Nitric Oxide Donor β_2-Agonists: Furoxan Derivatives Containing the Fenoterol Moiety and Related Furazans

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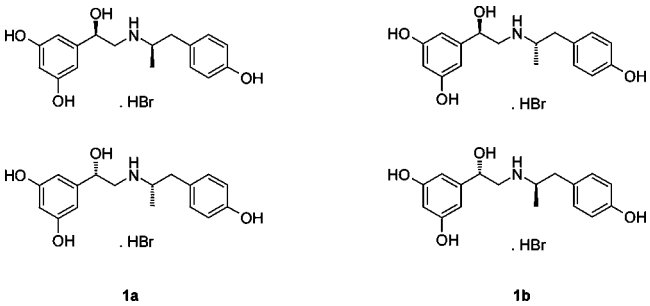
The structure of fenoterol, a β_2-adrenoceptor agonist used in therapy, has been joined with furoxan NO-donor moieties to give new NO-donor β_2-agonists. The furazan analogues, devoid of the property to release NO, were also synthesized for comparison. All the compounds retained β_2-agonistic activity at micromolar or submicromolar concentration when tested on guinea pig tracheal rings precontracted with carbachol. Among the furoxan derivatives, the NO contribution to trachea relaxation was evident with product **15b** at micromolar concentrations. All the new NO-donor hybrids were able to dilate rat aortic strips precontracted with phenylephrine. Both furoxan and furazan derivatives displayed antioxidant activity greater than that of fenoterol.

Introduction

Bronchial constriction is characteristic of asthmatic airway obstruction and of chronic obstructive pulmonary diseases (COPD). Selective β_2-adrenoceptor agonists are certainly among the most effective and safest drugs to treat these pathologies. By activating the β_2-receptors positioned on the smooth muscle bronchial cells, they induce an increase in cAMP with consequent muscle relaxation. A wide number of these drugs, characterized by having different pharmacokinetic properties, have been developed in the past.[1] These products also display in vitro antioxidant properties with respect to reactive oxygen species-mediated cytotoxicity.[2] Nitric oxide (NO) is a physiological messenger that triggers a variety of actions in several systems;[3] they include relaxation of vascular smooth muscle, inhibitory effects on smooth muscle proliferation, inhibition of platelets aggregation and adhesion, and bronchodilation. NO triggers all these actions through a mechanism which involves the activation of the soluble guanylate cyclase (sGC). NO plays relevant roles in the respiratory system, and its absence or dysfunction is implicated in a number of airway diseases.[4,5] The therapeutic potential of NO donors in the treatment of respiratory diseases has recently been discussed.[5−8] During the past few years, a link between β-adrenergic receptors and NO generation in the cardiovascular system has been evidenced.[9] In particular, the role of β_2-adrenoceptors under this aspect has been discussed.[10] There is evidence that β_2-mediated vasodilation is partly or completely NO-mediated in resistance vasculature in vivo as well as the effects of β_2-agonists on arterial pulse wave reflection. Fenoterol is a short-acting β_2-selective agonist characterized by a prompt onset action, currently used in therapy.[11] It has two chiral centers and exists in two diastereoisomers, **1a** and **1b**, each of them is a 50/50 mixture of two enantiomers (Chart 1). The *RR,SS* diastereoisomer **1a** is the marketed product. As development of our research on multifunctional drugs containing NO-donor moieties, we designed the new NO-donor β_2-agonists **15a**, **34a**, **36a**, **15b**, **34b**, and **36b**, deriving from the conjugation of diastereoisomers **1a** and **1b**, respectively, to NO-donor furoxan substructures, endowed with different capabilities of producing NO. This paper describes the synthesis, structural characterization, dilator, and antioxidant properties of these products. The furazan analogues **16a**, **35a**, **37a**, **16b**, **35b**, and **37b**, devoid of the property to release NO, are also considered for comparison.

Chart 1. Structures of (*RR,SS*)-Fenoterol (**1a**) and (*RS,SR*)-Fenoterol (**1b**)



Results and Discussion

Chemistry. The synthesis of the final drugs required preliminary preparation of the oxirane derivative **6** and of the amino derivative **10** (Scheme 1). The reaction of 3,5-diacetoxyacetophenone **2** with bromine in $CHCl_3$ solution, followed by heating with HBr in ethanol solution, gave the crude bromo derivative **3**. The protection of the hydroxyl groups of **3** was accomplished with *tert*-butyldimethylchlorosilane (TBDMSCl) in DMF in the presence of diisopropylethylamine (DIPEA). Chloride **4** was the principal product of this reaction, not the expected bromide, because a halogen exchange reaction occurs with the excess of TBDMSCl used. Chloride **4** was treated with $NaBH_4$ to give alcohol **5**, which was transformed into **6** by action of NaH in THF. Amine **10** was obtained by starting with *p*-methoxybenzyl methyl ketone **7**. This ketone was heated in ethanol with NH_2OH to obtain the corresponding oxime **8** that, in turn, was reduced to amine **9** in NH_3-saturated methanol solution, using H_2 in the presence of Ni W-6[12] as the catalyst. The hydrochloride of amine **10** was obtained by modified literature method,[13] hydrolyzing the methoxy group of **9** with 37% HCl at 140 °C in a closed vessel.

Oxirane derivative **6** was treated with the amine hydrochloride **10** in methanol solution in the presence of triethylamine to give the 1:1 mixture of the two diastereoisomers **11** (Scheme 2). Reaction of this mixture with *tert*-butyldicarbonate ($(Boc)_2O$)

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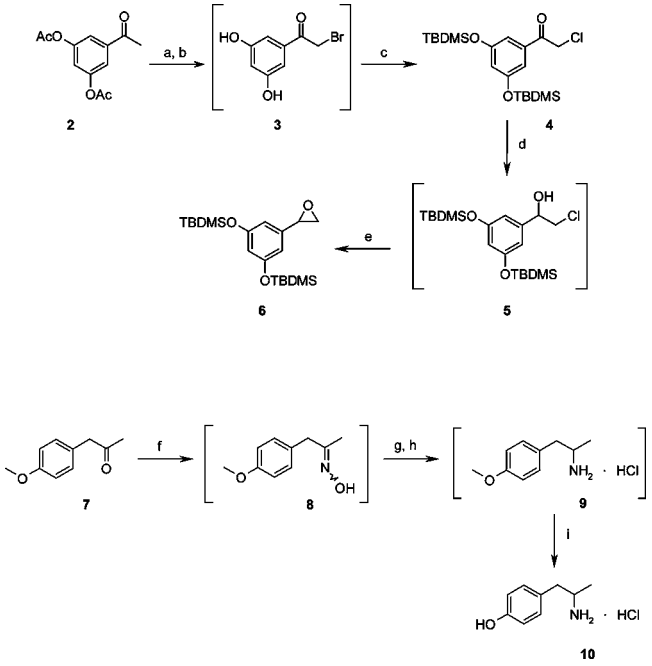
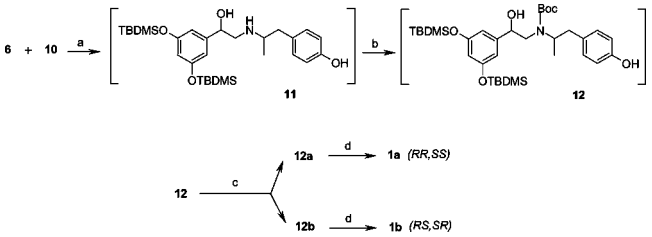
Scheme 1. Synthesis of Intermediates **6** and **10**[a]



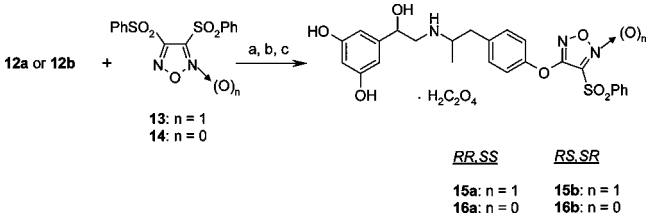
[a] Reagents and conditions: (a) Br_2, $CHCl_3$; (b) 5 N HBr, EtOH, reflux; (c) TBDMSCl, DIPEA, DMF; (d) $NaBH_4$, MeOH; (e) NaH, dry THF, N_2; (f) $NH_2OH{\cdot}HCl$, 10% aq NaOH, EtOH, reflux; (g) H_2, 10% Ni W-6, $MeOH_{(NH_3)}$, 30 bar, 68 °C; (h) $Et_2O_{(HCl)}$; (i) 37% aq HCl, 140 °C, closed vessel.

