t, 2H), 6.6–7.2 (m, 4H), 7.34 (d, 2H), 7.85 (d, 2H). Anal. Calcd for $C_{17}H_{20}O_4S$ (320.40).

5.1.7. 2-(2-Methylthiophenoxy)-1-tosyloxyethane (23). Prepared from **19** as described for **22** and isolated after crystallization from cyclohexane/ethyl acetate (1:1) as a white solid (64%): mp 71–73 °C; ^{1}H NMR ($CDCl_3$) δ 2.37 (s, 3H), 2.42 (s, 3H), 4.0–4.6 (t, 4H), 6.0–7.2 (m, 4H), 7.32 (d, 2H), 7.85 (d, 2H). Anal. Calcd for $C_{16}H_{18}O_4S_2$ (338.44).

5.1.8. 2-(2-Acetylphenoxy)-1-tosyloxyethane (24). Prepared from **20** as described for **22** and isolated after crystallization from hexane/ethyl acetate (1:1) as a white solid (43%): mp 87 °C; ^{1}H NMR ($CDCl_3$) δ 2.4 (s, 3H), 2.50 (s, 3H), 4.60–4.80 (m, 4H), 6.80–8.00 (m, 8H). Anal. Calcd for $C_{17}H_{18}O_5S$ (334.39).

5.1.9. 2-(2-Acetylphenoxy)-1-iodoethane (25). A mixture of **24** (4.7 g, 14 mmol) and sodium iodide (24.0 g, 140 mmol) in acetone (50 mL) was refluxed for 12 h. The solvent was evaporated and the residue treated with 10% HCl and diethyl ether. The aqueous layer was separated and extracted with diethyl ether twice. The organic phases were combined, dried and concentrated to give the crude product, which was purified by chromatography on silica gel. Elution with *n*-hexane/ethyl acetate (90:10) afforded 3.5 g (85%) of **25** as a solid: mp 45–47 °C; ^{1}H NMR ($CDCl_3$) δ 2.70 (s, 3H), 3.50 (t, 2H), 4.40 (t, 2H), 6.80–7.90 (m, 4H). Anal. Calcd for $C_{10}H_{11}IO_2$ (290.10).

5.1.10. 2-(2-Trifluoromethylphenoxy)-1-mesyloxyethane (26). Mesyl chloride (0.98 mL, 12.6 mmol) was added dropwise to a stirred solution of **18** (2.47 g, 11.98 mmol) and TEA (1.75 mL) in dichloromethane at 0 °C. After 2 h at room temperature the reaction mixture was washed with a saturated solution of $NaHCO_3$, dried and concentrated to give the crude product 3.35 g (98.5%) as a white solid which was used for the subsequent step without further purification: mp 51.5 °C; ^{1}H NMR ($CDCl_3$) δ 3.07 (s, 3H), 4.35 (m, 2H), 4.58 (m, 2H), 6.97–7.10 (m, 2H), 7.48–7.60 (m, 2H). Anal. Calcd for $C_{10}H_{11}F_3O_4S$ (284.25).

5.1.11. 2-(2-Nitrophenoxy)-1-mesyloxyethane (27). Prepared from **21** as described for **26** and isolated as an orange oil (98%): ^{1}H NMR ($CDCl_3$) δ 3.04 (s, 3H), 4.32 (m, 2H), 4.52 (m, 2H), 6.98–7.08 (m, 2H), 7.5 (t, 1H), 7.74 (d, 1H). Anal. Calcd for $C_9H_{11}NO_6S$ (261.16).

5.1.12. *N*-[2-(2-Ethylphenoxy)ethyl]phthalimido (28). Sodium hydride (1.46 g, 50.8 mmol) was suspended in DMF (35 mL) under N_2 atmosphere. A solution of phthalimide (7.46 g, 50.8 mmol) in DMF (35 mL) was added dropwise and the resulting mixture was heated at 70 °C for 1 h. Then **22** (19.0 g, 59.3 mmol) was diluted in DMF (25 mL) and added stepwise to the stirring solution. The resulting mixture was refluxed for 3 h and after cooling at room temperature was quenched into frozen water (150 mL). The resulting precipitate was isolated yielding 15.60 g (90%) of **28** as a white solid: mp 61–63 °C; ^1H NMR ($CDCl_3$) δ 1.1 (t, 3H), 2.24 (d, 2H), 4.18 (s, 4H), 6.6–7.4 (m, 4H), 7.5–8.0 (m, 4H). Anal. Calcd for $C_{18}H_{17}NO_3$ (295.34).

5.1.13. *N*-[2-(2-Methylthiophenoxy)ethyl]phthalimido (29). Prepared from **23** as described for **28**: mp 117–120 °C: ^1H NMR ($CDCl_3$) δ 2.3 (s, 3H), 4.22 (m, 4H), 6.7–7.4 (m, 4H), 7.5–8.15 (m, 4H). Anal. Calcd for $C_{17}H_{15}NO_3S$ (313.37).

5.1.14. *N*-[2-(2-Trifluoromethylphenoxy)ethyl]phthalimido (30). Prepared from **26** as described for **28**: mp 126 °C; ^1H NMR ($CDCl_3$) δ 4.17 (t, 2H), 4.32 (t, 2H), 6.95–7.00 (m, 2H), 7.46 (m, 2H), 7.71 (m, 2H), 7.85 (m, 2H). Anal. Calcd for $C_{17}H_{12}F_3O_3N$ (335.28).