Scheme 2. Synthesis of Fenoterol Diastereoisomers **1a** and **1b**[a]



[a] Reagents and conditions: (a) Et_3N, MeOH, reflux; (b) $(Boc)_2O$, CH_2Cl_2, rt; (c) HPLC separation; (d) 1 N HBr, MeOH.

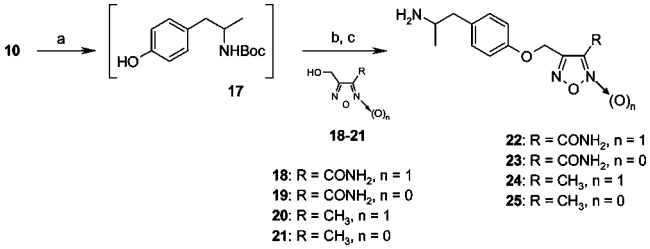
Scheme 3. Synthesis of Final Compounds **15a**, **15b**, **16a**, and **16b**[a]



[a] Reagents and conditions: (a) 50% aq NaOH, dry THF, N_2; (b) $MeOH_{(HCl)}$; (c) 10% aq $NaHCO_3$, $H_2C_2O_4$/EtOAc.

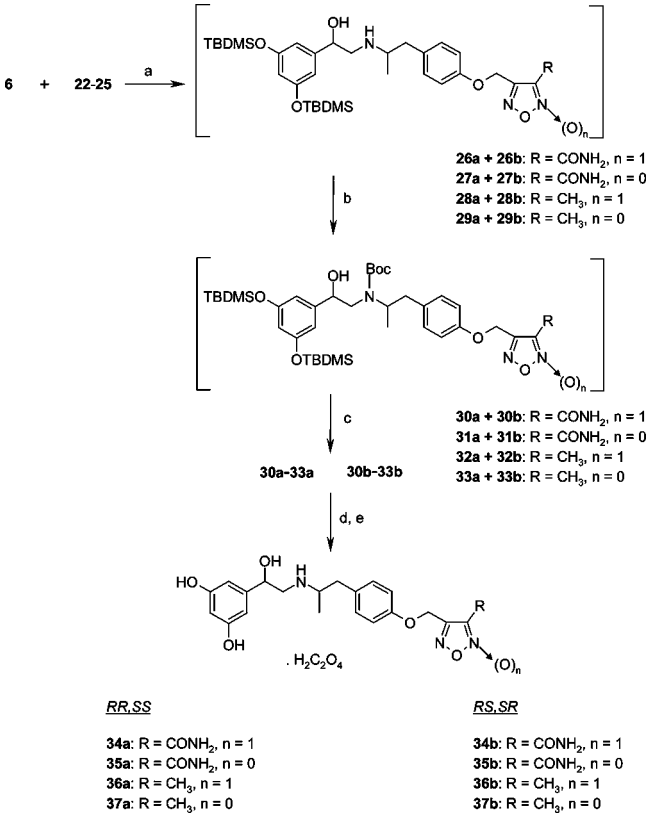
gave mixture **12**, which was subsequently separated by HPLC to give **12a** (first eluted) and **12b**. Treatment with 1 N HBr gave rise from **12a** to the fenoterol diastereoisomer **1a**, identical to the *RR*,*SS* marketed drug (mixed mp, HPLC, ^{13}C NMR spectra) and from **12b** to the diastereoisomer **1b** to which the *RS*,*SR* configuration was consequently assigned. The protected fenoterol diastereoisomers **12a** and **12b** were reacted with either bis(phenylsulfonyl)furoxan **13** or the analogue furazan **14** in THF solution in the presence of 50% NaOH (Scheme 3). Treatment with HCl-saturated methanol and then with 10% $NaHCO_3$ gave rise to the final target compounds **15a**, **16a**, **15b**, **16b**, which were isolated as oxalates. The protection of the amino group of **10** was accomplished with $(Boc)_2O$ to give **17** (Scheme 4). Finally, the appropriately substituted furoxan and furazan derivatives **18**−**21** were coupled to the protected amine **17** in THF solution in the presence of PPh_3 and diisopropyl azodicarboxylate (DIAD; Mitsunobu reaction), affording **22**−**25**. The reaction of these latter amines with **6** in refluxing methanol yielded the intermediate mixtures of diastereoisomers **26a**−**29a** + **26b**−**29b**, which were treated with $(Boc)_2O$ in CH_2Cl_2 to give the corresponding mixtures **30a**−**33a** + **30b**−**33b** (Scheme 5). Each mixture was resolved into the two corresponding diastereoisomers by direct phase HPLC method (see Experimental Section), giving **30a**−**33a** and **30b**−**33b**. The single diastereoisomers were deprotected in acidic conditions, affording the final pure diastereoisomers **34a**−**37a** and **34b**−**37b**.

Scheme 4. Synthesis of Intermediates **22**−**25**[a]



[a] Reagents and conditions: (a) Et_3N, $(Boc)_2O$, H_2O, CH_3CN; (b) PPh_3, DIAD, compound **18**−**21**, dry THF, 0 °C to rt; (c) TFA, CH_2Cl_2, 10% aq Na_2CO_3.

Scheme 5. Synthesis of Final Compounds **34a**−**37a** and **34b**−**37b**[a]



[a] Reagents and conditions: (a) MeOH, reflux; (b) $(Boc)_2O$, CH_2Cl_2, rt; (c) HPLC separation; (d) 1 N HCl; (e) 10% aq $NaHCO_3$, $H_2C_2O_4$/EtOAc.

The assignment of the configurations to the couples of diasteroisomers **15a**, **15b** and **16a**, **16b** follows immediately

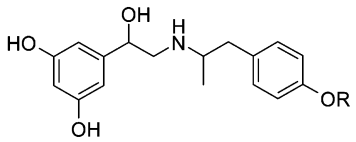
from the synthetic pathway used to prepare these products from *RR*,*SS* **12a** and *RS*,*SR* **12b**. The configurations of the other diastereoisomer fenoterol derivative pairs were assigned on the basis of a comparison of their ^{13}C NMR spectra with those of **1a** and **1b**. On this subject, the signals belonging to *C*HOH, *C*HCH$_3$, and *C*H$_3$ groups, spatially close to the chiral centers, are of particular utility. In the case of **1a**, **1b**, and in the related stereoisomers **15a**, **16a**, **15b**, and **16b**, the carbon chemical shifts of all these groups are always upfield in the *RR*,*SS* forms with respect to *RS*,*SR* forms (see Experimental Section). Consequently, the configurations of all other diastereoisomers were assigned on the basis of this statement.

NO Release. The ability of the furoxan diastereoisomers **15a**, **15b**, **34a**, **34b**, **36a**, and **36b** to release NO was indirectly evaluated through their capacity to produce nitrite in 37 °C water solution, at physiological pH (7.4), in the presence of a strong excess of cysteine. Nitrite is the most important product of the oxidation of NO in aerobic aqueous solution. It was detected by Griess reaction, according to the procedure we previously used.[14] The results, expressed as NO_2^-% (mol/mol) are described in Table 1. The ability of the products to generate nitrite generates the rank order **15a**, **15b** > **34a**, **34b** > **36a**, **36b**.