5.1.15. 2-(2-Ethylphenoxy)ethylamine (31). Hydrazine hydrate (2.7 mL) was added dropwise to a stirred mixture of **28** (15 g, 50.8 mmol) in 2-methoxyethanol at 125 °C. After refluxing for 2 h 10% HCl was added until pH = 3. After cooling, the reaction mixture was concentrated, the residue treated with 10% NaOH (100 mL) and washed with diethyl ether. The organic phases were combined, dried and concentrated. Distillation under vacuum (bp 75 °C at 0.5 mbar) of the residue gave 5.2 g (62%) of **31** as a colourless oil: ^1H NMR ($CDCl_3$) δ 1.04 (t, 3H), 1.30 (br s, 2H), 2.52 (q, 2H) 2.91 (t, 2H), 3.81 (t, 2H), 6.5–7.4 (m, 4H). Anal. Calcd for $C_{10}H_{15}NO$ (165.23).

5.1.16. 2-(2-Trifluoromethylphenoxy)ethylamine (32). Prepared from **30** as described for **31** and isolated, after chromatography on silica gel (dichloromethane/isopropanol 95:5), as a yellow oil (88.7%): ^1H NMR ($CDCl_3$) δ 1.62 (br s, 2H), 3.11 (t, 2H), 4.07 (t, 2H), 6.96–7.05 (m, 2H), 7.41–7.58 (m, 2H). Anal. Calcd for $C_9H_{10}F_3ON$ (205.18).

5.1.17. 2-(2-Methylthiophenoxy)ethylamine (33). Prepared from **29** as described for **31** and isolated after distillation under vacuum (bp 108 °C at 0.8 mbar) as an oil (42%): ^1H NMR ($CDCl_3$) δ 1.7–2.1 (m, 1H), 2.3–2.6 (m, 1H), 2.4 (s, 3H), 3.12 (t, 2H), 4.12 (t, 2H), 6.7–7.5 (m, 4H). Anal. Calcd for $C_9H_{13}NOS$ (183.27).

5.1.18. 2-(2-Methylsulfinylphenoxy)ethylamine (34). 30% H_2O_2 (10.2 mL) was added to a solution of **33** hydrochloride (14 g, 63.70 mmol) in water (40 mL). The resulting mixture was heated at 90 °C for 4 h. After cooling at room temperature, water was removed and the residue was dissolved with ethanol, which was then concentrated. The same treatment was done for four times. At the end **34** hydrochloride was isolated as white solid 10 g (67%): ^1H NMR ($CDCl_3$) δ 2.60 (s, 3H), 4.05 (m, 2H), 4.25 (m, 2H), 6.60–7.79 (m, 4H). Anal. Calcd for $C_9H_{13}O_2SN$ (199.26).

5.1.19. 2-(2-Methylsulfonylphenoxy)ethylamine (35). 3-Chloroperbenzoic acid (12 g, 28 mmol) was added to a stirring solution of **34** (5 g, 27.5 mmol) in dichloromethane (40 mL) at 0 °C. The reaction mixture was stirred at room temperature for 24 h. At the end of the reaction, the mixture was washed with a saturated solution of $NaHCO_3$. The organic phase was separated, dried and concentrated to give **35** (3.75 g, 63%) as an oil: ^1H NMR ($CDCl_3$) δ 2.10 (s, 2H), 3.25 (s, 3H), 4.25 (t, 2H), 7.00–8.15 (m, 4H). Anal. Calcd for $C_9H_{13}O_3SN$ (215.26).

5.1.20. 2-(2-Aminophenoxy)ethanol (36). A solution of **21** (11 g, 60 mmol) in methanol (220 mL) was added with 10% Pd/C (1 g) and vigorously shaken under hydrogen at room temperature. The catalyst was removed by filtration and the filtrate concentrated to give **36** (9.02 g) as a red solid: mp 90.9 °C; ^1H NMR ($CDCl_3$) δ 2.6–3.3 (br s, 3H), 3.97 (t, 2H), 4.12 (t, 2H), 6.6–6.9 (m, 4H). Anal. Calcd for $C_8H_{11}NO_2$ (153.18).

5.1.21. 2-(2-Acetlyaminophenoxy)ethanol (37). Acetyl chloride (7.82 mL, 0.110 mmol) was added stepwise to a cool solution of **36** (7.99 g, 52 mmol) and TEA (15.33 mL, 0.110 mmol) in dichloromethane (48 mL). The reaction mixture was stirred at room temperature for 2 h, treated with water (50 mL) dried and concentrated. The residue obtained was dissolved in methanol (40 mL) and 10% NaOH (21.5 mL). After stirring at room temperature for 30 min the solvent was removed. The residue was treated with 10% HCl (30 mL) and dichloromethane (50 mL). The organic phase was separated, dried and concentrated to give **37** (10.83 g, 98%) as an oil: ^1H NMR ($CDCl_3$) δ 2.14 (s, 3H), 3.97 (t, 2H), 4.12 (t, 2H), 6.86–7.02 (m, 3H), 8.2 (br s, 1H), 8.24 (m, 1H). Anal. Calcd for $C_{10}H_{13}NO_3$ (195.22).

5.1.22. 2-(2-Acetylaminophenoxy)-1-mesyloxyethane (38). Prepared from **37** as described for **26** and isolated after crystallization from diisopropyl ether/2-propanol (120 mL:100 mL) as a white solid (53%): mp 87.9 °C ^1H NMR ($CDCl_3$) δ 1.2 (s, 3H), 3.1 (s, 3H), 4.23 (t, 2H), 4.65 (t, 2H), 6.8 (m, 1H), 6.98 (m, 2H), 8.29 (br s, 1H), 8.39 (m, 1H). Anal. Calcd for $C_{11}H_{15}NO_5S$ (273.39).

5.1.23. 2-(2-Acetylaminophenoxy)-1-dibenzylaminoethane (39). A solution of **38** (8.6 g, 32 mmol) and dibenzylamine (12.21 mL, 64 mmol) in isopropanol (50 mL) was refluxed overnight. At the end, the solvent was removed and the residue dissolved in dichloromethane (50 mL) and treated with 10% NaOH (50 mL). The organic phase was separated, washed with water (50 mL), dried and concentrated to give the crude product, which was purified by chromatography on silica gel.

Elution with cyclohexane/ethyl acetate (1:1) afforded **39** as an oil. After crystallization from cyclohexane (50 mL) **39** was isolated (7.63 g, 64%) as a white solid: mp 98.1 °C; ^{1}H NMR ($CDCl_3$) δ 2.09 (s, 3H), 2.94 (t, 2H), 3.73 (s, 4H), 4.1 (t, 2H), 6.77 (m, 1H), 6.96 (m, 2H), 7.23–7.42 (m, 10H), 7.79 (br s, 1H), 8.36 (m, 1H). Anal. Calcd for $C_{24}H_{26}N_2O_2$ (374.48).

5.1.24. 2-(Acetylaminophenoxy)ethylamine (40). A solution of **39** (7.63 g, 20 mmol) in ethyl acetate (100 mL) and ethanol (250 mL) was added with 10% Pd/C (0.5 g) and vigorously shaken under hydrogen at room temperature. The catalyst was removed by filtration, the filtrate concentrated and dissolved with dichloromethane (100 mL). The solvent was then removed to give **40** (3.88 g) as a white solid: mp 62.4 °C: ^{1}H NMR ($CDCl_3$) δ 1.49 (s, 2H), 2.18 (s, 3H), 3.09 (t, 2H), 4.04 (t, 2H), 6.88–7.04 (m, 3H), 8.32 (m, 1H), 8.44 (br s, 1H): $C_{10}H_{14}N_2O_2$ (194.23).

5.1.25. (2*S*)-2-Benzylaminomethyl-1,4-benzodioxane [(*S*)-41]. A solution of (2*R*)-2-iodomethyl-1,4-benzodioxane (1.5 g, 5.43 mmol) and benzylamine (1.18 mL, 10.86 mmol) in isopropanol was refluxed for 18 h. After cooling at room temperature, Na_2CO_3 (700 mg) and silica gel (10 g) was added. The solvent was removed and the residue was purified by chromatography on silica gel. Elution with hexane/ethyl acetate (85:15) afforded 850 mg (81.2%) of (*S*)-**41** as a colourless oil: $[\alpha]_D^{25} = -41.4$ (*c* 1, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 2.90 (d, 2H), 3.85 (s, 2H), 3.95–4.50 (m, 3H), 6.90 (s, 4H), 7.35 (s, 5H). Anal. Calcd for $C_{16}H_{17}NO_2$ (255.31).

5.1.26. (*S*)-2-[((2-(Phenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*S*)-1]. A solution of (2*R*)-2-iodomethyl-1,4-benzodioxane (1.7 g, 6.16 mmol) and 2-phenoxyethylamine (1.69 g, 12.32 mmol) in EtOH (5 mL) was refluxed for 48 h. After removing the solvent, 10% NaOH and diethyl ether were added. The aqueous layer was separated and extracted with diethyl ether again. The organic phases were combined, dried and concentrated to give a residue, which was purified by chromatography on silica gel. Elution with cyclohexane/ethyl acetate–isopropanol–NH_4OH (8:6:2:0.03) afforded 1.37 g (78%) of (2*S*)-2-[((2-(Phenoxy)ethyl)amino)methyl]-1,4-benzodioxane as an oil: $[\alpha]_D^{25} = -33.44$ (*c* 1, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 1.7 (s, 1H), 2.7–3.35 (m, 4H), 3.8–4.6 (m, 5H), 6.7–7.1 (m, 9H). The secondary amine was dissolved in EtOH (10 mL) and 4 N HCl/EtOH (5 mL) was slowly added. The resulting precipitate was dried yielding 0.995 g (64%, based on the starting amount of (2*R*)-2-iodomethyl-1,4-benzodioxane) of (*S*)-**1** as a white solid: mp 233 °C; $[\alpha]_D^{25} = -67.0$ (*c* 1, methanol); ^{1}H NMR (DMSO-d_6) δ 2.9–3.8 (m, 4H), 3.8–5.2 (m, 5H), 6.5–7.7 (m, 9H), 9.82 (br s, 2H). Anal. Calcd for $C_{17}H_{20}ClNO_3$ (285.34).

5.1.27. (*R*)-2-[((2-(Phenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*R*)-1]. Prepared from (2*S*)-2-iodomethyl-1,4-benzodioxane and 2-phenoxyethylamine as described (*S*)-**1**: mp 230 °C; $[\alpha]_D^{25} = +65.74$ (*c* 1, methanol) ($[\alpha]_D^{25} = +35.22$ (*c* 1, $CHCl_3$) for the free amine); ^{1}H NMR identical to that of (*S*)-**1**.

5.1.28. (*S*)-2-[((2-(2-Methylphenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*S*)-2]. A solution of 2-(2-methylphenoxy)ethylamine (1.4 g, 9.25 mmol) and (2*R*)-mesyloxymethyl-1,4-benzodioxane (1.2 g, 4.91 mmol) in 2-methyl-1-propanol (4 mL) was refluxed for 48 h. After removing the solvent, 10% Na_2CO_3 and dichloromethane were added. The aqueous layer was separated and extracted with dichloromethane again. The organic phases were combined, dried and concentrated to give a residue, which was purified by chromatography on silica gel. Elution with cyclohexane/ethyl acetate (1:1) afforded 0.44 g (30.13%) of (2*S*)-2-[((2-(2-methylphenoxy)ethyl)amino)methyl]-1,4-benzodioxane: $[\alpha]_D^{25} = -32.2$ (*c* 1, $CHCl_3$);^{1}H NMR ($CDCl_3$) δ 1.40 (br s, 1H), 2.15 (s, 3H), 3.00 (m, 2H), 3.15 (m, 2H), 4.00–4.10 (m, 3H), 4.10–4.25 (m, 2H), 6.80 (m, 6H), 7.15 (m, 2H). The secondary amine was dissolved in ethyl acetate/chloroform (5:1) (5 mL) and 4 N HCl/EtOH was added. Ethyl acetate (10 mL) was added and evaporated for three times. After crystallization with ethyl acetate/ethanol (10:1), (*S*)-**2** was isolated as a white solid: mp 167.7 °C; $[\alpha]_D^{25} = -61.34$ (*c* 1, methanol); ^{1}H NMR (DMSO-d_6) δ 2.18 (s, 3H), 3.27–3.46 (m, 4H), 4.08 (dd, 1H), 4.31 (t, 2H), 4.40 (dd, 1H), 4.72 (m, 1H), 6.80–6.95 (m, 6H), 7.14 (m, 2H), 9.74 (br s, 2H). Anal. Calcd for $C_{18}H_{22}ClNO_3$ (335.82).