NO release was also evaluated on **15a**, **34a**, and **36a** under conditions more similar to physiological ones, using a Clark-type electrode. The experiments were carried out in phosphate buffer (pH 7.4) in the presence of ascorbate (1 mM) and glutathione (3 mM), following the release over 10 min. The release profile for **15a** is reported in Figure 1. The product when tested at 1 μM concentration showed a peak concentration of 0.31 ± 0.03 μM after about half a minute, and after about 2 min, the release was complete. The NO release from **34a** was evaluated under the same conditions. When **34a** was tested at 1, 10, and 100 μM concentrations, the NO released was not detectable, however, when the product was tested at 500 μM concentration a signal corresponding to the NO release was detected (Figure 1). The peak concentration was 0.64 ± 0.04 μM after about 10 min, and the release, after this time, was not complete. NO release from **36a** (500 μM) was undetectable when the product was tested under the aforementioned conditions. In conclusion, **15a** appears to be a more potent and faster NO donor with respect to **34a**.

Biological Results. Myorelaxing Activity. The myorelaxing effects of the two diastereoisomers of fenoterol **1a** and **1b**, the target furoxan products **15a**, **34a**, **36a**, **15b**, **34b**, and **36b**, and related furazans **16a**, **35a**, **37a**, **16b**, **35b**, and **37b** were assessed on guinea pig tracheal rings precontracted with 1 μM carbachol. All the products were able to relax the contracted tissue in a concentration-dependent manner and produced the same maximum response as (−)-isoprenaline. The potencies of the products expressed as EC_{50} value are described in Table 1. The *RR*,*SS* diastereoisomer **1a**, the marketed form of fenoterol, displays a potent relaxant effect, about 450 times higher than that triggered by the *RS*,*SR* diastereoisomer **1b**. As expected, the higher activity of the *RR*,*SS* diasteroisomer with respect to the *RS*,*SR* diastereoisomer occurs in all of the other diastereoisomer couples studied, but in a less-pronounced manner. Both in the furoxan and in the furazan series, the potencies of the *RR*,*SS* diastereoisomers are lower than the potency of **1a**, with the only exception of phenylsulfonylfuroxan derivative **15a**, which is equipotent. By contrast, the potencies of the *RS*,*SR* diastereoisomers are higher than the potency of **1b**. The ratio between these figures ($EC_{50\mathbf{b}}/EC_{50\mathbf{a}}$) ranges from about 6 to 46. The different binding modes of the two diastereoisomers should be responsible for this behavior.

Table 1. Pharmacological Profile and Nitrite Formation of Fenoterol Derivatives and References **1a**, **1b**, **18**, **20**, and CHF 2363



Compound	Myorelaxing activity[a]			Vasodilating activity[b]	Antioxidant activity[c]	Nitrite detection[d]	CLOGP[e]
	EC_{50} (± SE μM)			EC_{50} ± SE μM	IC_{50} (95%CL) μM	$\%NO_2^-$ (± SE) mol/mol	
	10μM ODQ	100μM Propranolol		[10μM ODQ]		Cys 50×	
 **RR, SS 16a**	0.039 (± 0.004)	-	#[g]	-	57 (52-62)	-	3.84
 **RS, SR 16b**	1.8 (± 0.3)	-	#[g]	-	-	-	
 **RR, SS 15a**	0.018 (± 0.002)	0.017 (± 0.003)	2.9 (± 0.3)	0.014 ± 0.003 [0.86 ± 0.15]	13.5 (12.7-14.3)	28.4 (± 0.3)	3.73
 **RS, SR 15b**	0.25 (± 0.02)	0.44**[f] (± 0.05)	3.2 (± 0.3)	0.014 ± 0.004 [1.3 ± 0.2]	-	27.5 (± 0.3)	
 **RR, SS 35a**	0.029 (± 0.005)	-	#[g]	-	286 (284-310)	-	0.87
 **RS, SR 35b**	0.17 (± 0.02)	-	#[g]	-	-	-	
 **RR, SS 34a**	0.11 (± 0.02)	0.13 (± 0.02)	24 (± 3)	1.2 ± 0.2 [14 ± 6]	156 (147-164)	11.5 (± 0.3)	0.64
 **RS, SR 34b**	1.0 (± 0.2)	1.1 (± 0.1)	22 (± 2)	1.2 ± 0.2 [16 ± 1]	-	10.4 (± 0.2)	
 **RR, SS 37a**	0.11 (± 0.02)	-	#[g]	-	123 (110-139)	-	1.58
 **RS, SR 37b**	2.2 (± 0.4)	-	#[g]	-	-	-	
 **RR, SS 36a**	0.22 (± 0.02)	0.23 (± 0.06)	54 (± 1)	18 ± 4 [#[g]]	298 (284-312)	< 1%	1.21
 **RS, SR 36b**	1.3 (± 0.1)	1.2 (± 0.2)	55 (± 2)	20 ± 2 [#[g]]	-	< 1%	
R = H **RR, SS 1a**	0.018 (± 0.002)	0.019 (± 0.003)	#[g]	Inactive	229 (201-261)	-	0.83 (MlogP)
R = H **RS, SR 1b**	8.2 (± 0.5)	-	-	-	215 (190-243)	-	0.98
 **CHF 2363**	2.3 (± 0.2)	#[g]	2.2 (± 0.1)	0.012 ± 0.002 [1.2 ± 0.2]	110 (98-122)	36.0 (± 6.0)	
 **18**	86 (± 7)	-	86 (± 10)	6.3 ± 0.8 [#[g]]	Inactive at 1mM	12.0 (± 1.0)	
 **20**	#[g]	-	-	#[g]	Inactive at 1 mM	< 1%	

[a] Determined on guinea pig tracheal rings precontracted with 1 μM carbachol. [b] Determined on rat thoracic aorta precontracted with 1 μM phenylephrine. [c] Measured as the ability of the compounds **15a**, **34a**, **35a**, **36a**, and **37a** tested as the hydrochlorides to inhibit lipid peroxidation in rat hepatic microsomial membranes. [d] A solution of the appropriate compound (100 μM) in 50 mM phosphate buffer (pH 7.4) and 1% DMSO containing 5 mM L-cys was incubated at 37 °C for 1 h; 1 mL of the reaction mixture was treated with the Griess reagent (250 μL).[14] No production of nitrite was observed in the absence of l-cys. [e] Calculated with Bio-Loom for Windows v. 1.5, BioByte Corp. [f] $P < 0.01$ when compared to EC_{50} value in the absence of ODQ, Student's *t*-test for unpaired values. [g] Myorelaxation did not reach 50% at maximal concn tested (100 μM).

The furoxan derivatives **15a**, **15b**, **34a**, **34b**, **36a**, and **36b** and fenoterol diastereoisomer **1a** (as the reference) were then tested for their myorelaxing activity in the presence of 10 μM 1*H*-[1,2,4]oxadiazolo-[4,3-*a*]quinoxalin-1-one (ODQ), a well-known inhibitor of soluble guanylate cyclase (sGC), to evidence the possible contribution to the relaxation exerted by the released NO.[15] No variation in the concentration−response curves was observed when products **15a**, **34a**, **34b**, **36a**, and **36b** and the reference **1a** were employed; this excludes any valuable contribution to the relaxation by NO. The only exception is the

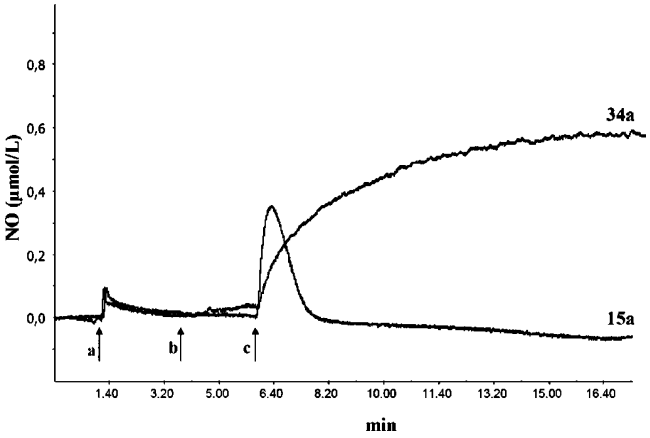


Figure 1. Release of NO from **15a** (1 μM) and **34a** (500 μM). The NO electrode was inserted into a 10 mL (final volume) aliquot of phosphate buffer, pH 7.4. Arrows indicate the time points of consecutive additions of tested compound (a), sodium ascorbate (1 mM; b) and glutathione (3 mM; c).