5.1.29. (*R*)-2-[((2-(2-Methylphenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*R*)-2]. Prepared from 2-(2-methylphenoxy)ethylamine and (2*S*)-mesyloxymethyl-1,4-benzodioxane as described for (*S*)-**2**: mp 167.3 °C; $[\alpha]_D^{25} = +60.1$ (*c* 1, ethanol) ($[\alpha]_D^{25} = +32.9$ (*c* 1, $CHCl_3$) for the free amine); ^{1}H NMR identical to that of (*S*)-**2**.

5.1.30. (*S*)-2-[((2-(2-Ethylphenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*S*)-3]. Prepared from (2*R*)-2-iodomethyl-1,4-benzodioxane (1.5 g, 5.43 mmol) and **31** (1.8 g, 10.86 mmol) as described for (*S*)-**1**. (2*S*)-2-[((2-(2-Ethylphenoxy)ethyl)amino)methyl]-1,4-benzodioxane was isolated as an oil: $[\alpha]_D^{25} = -25.3$ (*c* 1, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 1.2 (t, 3H), 1.9 (s, 1H), 2.68 (q, 2H), 2.8–3.25 (m, 4H), 3.65–4.55 (m, 5H), 6.4–7.4 (m, 8H). (*S*)-**3** was isolated as a white solid (17% based on the starting amount of (2*R*)-2-iodomethyl-1,4-benzodioxane): mp 141 °C; $[\alpha]_D^{25} = -55.22$ (*c* 1, methanol) ^{1}H NMR (DMSO-d_6) δ 0.92 (t, 3H), 2.47 (q, 2H), 2.95–3.53 (m, 4H), 3.6–4.3 (m, 5H), 4.62 (s, 2H), 6.5–7.2 (m, 8H). Anal. Calcd for $C_{19}H_{24}ClNO_3$ (385.86).

5.1.31. (*R*)-2-[((2-(2-Ethylphenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*R*)-3]. Prepared from (2*S*)-2-iodomethyl-1,4-benzodioxane and **31** as described for (*S*)-**3**: mp 141–142 °C; $[\alpha]_D^{25} = +55.19$ (*c* 1, methanol) ($[\alpha]_D^{25} = +25.25$ (*c* 1, $CHCl_3$) for the free amine); ^{1}H NMR identical to that of (*S*)-**3**.

5.1.32. (*S*)-2-[((2-(2-*t*-Butoxyphenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*S*)-4]. Prepared from (2*R*)-mesyloxymethyl-1,4-benzodioxane (1 g, 8.17 mmol) and 2-(2-*t*-butoxyphenoxy)ethylamine (1.58 g, 8.17 mmol) as described for (*S*)-**2**. The free amine was isolated as an oil: $[\alpha]_D^{25} = -24.4$ (*c* 1, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 1.40 (s, 9H), 1.95 (br s, 1H), 3.00 (m, 2H), 3.15 (m, 2H), 3.95–4.19 (m, 3H), 4.20–4.40 (m, 2H), 6.75–6.95 (m, 6H), 7.10–7.35 (m, 2H). (*S*)-**4** was isolated as a white solid (37% based on the starting amount of (2*R*)-mesyloxymethyl-1,4-benzodioxane): mp 156.8 °C; $[\alpha]_D^{25} = -63.7$ (*c* 1, methanol) ^1H NMR (DMSO-d_6) δ 1.32 (s, 9H), 3.30–3.50 (m, 4H), 4.08 (dd, 1H), 4.34–4.40 (m, 3H), 4.73 (m, 1H), 6.88 (m, 5H), 7.00 (d, 1H), 7.19 (m, 2H), 9.80 (br s, 2H). Anal. Calcd for $C_{21}H_{28}ClNO_3$ (377.90).

5.1.33. (*R*)-2-[((2-(2-*t*-Butoxyphenoxy)ethyl)amino)-methyl]-1,4-benzodioxane hydrochloride [(*R*)-4]. Prepared from (2*S*)-mesyloxymethyl-1,4-benzodioxane and 2-(2-*t*-butoxyphenoxy)ethylamine as described for (*S*)-**4**: mp 156.8 °C; $[\alpha]_D^{25} = +55.71$ (*c* 1, methanol) ($[\alpha]_D^{25} = +23.45$ (*c* 1, $CHCl_3$) for the free amine); ^1H NMR identical to that of (*S*)-**4**.

5.1.34. (*S*)-2-[((2-(2-Cyanophenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*S*)-5]. Prepared from (2*R*)-2-iodomethyl-1,4-benzodioxane (1.47 g, 5.32 mmol) and 2-(2-cyanophenoxy)ethylamine (1.08 g, 6.65 mmol) as described for (*S*)-**1**. After chromatography on silica gel (dichloromethane/methanol 97:3) the secondary amine was isolated as an oil: $[\alpha]_D^{25} = -26.4$ (*c* 1, $CHCl_3$). (*S*)-**5** was obtained as a white solid: mp 163–166 °C; $[\alpha]_D^{25} = -60.29$ (*c* 1, methanol) ^1H NMR ($CDCl_3$) δ 2.10 (br s, 1H), 2.90–3.35 (m, 4H), 3.90–4.60 (m, 5H), 6.80–7.20 (m, 6H), 7.35–7.80 (m, 2H). Anal. Calcd for $C_{18}H_{19}N_2ClO_3$ (346.81).

5.1.35. (*R*)-2-[((2-(2-Cyanophenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*R*)-5]. Prepared from (2*S*)-2-iodomethyl-1,4-benzodioxane and 2-(2-cyanophenoxy)ethylamine as described for (*S*)-**5**: mp 165.7 °C; $[\alpha]_D^{25} = +59.2$ (*c* 1, methanol) ($[\alpha]_D^{25} = +25.05$ (*c* 1, $CHCl_3$) for the free amine); ^1H NMR identical to that of (*S*)-**5**.

5.1.36. (*S*)-2-[((2-(2-Trifluoromethylphenoxy)ethyl)amino)-methyl]-1,4-benzodioxane hydrochloride [(*S*)-6]. Prepared from (2*R*)-2-iodomethyl-1,4-benzodioxane (1.02 g, 37 mmol) and **32** (0.95 g, 4.63 mmol) as described for (*S*)-**1**. After chromatography on silica gel (dichloromethane/methanol 98:2) the free amine was isolated as an oil: $[\alpha]_D^{25} = -28.3$ (*c* 1, $CHCl_3$). (*S*)-**6** was isolated as a white solid (370 mg, 26% based on the starting amount of (2*R*)-2-iodomethyl-1,4-benzodioxane: mp 150–152 °C; $[\alpha]_D^{25} = -48.1$ (*c* 1, methanol) ^1H NMR (DMSO-d_6) δ 3.60–3.33 (m, 4H), 4.10 (dd, 1H), 4.43 (d, 1H), 4.56 (m, 2H), 4.75 (m, 1H), 6.93 (m, 4H), 7.20 (t, 1H), 7.37 (d, 1H), 7.70 (m, 2H), 9.90 (m, 2H). Anal. Calcd for $C_{18}H_{19}ClF_3NO_3$ (389.79).

5.1.37. (*R*)-2-[((2-(2-Trifluoromethylphenoxy)ethyl)-amino)methyl]-1,4-benzodioxane hydrochloride [(*R*)-6]. Prepared from (2*S*)-2-iodomethyl-1,4-benzodioxane and **32** as described for (*S*)-**6**: mp 150.7 °C; $[\alpha]_D^{25} = +48.08$ (*c* 1, methanol) ($[\alpha]_D^{25} = +27.19$ (*c* 1, $CHCl_3$) for the free amine); ^1H NMR identical to that of (*S*)-**6**.

5.1.38. (*S*)-2-[((2-(2-Acetylphenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*S*)-7]. A solution of (*S*)-**41** (800 mg, 3.13 mmol), **25** (910 mg, 3.13 mmol) and K_2CO_3 (500 mg) in toluene (15 mL) was refluxed for 48 h. The solvent was removed to give the residue, which was purified by chromatography on silica gel. Elution with hexane/ethyl acetate (90:10) afforded 560 mg of tertiary amine, which was dissolved in a suspension of 10% Pd/C in ethanol. At the end the catalyst was filtrate and the solvent removed. The residue was dissolved in ethyl acetate (30 mL) and treated with 5% NaOH (30 mL). The aqueous layer was separated and extracted with ethyl acetate again. The organic phases were combined, washed with water, dried and concentrated to give the residue, which was purified by chromatography on silica gel. Elution with dichloromethane/methanol (97:3) afforded (*S*)-2-[((2-(2-acetylphenoxy)ethyl)amino)methyl]-1,4-benzodioxane (340 mg, 43%) as a yellow oil. The secondary amine was dissolved in ethanol (5 mL) and 4 N HCl/EtOH (5 mL) was added. The resulting precipitate was filtrated and dried to give (*S*)-**7** (320 mg, 26% based on the starting amount of (*S*)-**41**): mp 118 °C; $[\alpha]_D^{25} = -53.23$ (*c* 1, methanol); ^1H NMR ($CDCl_3$) δ 2.58 (s, 3H), 3.30 (m, 2H), 3.40–3.52 (m, 2H), 4.16 (dd, 1H), 4.31–4.40 (m, 1H), 4.50–4.54 (m, 1H), 6.84–6.90 (m, 5H), 7.10 (t, 1H), 7.45 (t, 1H), 7.70 (dd, 1H), 9.7 (br s, 1H), 10.7 (br s, 1H). Anal. Calcd for $C_{19}H_{23}N_2O_4Cl$ (378.84).

5.1.39. (*R*)-2-[((2-(2-Acetylphenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*R*)-7]. Prepared from (*R*)-**41** and **25** as described for (*S*)-**7**: mp 97.8 °C; $[\alpha]_D^{25} = +60.12$ (*c* 1, methanol); ^1H NMR ($CDCl_3$) identical to that of (*S*)-**7**.

5.1.40. (*S*)-2-[((2-(2-Fluorophenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*S*)-8]. Prepared from (2*R*)-2-iodomethyl-1,4-benzodioxane (1 g, 3.62 mmol) and 2-(2-fluorophenoxy)ethylamine (1.2 g, 7.24 mmol) as described for (*S*)-**1** but replacing ethanol with isopropanol. The secondary amine was isolated as an oil: $[\alpha]_D^{25} = -31.72$ (*c* 1, $CHCl_3$). (*S*)-**8** was obtained as a white solid (0.51 g, 42% based on the starting amount of (2*R*)-2-iodomethyl-1,4-benzodioxane): mp 148–154 °C; $[\alpha]_D^{25} = -62.79$ (*c* 1, $CHCl_3$); ^1H NMR (DMSO-d_6) δ 3.26–3.31 (m, 1H), 3.41–3.46 (m, 3H), 4.03 (dd, 1H), 4.35–4.41 (m, 3H), 4.65–4.69 (m, 1H), 6.83–6.9 (m, 4H), 6.92–7.0 (m, 1H), 7.14–7.27 (m, 3H), 9.54 (br s, 2H): Anal. Calcd for $C_{17}H_{19}O_3FNCl$ (339.78).