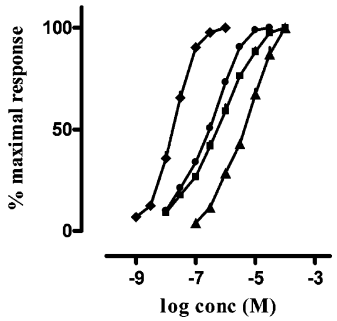


Figure 2. Concentration−response curves for myorelaxing activity of **1a** (diamond), **15b** (circle), **15b** in the presence of 10 μM ODQ (square), and **15b** in the presence of 100 μM propranolol (triangle) on precontracted guinea pig trachea.

RS,*SR* diastereoisomer **15b**, for which a decrease in the relaxing potency was observed. To determine whether there was any NO-dependent myorelaxant activity, the NO-donor hybrids were then studied in the presence of 100 μM propranolol, a well-known potent β-antagonist.[16] In our experimental conditions, this high concentration of propranolol proved to be able to almost completely abolish the β_2-mediated relaxant activity on guinea pig trachea of fenoterol **1a** and of the furazan derivatives **16a**, **16b**, **35a**, **35b**, **37a**, and **37b**, which are structurally related to furoxan analogues but devoid of the ability to release NO.

Under these experimental conditions, the activity of furoxan hybrids was not completely abolished, and EC_{50} values in the μM range were obtained. This residual activity is likely to be NO-dependent because similar myorelaxing potency (EC_{50}s) was obtained for the simple NO donor furoxan models **18** and CHF 2363. To further confirm this hypothesis, we repeated the experiments in the presence of both 100 μM propranolol and 10 μM ODQ. Under these conditions, EC_{50} values were not calculable for all the compounds because myorelaxation did not reach 50% at maximal concentration tested (100 μM).

Analysis of the concentration−response curves obtained under different conditions used showed that there is no overlapping between the concentration range in which the two relaxations, β_2-receptor-dependent and NO-dependent, respectively, occur. The only exception is product **15b** in which partial overlapping occurs (Figure 2). Consequently, this compound can be considered a "well-balanced" hybrid in vitro in a limited concentration range.

Antioxidant Properties. As outlined above, β_2-agonists have in vitro antioxidant functions. These are strictly correlated to the presence of phenol groups within their structures. Phenols are easily oxidized; this capacity is higher for the products containing more than an aromatic hydroxyl group. The antioxidant function of β_2-agonists may be of interest for their application in the treatment of a number of airways diseases.[17] As far as fenoterol is concerned, its ability to inactivate superoxide anion, hydrogen peroxide, hydroxyl radical, and hypochlorous acid has been established.[2] Consequently, we evaluated the antioxidant properties of fenoterol diastereoisomers **1a**, **1b**, and of all the fenoterol derivatives of series **a**, possessing the same configuration of marketed fenoterol, through their ability to inhibit the ferrous salt/ascorbate-induced peroxidation of lipids present in microsomial membranes of rat hepatocytes. The progress of this reaction was followed using visible spectroscopy to detect the thiobarbituric reactive species (TBARS) produced in the oxidation. This is a very common technique used in this kind of work.[18] The antioxidant potencies of the two diastereoisomers of fenoterol and of the *RR*,*SS* diastereoisomer of each furazan and furoxan derivative of fenoterol, expressed as IC_{50}, namely, the concentration able to inhibit 50% of the TBARS formation, are collected in Table 1 together with the calculated log P. As expected, the antioxidant potencies of **1a** and **1b** are the same. All the other products display antioxidant activities in a range roughly near to that of fenoterol, with the exception of **15a** and **16a**. It is likely that the higher antioxidant potencies of these two products are principally connected with their definitively higher lipophilicity. Direct measurements of log P, as well as evaluation of other molecular descriptors that could influence the antioxidant behavior of a phenol derivative carried out on a more extended series of products, are needed to confirm this hypothesis.

Vasodilator Activity. The vasodilator effects of target NO donor furoxan derivatives were assessed on denuded rat aorta strips precontracted with phenylephrine. The vasodilator potencies of the products are described in Table 1. Within a fixed couple of diastereoisomers, the vasodilator potencies are the same, according to the presence of the same NO donor moiety. The most potent vasodilators were the couple of diastereoisomers containing the phenylsulfonylfuroxan substructure **15a** and **15b**, followed by the furoxancarboxamide diasteroisomers **34a** and **34b**, and then by the methylfuroxan ones **36a** and **36b**, keeping with the vasodilator potency of the simple related furoxans CHF 2363, **18**, and **20**. This sequence parallels their capacity to produce nitrite in 37 °C aqueous solution, at physiological pH (7.4), in the presence of a relevant excess of cysteine. When the vasodilation experiments were repeated in the presence of ODQ, a strong decrease in the potencies occurred. This is in keeping with the NO-mediated activation of the sGC.

Conclusion

We have synthesized a series of diasteroisomer couples of fenoterol analogues containing NO donor furoxan moieties and a series of the related furazans. The configuration of the single diastereoisomers was assigned by chemical or spectroscopic correlations with the two diastereoisomers of fenoterol. Almost all the products were able to relax precontracted guinea pig tracheal rings through β_2-mediated receptor mechanism at submicromolar concentrations. They were full β_2-agonists in this preparation possessing the same efficacy as (−)-isoprena-

line. The *RR*,*SS* series were more potent than the *RS*,*SR* ones. In the case of the NO donor furoxan derivatives, NO-dependent relaxation also occurs at higher concentrations and in equipotent manner in the two diastereoisomers of each couple. The only exception is the furoxan derivative **15b**, which triggers relaxation simultaneously through both of the mechanisms in a limited concentration range. All the analogues of fenoterol **1a**, the marketed drug, display antioxidant activity particularly marked in the quite lipophilic phenylsulfonyl-substituted models. The products containing NO donor furoxan moieties were able to dilate precontracted rat aorta strips in keeping with a NO-mediated activation of the sGC. The NO-donor analogues of fenoterol described in the present paper represent a new interesting class of polyvalent drugs potentially useful in a number of airways and cardiovascular disorders.

Experimental Section

Chemistry. Melting points were determined with a capillary apparatus (Büchi B-540). ^{1}H and ^{13}C NMR spectra were obtained on a Bruker Avance 300, at 300 and 75 MHz, respectively; δ in ppm relative to $SiMe_4$ as the internal standard; coupling constants J in Hz. ^{13}C NMR spectra were fully decoupled. The following abbreviations are used: s, singlet; d, doublet; t, triplet; q, quartet; dd, double doublet; m, multiplet. Mass spectra were recorded on a Finnigan-Mat TSQ-700.

HPLC measurements were carried out on a Varian Pro-Star 210 chromatograph equipped with a variable wavelength detector (Pro-Star 325). The chromatography was performed using a 7 μm particle size Hibar prepacked column (250 mm × 10 mm), with a flow rate of 20 mL/min.; the detection was performed at 280 nm. Samples of the mixtures were dissolved (ca 0.5%) in mobile phase and used in all chromatographic measurements.