5.1.41. (*R*)-2-[((2-(2-Fluorophenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*R*)-8]. Prepared from (2*S*)-2-iodomethyl-1,4-benzodioxane and 2-(2-fluorophenoxy)ethylamine as described for (*S*)-**8**: mp 213.4 °C; $[\alpha]_D^{25} = +58.06$ (*c* 1, $CHCl_3$) ($[\alpha]_D^{25} = +31.22$ (*c* 1, $CHCl_3$) for the free amine); ^1H NMR identical to that of (*S*)-**8**.

5.1.42. (*S*)-2-[((2-(2-Chlorophenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*S*)-9]. Prepared from

(2*R*)-2-iodomethyl-1,4-benzodioxane (1 g, 3.62 mmol) and 2-(2-chlorophenoxy)ethylamine (2.48 g, 14.48 mmol) as described for (*S*)-**1** but replacing ethanol with 2-methylpropanol. After purification by chromatography on silica gel (dichloromethane/methanol 98:2) the secondary amine was isolated as a yellow oil (880 mg, 76.5%): $[\alpha]_D^{25} = -26.56$ (*c* 1, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 1.95 (br s, 1H), 2.95 (m, 2H), 3.12 (m, 2H), 4.05 (dd, 1H), 4.13–4.20 (m, 2H), 4.25–4.40 (m, 2H), 6.83–6.95 (m, 6H), 7.21 (t, 1H), 7.36 (d, 1H). (*S*)-**9** was isolated as a white solid (460 mg, 38.3% based on the starting amount of (2*R*)-2-iodomethyl-1,4-benzodioxane): mp 164.6 °C; $[\alpha]_D^{25} = -53.33$ (*c* 1, methanol); ^{1}H NMR (DMSO-d_6) δ 3.42–3.57 (m, 4H), 4.13 (dd, 1H), 4.42–4.48 (m, 3H), 4.77 (m, 1H), 6.88–7.09 (m, 5H), 7.25 (m, 1H), 7.38 (t, 1H), 7.49 (d, 1H), 9.78 (br s, 2H). Anal. Calcd for $C_{17}H_{19}Cl_2NO_3$ (356.23).

5.1.43. (*R*)-2-[((2-(2-Chlorophenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*R*)-9]. Prepared from (2*S*)-2-iodomethyl-1,4-benzodioxane and 2-(2-chlorophenoxy)ethylamine as described for (*S*)-**9**: mp 164.6 °C; $[\alpha]_D^{25} = +53.28$ (*c* 1, methanol) ($[\alpha]_D^{25} = +29.22$ (*c* 1, CHCl$_3$) for the free amine); ^{1}H NMR identical to that of (*S*)-**9**.

5.1.44. (*S*)-2-[((2-(2-Methylthiophenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*S*)-10]. Prepared from (2*R*)-2-iodomethyl-1,4-benzodioxane (1.11 g, 4.02 mmol) and **33** (1.47 g, 8.05 mmol) as described for (*S*)-**1**. The secondary amine was isolated as an oil (442 mg, 34%): $[\alpha]_D^{25} = -23.79$ (*c* 1, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 2.04 (s, 1H), 2.4 (s, 3H), 2.8–3.3 (m, 4H), 3.8–4.55 (m, 5H), 6.6–7.5 (m, 8H). (*S*)-**10** was isolated as a white solid (387 mg, 29% based on the starting amount of (2*R*)-2-iodomethyl-1,4-benzodioxane): mp 167-169 °C; $[\alpha]_D^{25} = -50.47$ (*c* 1, methanol); ^{1}H NMR (DMSO-d_6) δ 2.4 (s, 3H), 3.1–3.8 (m, 5H), 6.7–7.45 (m, 8H), 9.85 (m, 1H). Anal. Calcd for $C_{18}H_{22}ClNO_3S$ (367.88).

5.1.45. (*S*)-2-[((2-(2-Methylthiophenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*R*)-10]. Prepared from (2*S*)-2-iodomethyl-1,4-benzodioxane and **33** as described for (*S*)-**10**: mp 167–170 °C; $[\alpha]_D^{25} = +49.3$ (*c* 1, methanol) ($[\alpha]_D^{25} = +23.7$ (*c* 1, CHCl$_3$) for the free amine); ^{1}H NMR identical to that of (*S*)-**10**.

5.1.46. (*S*)-2-[((2-(2-Methanesulfinylphenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*S*)-11]. Prepared from (2*R*)-2-iodomethyl-1,4-benzodioxane (1.04 g, 3.76 mmol) and **34** (1.5 g, 7.52 mmol) as described for (*S*)-**1** but replacing ethanol with isopropanol (5 mL). The secondary amine was isolated as an oil (1.2 g, 92%): $[\alpha]_D^{25} = -27.46$ (*c* 1, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 2.20 (s, 1H), 2.85 (s, 3H), 2.9–3.4 (m, 4H), 4.0–4.6 (m, 5H), 6.9–8.0 (m, 8H). (*S*)-**11** was isolated as a white solid (510 mg, 35% based on the starting amount of (2*R*)-2-iodomethyl-1,4-benzodioxane): mp 134-136 °C; $[\alpha]_D^{25} = -48.06$ (*c* 1, methanol); ^{1}H NMR (DMSO-d_6) δ 2.73 (s, 3H), 3.26–3.33 (m, 2H), 3.40–3.46 (m, 3H), 4.0–4.1 (m, 1H), 4.36–4.40 (m, 3H), 4.41(m, 1H), 6.83–6.91 (m, 4H), 7.17–7.16 (m, 2H), 7.50–7.55 (m, 1H), 7.64–7.67 (m, 1H). Anal. Calcd for $C_{18}H_{22}O_4NCl$ (383.87).