Silica gel 60 F254 precoated plates on glass from Merck was used for thin-layer chromatography. Flash chromatography (FC) was performed on BDH silica gel (particle size 40−63 μm). When not otherwise specified, anhydrous magnesium sulfate ($MgSO_4$) was used as the drying agent of organic phases. Analysis (C, H, N) of the new compounds was performed by REDOX (Monza), the analytical results obtained were within ±0.4% of the theoretical value. Compounds **13**,[19] **14**,[20] **19**,[21] **20**,[22] **21**,[23] and CHF 2363[19] were synthesized according to literature. Product **18** (CAS 1609) was a kind gift from Sanofi-Aventis Deutschland GmbH. *rac*-Fenoterol hydrobromide was purchased from Sigma Aldrich. The NO released was measured by means of an ISO-NO meter equipped with a 2 mm diameter shielded microsensor ISO-NOP and a ISO-NO Mark II data recording system from World Precision Instrument (Sarasota, FL).

1-(3,5-Bis{[*tert*-butyl(dimethyl)silyl]oxy}phenyl)-2-chloroethanone (4). To a stirred solution of **2** (5 g; 21.1 mmol) in $CHCl_3$ (500 mL), bromine (1.29 mL; 25.3 mmol) was added, after 30 min, the solution was washed with 10% $NaHCO_3$ (5 × 30 mL) and then with brine (2 × 30 mL), and the organic layer was dried and concentrated in vacuo to leave an oily residue, which was taken up with EtOH (50 mL) and treated with 5 N HBr (20 mL). The mixture was refluxed for 30 min, cooled, poured into water (500 mL), and then extracted with Et_2O (5 × 20 mL). The organic phase was worked up as described above to afford a pale yellow oil, which was readily dissolved in DMF (5 mL). The obtained solution was added dropwise into a solution of TBDMSCl (6.87 g; 45.6 mmol) and DIPEA (9.9 mL; 57 mmol) in DMF (20 mL) kept at 0 °C. When the addition was over, the reaction mixture was stirred at room temperature for 1 h, diluted with water (200 mL), and extracted with Et_2O (5 × 20 mL). The ethereal layer was washed with 10% NaOH (3 × 30 mL), 10% aqueous citric acid (3 × 30 mL), and brine (2 × 30 mL) and the organic phase was dried and evaporated to obtain the crude product as a yellow oil. The product was purified by FC on silica gel (eluent, petroleum ether/CH_2Cl_2 = 8/2 then 7/3) to give pure **4** as a pale yellow oil (7.00 g; 78%): ^{1}H NMR ($CDCl_3$) δ 7.04 (2H, d, Ph-2,6-H, J = 2.2 Hz), 6.59 (1H, t, Ph-4-H, J = 2.2 Hz), 4.65 (2H, s, CH_2Cl), 1.00 (18H, s, $C(CH_3)_3$), 0.24 (12H, s, CH_3Si). ^{13}C NMR ($CDCl_3$) δ 190.4, 160.4, 136.0, 117.7, 113.4, 46.1, 25.6, 18.2, −4.4. MS (CI) m/z 413−415 $(M + H)^+$. Anal. ($C_{20}H_{35}ClO_3Si_2 \cdot 1/3H_2O$, 421.06) C, H.

***tert*-Butyl-(3-{[*tert*-butyl(dimethyl)silyl]oxy}-5-oxiran-2-ylphenoxy)dimethylsilane (6).** To a solution of **4** (4.42 g; 10.7 mmol) in MeOH (50 mL), $NaBH_4$ (0.1 g; 2.7 mmol) was added at 0 °C, and the reaction was stirred at room temperature for 30 min. The solvent was evaporated in vacuo, and the residue was taken up with brine (100 mL) and extracted with EtOAc (3 × 20 mL). The organic layer was dried and concentrated in vacuo to leave an oil that was dissolved in dry THF (100 mL), treated with 60% NaH in mineral oil (0.35 g; 8.8 mmol), and stirred under nitrogen for 24 h. The reaction mixture was cooled to 0 °C and diluted with water (20 mL), and the THF was evaporated in vacuo. The residual product was taken up with brine (50 mL), extracted with Et_2O (3 × 10 mL), dried, and concentrated in vacuo to afford crude **6** as a cream-colored oil. The product was purified by FC on silica gel (eluent, petroleum ether/EtOAc = 9.5/0.5) to yield pure **6** as a colorless oil (1.64 g; 60%): ^{1}H NMR ($CDCl_3$) δ 6.39 (2H, d, Ph-2,6-H, J = 2.2 Hz), 6.26 (1H, t, Ph-4-H, J = 2.2 Hz), 3.73 (1H, q, PhCH, J = 4.1, 2.5 Hz), 3.08 (1H, dd, oxirane-cis, J_{gem} = 5.6, J_{cis} = 4.1 Hz), 2.71 (1H, dd, oxirane-trans, J_{gem} = 5.6, J_{trans} = 2.5 Hz), 0.97 (18H, s, $C(CH_3)_3$), 0.18 (12H, s, CH_3Si); ^{13}C NMR ($CDCl_3$) δ 156.8, 139.8, 111.9, 110.4, 52.2, 51.1, 25.7, 18.2, −4.2; MS (CI) m/z 381 $(M + H)^+$. Anal. ($C_{20}H_{36}O_3Si_2$, 380.67) C, H.

1-(4-Hydroxyphenyl)propan-2-amonium Chloride (10). To a stirred solution of **7** (4.68 mL; 30.5 mmol) in EtOH (40 mL), hydroxylamine hydrochloride (4.92 g; 71.3 mmol) in water (25 mL) was added, and the reaction pH was adjusted to 8 by the addition of 10% NaOH. The mixture was refluxed for 15 min, cooled, and treated with water (100 mL) to obtain a white solid, which was collected. After drying, the oxime **8** was dissolved in NH_3-saturated methanol solution (50 mL) and transferred in the Parr hydrogenator. Ni catalyst[12] (10%) was added, and the mixture was hydrogenated for 24 h at 30 bar, keeping the temperature at 68 °C. The mixture was filtered to remove the catalyst, and the solvent was evaporated in vacuo. The residual semisolid material was taken up with HCl-saturated MeOH and precipitated by treatment with Et_2O to obtain the hydrochloride salt (**9**). This salt was dissolved in concd HCl (50 mL) and heated at 140 °C in a closed vessel for 24 h. The dark solution was evaporated in vacuo, the residual solid was dissolved in abs EtOH (100 mL), treated with decolorizing charcoal, and refluxed for an additional 24 h. After filtering through celite, the solvent was evaporated in vacuo to leave a cream-colored solid, which was recrystallized from abs EtOH/Et_2O to give **10** (4.25 g; 75%) as a white solid, mp 172.9−173.8 °C (lit.[13] 171−172 °C); ^{1}H NMR (CD_3OD) δ 7.08 (2H, d, Ph-2,6-H, J = 8.4 Hz), 6.78 (2H, d, Ph-3,5-H, J = 8.4 Hz), 3.45 (1H, m, CH), 2.93 (1H, dd, (C*H*)H, J = 13.6, 8.1 Hz), 2.71 (1H, dd, (C*H*)H, J = 13.6, 8.3 Hz), 1.25 (3H, d, CH_3, J = 6.6 Hz). ^{13}C NMR (CD_3OD) δ 158.3, 132.0, 128.5, 117.2, 51.1, 41.5, 18.8.

General Procedure for the Synthesis of Fenoterol Diastereoisomers (1a,b). To a solution of **10** (1.19 g; 6.4 mmol) and Et_3N (0.88 mL; 6.4 mmol) in MeOH (15 mL), oxirane **6** (0.8 g; 2.1 mmol) was added and the mixture was refluxed for 24 h. The solvent was removed in vacuo to leave a yellowish oil (**11**), and the product, without further purification, was reacted with $(Boc)_2O$ in CH_2Cl_2 at room temperature to obtain **12a** + **12b** as a diastereoisomeric mixture (*RR*,*SS*; *RS*,*SR*). The separation of mixture into both racemic pairs (*RR*,*SS*)**a** and (*RS*,*SR*)**b** was carried out with a direct phase HPLC method, eluting with CH_2Cl_2/MeOH 99:1% v/v. The first eluted compound **12a** (t_R: 7.1) was then hydrolyzed in MeOH/1 N HBr to give **1a** (*RR*,*SS*); with the same acidic treatment, the second eluted compound **12b** (t_R: 8.3) was hydrolyzed to give **1b** (*RS*,*SR*). The obtained compounds were freeze-dried.

***rac*-(2*R*)-*N*-[(2*R*)-2-(3,5-Dihydroxyphenyl)-2-hydroxyethyl]-1-(4-hydroxyphenyl)propan-2-amonium Bromide (1a).** White solid (1.02 g, 46%); mp 223.5−225 °C (lit.[24] 223.8−224.5 °C); ^{1}H NMR (CD_3OD) δ 7.08 (2H, d, H-2,6-PhOH, J = 8.4 Hz), 6.76 (2H, d,

H-3,5-PhOH, $J = 8.4$ Hz), 6.40 (2H, d, Ph-2,6-H, $J = 2.1$ Hz), 6.23 (1H, m, Ph-4-H), 4.90 (1H, d, C*H*OH, $J = 3.3$ Hz), 3.52−3.48 (1H, m, C*H*CH_3), 3.24−3.08 (3H, m, C*H*$_2$NH, H(C*H*)PhOH), 2.71−2.63 (1H, m, H(C*H*)PhOH), 1.22 (3H, d, CH_3, $J = 6.6$). ^{13}C NMR (CD_3OD) δ 159.9, 157.7, 144.7, 131.4, 128.2, 116.6, 105.3, 103.3, *70.1*, *57.2*, 52.5, 39.6, *15.5* (chemical shifts, which are italicized, were used for assigning the diastereoisomers).

***rac*-(2*S*)-*N*-[(2*R*)-2-(3,5-Dihydroxyphenyl)-2-hydroxyethyl]-1-(4-hydroxyphenyl)propan-2-amonium Bromide (1b).** White solid (1.02 g, 46%), mp 187−188.4 °C (lit.[24] 187.4−188.2 °C); ^{1}H NMR (CD_3OD) δ 7.07 (2H, d, H-2,6-PhOH, $J = 8.4$ Hz), 6.76 (2H, d, H-3,5-PhOH, $J = 8.1$ Hz), 6.38 (2H,m, Ph-2,6-H), 6.22 (1H, m, Ph-4-H), 4.83−4.79 (1H, m, C*H*OH), 3.45−3.34 (1H, m, C*H*CH_3), 3.23−3.18 (1H, m, H(C*H*)PhOH), 3.09−2.99 (2H, m, C*H*$_2$NH), 2.69−2.61 (1H, m, H(C*H*)PhOH), 1.20 (3H, d, CH_3, $J = 5.7$). ^{13}C NMR (CD_3OD) δ 160.2, 157.8, 145.3, 131.8, 128.6, 116.9, 105.7, 103.6, *71.1*, *57.5*, 53.2, 39.9, *16.3*. Anal. ($C_{17}H_{21}NO_4$·HBr·1/3H_2O, 390.9) C, H, N. (chemical shifts, which are italicized, were used for assigning the diastereoisomers).

General Procedure for the Synthesis of Derivatives 15a, 15b, 16a, and 16b. To a solution of **12a** or **12b** (0.94 mmol) and furoxan **13** or furazan **14** (1.13 mmol) in distilled THF (30 mL), stirred under nitrogen, 50% NaOH (0.22 g; 2.8 mmol) was added dropwise. After 1 h at room temperature, the solvent was evaporated in vacuo, and the residue was taken up with water (50 mL) and extracted with EtOAc (4 × 10 mL). The organic phase was dried (K_2CO_3), and the solvent was concentrated in vacuo to give a pale yellow oil. This oil was readily dissolved in HCl-saturated MeOH (30 mL), stirred for 2 h, and concentrated in vacuo to leave a yellow foam. The product was taken up with 10% $NaHCO_3$ and extracted with EtOAc (5 × 10 mL). The organic layer was dried, the solvent was evaporated in vacuo, and the obtained residue was purified by FC on silica gel (eluent, $CH_2Cl_2/MeOH_{(NH_3)} = 9.5/0.5$) to give the expected product as the free base. The product was converted into the corresponding oxalate, recrystallized from MeOH/Et_2O, and freeze-dried.

***rac*-(2*R*)-*N*-[(2*R*)-2-(3,5-Dihydroxyphenyl)-2-hydroxyethyl]-1-(4-[(3-phenylsulfonylfuroxan-4-yl)]oxy]phenyl)propan-2-amonium Oxalate (15a).** White solid (0.61 g; 42%), mp 128.3−129.6 °C; ^{1}H NMR (CD_3OD) δ 8.03 (2H, d, H-2,6-PhSO_2, $J = 7.5$ Hz), 7.81 (1H, t, H-4-PhSO_2, $J = 7.5$ Hz), 7.66 (2H, t, H-3,5-PhSO_2, $J = 7.5$ Hz), 7.29 (4H, m, H aromatics phenylpropylaminic substructure, $J = 8.7$ Hz), 6.39 (2H, m, 2,6-H aromatics phenylethanolaminic substructure), 6.20 (1H, m, 4-H aromatic phenylethanolaminic substructure), 4.79 (1H, m, C*H*OH overlapping with HOD), 3.54 (1H, m, C*H*CH_3), 3.32−3.09 (3H, m, C*H*$_2$NH, H(C*H*)-PhO-), 2.81−2.73 (1H, m, H(C*H*)PhO-), 1.20 (3H, d, CH_3, $J = 6.3$ Hz). ^{13}C NMR (CD_3OD) δ 168.7, 159.9, 159.8, 153.3, 144.8, 139.2, 137.1, 136.4, 132.1, 131, 129.7, 121.2, 112.2, 105.3, 103.3, 70.3, 56.7, 52.7, 39.6, 15.4. Anal. ($C_{25}H_{25}N_3O_8S$·$H_2C_2O_4$·2/3H_2O, 629.48) C, H, N.

***rac*-(2*S*)-*N*-[(2*R*)-2-(3,5-Dihydroxyphenyl)-2-hydroxyethyl]-1-(4-[(3-phenylsulfonylfuroxan-4-yl)]oxy]phenyl)propan-2-amonium Oxalate (15b).** White solid (0.61 g; 42%), mp 94.4−95.6 °C; ^{1}H NMR (CD_3OD) δ 8.06 (2H, d, H-2,6-PhSO_2, $J = 7.6$ Hz), 7.84 (1H, t, H-4-PhSO_2, $J = 7.6$ Hz), 7.69 (2H, t, H-3,5-PhSO_2, $J = 7.6$ Hz), 7.33 (4H, m, H aromatics phenylpropylaminic substructure, $J = 8.5$ Hz), 6.40 (2H, m, 2,6-H aromatics phenylethanolaminic substructure), 6.22 (1H, m, 4-H aromatic phenylethanolaminic substructure), 4.79 (1H, m, C*H*OH overlapping with HOD), 3.57 (1H, m, C*H*CH_3), 3.27−3.22 (2H, m, C*H*$_2$NH), 3.16−3.08 (1H, m, H(C*H*)PhO-), 2.78 (1H, m, H(C*H*)PhO-), 1.24 (3H, d, CH_3, $J = 6.2$ Hz). ^{13}C NMR (CD_3OD) δ 168.5, 159.9, 159.8, 153.4, 144.8, 139.3, 137.1, 136.3, 132.1, 131, 129.7, 121.2, 112.2, 105.3, 103.3, 70.5, 56.9, 52.7, 39.1, 16.1. Anal. ($C_{25}H_{25}N_3O_8S$·$H_2C_2O_4$·1.5H_2O, 644.59) C, H, N.