5.1.47. (*R*)-2-[((2-(2-Methanesulfinylphenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*R*)-11]. Prepared from (2*S*)-2-iodomethyl-1,4-benzodioxane and **34** as described for (*S*)-**11**: mp 136 °C; $[\alpha]_D^{25} = +47.22$ (*c* 1, methanol) ($[\alpha]_D^{25} = +28.6$ (*c* 1, CHCl$_3$) for the free amine); ^{1}H NMR identical to that of (*S*)-**11**.

5.1.48. (*S*)-2-[((2-(2-Methanesulfonylphenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*S*)-12]. Prepared from (2*R*)-2-iodomethyl-1,4-benzodioxane (520 mg, 1.88 mmol) and **35** (750 mg, 3.76 mmol) as described for (*S*)-**1** but replacing ethanol with isopropanol. The secondary amine was isolated (460 mg, 67%) as an oil: $[\alpha]_D^{25} = -15.91$ (*c* 1, CHCl$_3$); ^{1}H NMR (CDCl$_3$) δ 2.15 (s, 1H), 2.90–3.40 (m, 4H), 3.25 (s, 3H), 4.00–4.60 (m, 5H), 6.80–8.10 (m, 4H). (*S*)-**12** was isolated as a white solid (250 mg, 39% based on the starting amount of (2*R*)-2-iodomethyl-1,4-benzodioxane): mp 94–97 °C; $[\alpha]_D^{25} = -44.09$ (*c* 1, methanol); ^{1}H NMR (DMSO-d_6) δ 3.29–3.31 (m, 6H), 3.40–3.53 (m, 2H), 4.0–4.06 (m, 1H), 4.33–4.37 (m, 1H), 4.53–4.63 (m, 2H), 6.8–7.00 (m, 4H), 7.19–7.24 (m, 1H), 7.37–7.36 (m, 1H), 7.72–7.80 (m, 1H), 7.80–7.81 (m, 1H). Anal. Calcd for $C_{18}H_{22}ClNO_5S$ (399.47).

5.1.49. (*R*)-2-[((2-(2-Methanesulfonylphenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*R*)-12]. Prepared from (2*S*)-2-iodomethyl-1,4-benzodioxane and **35** as described for (*S*)-**12**: mp 93–96 °C; $[\alpha]_D^{25} = +45.84$ (*c* 1, methanol) ($[\alpha]_D^{25} = +17.43$ (*c* 1, CHCl$_3$) for the free amine); ^{1}H NMR identical to that of (*S*)-**12**.

5.1.50. (*S*)-2-[((2-(2-Nitrophenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*S*)-13]. A solution of (2*S*)-aminomethyl-1,4-benzodioxane (1.5 g, 9.08 mmol) and **27** (1.4 g, 5.45 mmol) in 2-methylpropanol was refluxed for 24 h. At the end, the solvent was removed and the residue dissolved in dichloromethane (20 mL) and treated with NaHCO$_3$ (20 mL). The aqueous layer was separated and extracted with dichloromethane again. The organic phases were combined, washed with water, dried and concentrated to give a residue, which was purified by chromatography on silica gel. Elution with dichloromethane/methanol (98:2) afforded 1.06 g (59%) of (*S*)-2-[((2-(2-nitrophenoxy)ethyl)amino)methyl]-1,4-benzodioxane as a white solid: mp 75.6 °C; $[\alpha]_D^{25} = -51.3$ (*c* 1, methanol); ^{1}H NMR (CDCl$_3$) δ 1.95 (br s, 1H), 2.9–3.04 (m, 2H), 3.11 (t, 2H), 3.99–4.09 (dd, 1H), 4.2–4.33 (m, 4H), 6.8–6.99 (m, 4H), 7.03–7.09 (m, 2H), 7.52 (dt, 1H), 7.86 (dd, 1H). The secondary amine was dissolved in ethanol (2 mL) and 4.6 N HCl/EtOH (2 mL) was slowly added. The resulting precipitate was dried to give (S)-**13** (700 mg, 36% based on the starting amount of **27**) as a white solid: mp 155.1 °C; $[\alpha]_D^{25} = -55.74$ (*c* 1, methanol); ^{1}H NMR (DMSO-d_6) δ 3.38–3.52 (m, 4H), 4.06–4.15 (dd, 1H), 4.45 (d, 1H), 4.60 (m, 2H), 4.73 (m, 1H), 6.93 (m, 4H), 7.23 (t, 1H), 7.09 (d, 1H), 7.45 (t, 1H), 7.96 (d, 1H), 9.81 (br s, 2H). Anal. Calcd for $C_{17}H_{19}ClN_2O_5 \cdot 1/2H_2O$ (365.26).

5.1.51. (*R*)-2-[((2-(2-Nitrophenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*R*)-13]. Prepared from (2*R*)-aminomethyl-1,4-benzodioxane and **27** as described for (S)-**13**: mp 154.8 °C; $[\alpha]_{\mathrm{D}}^{25} = +57.2$ (*c* 1, methanol) (mp 75.7 °C; $[\alpha]_{\mathrm{D}}^{25} = +53.22$ (*c* 1, methanol) for the free amine); ^{1}H NMR identical to that of (S)-**13**.