***rac*-(2*R*)-*N*-[(2*R*)-2-(3,5-Dihydroxyphenyl)-2-hydroxyethyl]-1-(4-[(3-phenylsulfonylfurazan-4-yl)]oxy}phenyl)propan-2-amonium Oxalate (16a).** White solid (0.66 g; 46%), mp 100.4−101.0 °C; ^{1}H NMR (CD_3OD) δ 8.14 (2H, d, H-2,6-PhSO_2, $J = 7.6$ Hz), 7.85 (1H, t, H-4-PhSO_2, $J = 7.6$ Hz), 7.72 (2H, t, H-3,5-PhSO_2, $J = 7.6$ Hz), 7.34 (2H, d, 2,6-H aromatics phenylpropylaminic substructure, $J = 8.5$ Hz), 7.24 (2H, d, 3,5-H aromatics phenylpropylaminic substructure, $J = 8.5$ Hz), 6.40 (2H, m, 2,6-H aromatics phenylethanolaminic substructure), 6.20 (1H, m, 4-H aromatic phenylethanolaminic substructure), 4.86 (1H, m, C*H*OH overlapping with HOD), 3.55 (1H, m, C*H*CH_3), 3.34−3.00 (3H, m, C*H*$_2$NH, H(C*H*)PhO-), 2.79 (1H, m, H(C*H*)PhO-), 1.21 (3H, d, CH_3, $J = 6.4$ Hz). ^{13}C NMR (CD_3OD) δ 169.1, 162.2, 159.9, 154.5, 151.3, 144.9, 139.2, 137.1, 136.4, 132.2, 131.1, 130.3, 120.6, 105.3, 103.2, 70.3, 56.7, 52.7, 39.6, 15.5. Anal. ($C_{25}H_{25}N_3O_7S$·$H_2C_2O_4$·2/3H_2O, 613.6) C, H, N.

***rac*-(2*S*)-*N*-[(2*R*)-2-(3,5-Dihydroxyphenyl)-2-hydroxyethyl]-1-(4-[(3-phenylsulfonylfurazan-4-yl)]oxy}phenyl)propan-2-amonium Oxalate (16b).** White solid (0.66 g; 46%), mp 76.1−76.6 °C; ^{1}H NMR (CD_3OD) δ 8.17 (2H, d, H-2,6-PhSO_2, $J = 7.6$ Hz), 7.88 (1H, t, H-4-PhSO_2, $J = 7.6$ Hz), 7.74 (2H, t, H-3,5-PhSO_2, $J = 7.6$ Hz), 7.38 (2H, d, 2,6-H aromatics phenylpropylaminic substructure, $J = 8.0$ Hz), 7.29 (2H, d, 3,5-H aromatics phenylpropylaminic substructure, $J = 8.0$ Hz), 6.41 (2H, m, 2,6-H aromatics phenylethanolaminic substructure), 6.24 (1H, m, 4-H aromatic phenylethanolaminic substructure), 4.86 (1H, m, C*H*OH overlapping with HOD), 3.58 (1H, m, C*H*CH_3), 3.31−3.15 (3H, m, C*H*$_2$NH, H(C*H*)PhO-), 2.80 (1H, m, H(C*H*)PhO-), 1.26 (3H, m, CH_3). ^{13}C NMR (CD_3OD) δ 163.5, 162.2, 160, 154.6, 151.3, 144.7, 139.2, 137.1, 136.3, 132.2, 131.1, 130.1, 120.7, 105.3, 103.3, 70.5, 57.0, 52.6, 39, 16. Anal. ($C_{25}H_{25}N_3O_7S$·$H_2C_2O_4$·H_2O, 619.6) C, H, N.

General Procedure for the Synthesis of Amines 22−25. To a stirred solution of **10** (5.34 mmol) in CH_3CN/water (2/1) (15 mL) at 0 °C, Et_3N (13.4 mmol), and $(Boc)_2O$ (5.34 mmol) were added. After 1 h at room temperature, the solvent was removed in vacuo, and the residual oil was taken up with water (20 mL) and extracted with EtOAc (3 × 20 mL). The organic phase was dried and the solvent was removed in vacuo to leave a white solid. The solid was dissolved in dry THF (50 mL), and the appropriate alcohol **18**−**21** (8 mmol) and PPh_3 (10.7 mmol) were added. The mixture was cooled to 0 °C, and a solution of DIAD (10.7 mmol) in dry THF (2 mL) was added dropwise. The mixture was stirred at room temperature for 24 h, and the solvent was removed in vacuo to leave the product as a yellow oil, which was dissolved in CH_2Cl_2 (6 mL) and TFA (2 mL) and stirred for an additional 1 h. The solvent was removed in vacuo, and the residue was taken up with water (30 mL) and extracted with EtOAc (2 × 20 mL). The aqueous layer was treated with 10% Na_2CO_3 (20 mL) and extracted with EtOAc (5 × 20 mL), and the organic phase was dried over anhydrous K_2CO_3 and concentrated in vacuo to afford a white solid. The solid was purified by FC on silica gel (eluent, CH_2Cl_2/MeOH = 9.5/0.5 then $CH_2Cl_2/MeOH_{(NH_3)} = 9.5/0.5$) to give the expected product.

4-[4-(2-Aminopropyl)phenoxymethyl]furoxan-3-carboxamide (22). Colorless oil (66%); ^{1}H NMR (CD_3OD) δ 7.13 (2H, d, Ph-2,6-H, $J = 8.5$ Hz), 6.97 (2H, d, Ph-3,5-H, $J = 8.5$ Hz), 5.37 (2H, s, CH_2-furoxan), 3.09−3.00 (1H, m, C*H*CH_3), 2.64−2.51 (2H, m, CH_2Ph), 1.06 (3H, d, CH_3, $J = 6.3$ Hz). ^{13}C NMR (CD_3OD) δ 158.1 (two overlapping peaks), 156.6, 134.0, 131.4, 116.1, 111.8, 62.6, 49.6, 45.8, 22.5. MS (CI) *m*/*z* 293 $(M + H)^+$. Anal. ($C_{13}H_{16}N_4O_4$·0.3H_2O, 297.69) C, H, N.

3-[4-(2-Aminopropyl)phenoxymethyl]furazan-4-carboxamide (23). Colorless oil (60%); ^{1}H NMR (CD_3OD) δ 7.13 (2H, d, Ph-2,6-H, $J = 8.5$ Hz), 6.96 (2H, d, Ph-3,5-H, $J = 8.5$ Hz), 5.46 (2H, s, CH_2-furazan), 3.09−3.03 (1H, m, C*H*CH_3), 2.59−2.54 (2H, m, CH_2Ph), 1.06 (3H, d, CH_3, $J = 6.3$ Hz). ^{13}C NMR (CD_3OD) δ 160.9, 158.1, 153.5, 149.8, 133.8, 131.4, 116.1, 60.8, 49.6, 45.6, 22.4. MS (CI) *m*/*z* 277 $(M + H)^+$. Anal. ($C_{13}H_{16}N_4O_3$·1/2 H_2O, 285.3) C, H, N.

1-Methyl-2-[4-(3-methylfuroxan-4-ylmethoxy)phenyl]ethylamine (24). Colorless oil (40%); ^{1}H NMR (CD_3OD) δ 7.19 (2H, d, Ph-2,6-H, $J = 8.5$ Hz), 7.02 (2H, d, Ph-3,5-H, $J = 8.5$ Hz), 5.22 (2H, s, CH_2-furoxan), 3.15−3.09 (1H, m, C*H*CH_3), 2.64−2.62 (2H, m, CH_2Ph), 2.22 (3H, s, CH_3-furoxan) 1.11 (3H, d, CH_3, $J = 6.4$ Hz). ^{13}C NMR (CD_3OD) δ 156.7, 155.6, 133.1, 130.5, 115.0, 113.2,

61.3, 48.6, 44.5, 21.3, 6.8. MS (CI) *m*/*z* 264 $(M + H)^+$. Analytical sample was obtained as the oxalate ($C_{13}H_{17}N_3O_3 \cdot H_2C_2O_4$, 353.33) C, H, N.