5.1.52. (*S*)-2-[((2-(2-Acetylaminophenoxy)ethyl)amino)-methyl]-1,4-benzodioxane hydrochloride [(*S*)-14]. Prepared from (2*R*)-mesyloxymethyl-1,4-benzodioxane (1.25 g, 5.15 mmol) and **40** (2 g, 10.2 mmol) as described for (*S*)-**2**. After purification by chromatography on silica gel (dichloromethane/MeOH–TEA 97.5/2.5/0.5) (*S*)-2-[((2-(2-acetylaminophenoxy)ethyl)amino)methyl]-1,4-benzodioxane was isolated (1.14 g, 65%) as a yellow oil: $[\alpha]_{\mathrm{D}}^{25} = -43.9$ (*c* 1, methanol); ^{1}H NMR (CDCl$_3$) δ 2.13 (br s, 1H), 2.18 (s, 3H), 2.97 (t, 2H), 3.08 (t, 2H), 4.01 (dd, 1H), 4.13 (t, 2H), 4.27 (dd, 1H), 4.33 (m, 1H), 6.74–7.02 (m, 7H), 8.2 (s, 1H), 8.33 (dd, 1H). (*S*)-**14** was isolated as a white solid (650 mg, 33% based on the starting amount of (2*R*)-mesyloxymethyl-1,4-benzodioxane): mp 122.7 °C; $[\alpha]_{\mathrm{D}}^{25} = -56.84$ (*c* 1, water); ^{1}H NMR (DMSO-*d*$_6$) δ 2.24 (s, 3H), 3.41–3.53 (m, 4H), 4.13 (dd, 1H), 4.35 (t, 2H), 4.41 (dd, 1H), 4.9 (m, 1H), 6.94–6.99 (m, 5H), 7.07 (m, 2H), 8.05 (d, 1H), 9.67 (s, 1H), 9.8 (br s, 1H). Anal. Calcd for $C_{19}H_{23}N_2O_4Cl$ (378.86).

5.1.53. (*R*)-2-[((2-(2-Acetylaminophenoxy)ethyl)amino)-methyl]-1,4-benzodioxane hydrochloride [(*R*)-14]. Prepared from (2*S*)-mesyloxymethyl-1,4-benzodioxane and **40** as described for (*S*)-**14**: mp 117.1 °C; $[\alpha]_{\mathrm{D}}^{25} = +48.76$ (*c* 1, water) ($[\alpha]_{\mathrm{D}}^{25} = +41.61$ (*c* 1, water) for the free amine); ^{1}H NMR identical to that of (*S*)-**14**.

5.1.54. (*S*)-2-[((2-(2-Aminophenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*S*)-15]. A solution of (*S*)-**14** (0.85 g, 2.48 mmol) in 6 N HCl (5 mL) was heated for 1 h. At the end water (10 mL) was added and then concentrated to give the crude product, which was crystallized from ethanol (16 mL) affording (*S*)-**15** as a white solid: mp 217 °C; $[\alpha]_{\mathrm{D}}^{25} = -56.65$ (*c* 1, methanol); ^{1}H NMR (DMSO-*d*$_6$) δ 3.30–3.50 (m, 4H), 4.13 (dd, 1H), 4.46–4.51 (m, 3H), 4.92 (m, 1H), 6.93–6.98 (m, 4H), 7.10 (t, 1H), 7.25 (d, 1H), 7.43 (m, 2H), 10.0 (m, 5H). Anal. Calcd for $C_{17}H_{22}N_2O_3Cl_2$ (373.28).

5.1.55. (*R*)-2-[((2-(2-Aminophenoxy)ethyl)amino)methyl]-1,4-benzodioxane hydrochloride [(*R*)-15]. Prepared from (*R*)-**14** as described for (*S*)-**15**: mp 217 °C; $[\alpha]_{\mathrm{D}}^{25} = +51.63$ (*c* 1, methanol); ^{1}H NMR identical to that of (*S*)-**15**.

5.2. Biology

5.2.1. Binding assays. The pharmacological profile of both the *S* and *R* enantiomers of compounds **1**–**16** was assessed by measuring their affinities for α_{1a}, α_{1b}, α_{1d} AR-subtypes and 5-HT$_{1A}$ serotoninergic receptor with in vitro binding studies.

Briefly, membranes derived from Chinese Hamster Ovary (CHO) cells expressing α_1-AR subtypes (prepared as described by Testa et al.[31]) were resuspended in Tris HCl, 50 mM, pH = 7.7 containing 10 μM pargyline and 0.1% ascorbic acid and incubated for 30 min at 25 °C with 0.5 nM [^{3}H]-Prazosin (NEN, 80.5 Ci/mmol) in the absence or presence of different concentrations of the tested compounds. Prazosin 1 M was used to determine non-specific binding.

Binding studies at 5-HT$_{1A}$ receptors were carried out using crude membrane preparations from rat hippocampus, which were resuspended in Tris HCl 50 mM (pH = 7.7, 10 μM pargyline and 4 mM CaCl$_2$) and incubated for 30 min at 25 °C with 1 nM [^{3}H]-8-OH-DPAT, in the absence or presence of different concentrations of the tested compounds. 5-HT$_{1A}$ 1 μM was used to determine non-specific binding.

Incubations were stopped by rapid filtration, through GF/B fiber filters, which were then washed, dried and counted in a LK1214 rack β-liquid scintillation spectrometer.

At least three different experiments, in triplicate, were carried out for each compound and usually each compound was tested simultaneously on the different α_1-AR subtypes. Prazosin or 5-HT$_{1A}$ were always tested in parallel, as reference drugs. The percentage inhibitory effects obtained in the different experiments were pooled together and the inhibition curves were analyzed using the 'one-site competition' equation built into GraphPad Prism 4.0 (GraphPAD Software, San Diego, CA). This analysis gives the IC$_{50}$ (i.e., the drug concentration inhibiting specific binding by 50%), calculated with the relative standard error. K_i values were then calculated by IC$_{50}$ using the Cheng and Prusoff equation in which the K_d of [^{3}H]-Prazosin for α_{1a}, α_{1b}, α_{1d} AR-subtypes were 0.4, 0.4 and 0.7 nM, respectively, whereas the K_d of [^{3}H]-8-OH-DPAT for 5-HT$_{1A}$ receptors was 1.2 nM.

Acknowledgements

Financial support provided by the Italian Ministero dell'Istruzione, dell'Università e della Ricerca Scientifica is gratefully acknowledged.

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