1-Methyl-2-[4-(4-methylfurazan-3-ylmethoxy)phenyl]ethylamine (25). Colorless oil (35%); ^{1}H NMR (CD_3OD) δ 7.14 (2H, d, Ph-2,6-H, $J = 8.7$ Hz), 6.97 (2H, d, Ph-3,5-H, $J = 8.7$ Hz), 5.28 (2H, s, CH_2-furazan), 3.10−3.03 (1H, m, C*H*CH_3), 2.60−2.57 (2H, m, CH_2Ph), 2.42 (3H, s, CH_3-furazan) 1.06 (3H, d, CH_3, $J = 6.4$ Hz). ^{13}C NMR (CD_3OD) δ 158.1, 153.2, 153.0, 134.3, 131.9, 116.3, 60.5, 50.0, 45.9, 22.7, 8.6. MS (CI) *m*/*z* 248 $(M + H)^+$. Anal. ($C_{13}H_{17}N_3O_2 \cdot 1/3H_2O$, 253.24) C, H, N.

General Procedure for the Synthesis of Derivatives 34a−37a and 34b−37b. To a stirred solution of the appropriate amine **22**−**25** (4.10 mmol) in MeOH (15 mL), oxirane **6** (1.64 mmol) was added, and the mixture was refluxed for 24 h. The solvent was removed in vacuo to leave a yellowish oil; the product without further purification was reacted with $(Boc)_2O$ (2.46 mmol) in CH_2Cl_2 at room temperature to obtain **30a** + **30b**, **31a** + **31b**, **32a** + **32b**, and **33a** + **33b** as a diastereoisomeric mixtures (*RR*,*SS* + *RS*,*SR*). The separation of mixtures into both racemic pairs (*RR*,*SS*)-**a** and (*RS*,*SR*)**b** was carried out with a direct phase HPLC method. The mobile phase used for the mixture **30a** + **30b** and **31a** + **31b** was CH_2Cl_2/MeOH 99.5:0.5% v/v, while hex/EtOAc 85:15% v/v was used for derivatives **32a** + **32b**, **33a** + **33b**. The first eluted enantiomeric pair **30a** (t_R: 18), **31a** (t_R: 20), **32a** (t_R: 12), and **33a** (t_R: 11) was then hydrolyzed with 1 N HCl (10 mL) and stirred at room temperature for 1 h. With the same acidic treatment, the second eluted compounds **30b** (t_R: 25), **31b** (t_R: 26), **32b** (t_R: 14), and **33b** (t_R: 13) were then hydrolyzed. The aqueous solutions were basified with 10% $NaHCO_3$ and extracted with EtOAc (4 × 15 mL), and the organic layers were dried and concentrated in vacuo to leave colorless oils. The crude products were purified by FC on silica gel (eluent, $CH_2Cl_2/MeOH_{(NH_3)} = 9.5/0.5$) to give the desired products as the free base. The products were converted into the corresponding oxalates, recrystallized from MeOH/Et_2O, and freeze-dried to afford the desired product **34a**−**37a** and **34b**−**37b**.

***rac*-4-(4-{(2*R*)-[(2-(3,5-Dihydroxyphenyl)-(2*R*)-hydroxyethylamino]propyl}phenoxymethyl)furoxan-3-carboxamide Oxalate (34a).** White solid (46%), mp 131.1−132 °C; ^{1}H NMR (CD_3OD) δ 7.20 (2H, d, 2,6-H aromatics phenylpropylaminic substructure, $J = 8.1$ Hz), 7.02 (2H, d, 3,5-H aromatics phenylpropylaminic substructure, $J = 8.1$ Hz), 6.37 (2H, m, 2,6-H aromatics phenylethanolaminic substructure), 6.22 (1H, m, 4-H aromatic phenylethanolaminic substructure), 5.40 (2H, s, CH_2-furoxan), 4.80 (1H, m, C*H*OH), 3.51 (1H, m, C*H*CH_3), 3.16−3.12 (3H, m, C*H*$_2$NH, H(C*H*)PhO-), 2.72−2.64 (1H, m, H(C*H*)PhO-), 1.22 (3H, d, CH_3, $J = 6.3$ Hz). ^{13}C NMR (CD_3OD) δ 166.2, 160, 158.8, 158.1, 156.5, 144.7, 131.6, 130.6, 116.5, 111.7, 105.3, 103.3, 70.3, 62.6, 57.1, 52.5, 39.5, 15.6. Anal. ($C_{21}H_{24}N_4O_7 \cdot H_2C_2O_4 \cdot 1/2H_2O$, 543.48) C, H, N.

***rac*-4-(4-{(2*R*)-[2-(3,5-Dihydroxyphenyl)-(2*S*)-hydroxyethylamino]propyl}phenoxymethyl)furoxan-3-carboxamide Oxalate (34b).** White solid (46%), mp 116.5−117.5 °C; ^{1}H NMR (CD_3OD) δ 7.19 (2H, d, 2,6-H aromatics phenylpropylaminic substructure, $J = 8.5$ Hz), 7.00 (2H, d, 3,5-H aromatics phenylpropylaminic substructure, $J = 8.5$ Hz), 6.37 (2H, m, 2,6-H aromatics phenylethanolaminic substructure), 6.21 (1H, m, 4-H aromatic phenylethanolaminic substructure), 5.39 (2H, s, CH_2-furoxan), 4.81 (1H, m, C*H*OH), 3.49 (1H, m, C*H*CH_3), 3.28−3.05 (3H, m, C*H*$_2$NH, H(C*H*)PhO-), 2.73−2.66 (1H, m, H(C*H*)PhO-), 1.23 (3H, d, CH_3, $J = 6.4$ Hz). ^{13}C NMR (CD_3OD) δ 167.4, 160, 158.7, 158.1, 156.5, 144.7, 131.6, 130.7, 116.5, 111.7, 105.3, 103.3, 70.5, 62.6, 57.3, 52.6, 39.1, 16.2. Anal. ($C_{21}H_{24}N_4O_7 \cdot H_2C_2O_4 \cdot 1/2H_2O$, 543.48) C, H, N.

***rac*-4-(4-{(2*R*)-[2-(3,5-Dihydroxyphenyl)-(2*R*)-hydroxyethylamino]propyl}phenoxymethyl)furazan-3-carboxamide Oxalate (35a).** White solid (60%), mp 230 °C dec; ^{1}H NMR (CD_3OD) δ 7.19 (2H, d, 2,6-H aromatics phenylpropylaminic substructure, $J = 8.5$ Hz), 7.02 (2H, d, 3,5-H aromatics phenylpropylaminic substructure, $J = 8.5$ Hz), 6.37 (2H, t, 2,6-H aromatics phenylethanolaminic substructure), 6.22 (1H, t, 4-H aromatic phenylethanolaminic substructure), 5.48 (2H, s, CH_2-furazan), 4.70 (1H, m, C*H*OH overlapping with HOD), 3.51 (1H, m, C*H*CH_3), 3.30−3.15 (3H, m, C*H*$_2$NH, H(C*H*)PhO-), 2.72 (1H, m, H(C*H*)-PhO-), 1.29−1.22 (3H, m, CH_3). ^{13}C NMR (CD_3OD) δ 166.2, 160.9, 160.1, 158.8, 153.5, 149.8, 144.8, 131.6, 130.5, 116.5, 105.3, 103.4, 70.3, 60.8, 57.1, 52.5, 39.6, 15.6. Anal. ($C_{21}H_{24}N_4O_6 \cdot H_2C_2O_4 \cdot 0.2H_2O$, 522.08) C, H, N.

***rac*-4-(4-{(2*R*)-[2-(3,5-Dihydroxyphenyl)-(2*S*)-hydroxyethylamino]propyl}phenoxymethyl)furazan-3-carboxamide Oxalate (35b).** White solid (60%), mp 111.3−112.1 °C; ^{1}H NMR (CD_3OD) δ 7.18 (2H, d, 2,6-H aromatics phenylpropylaminic substructure, $J = 8.5$ Hz), 6.98 (2H, d, 3,5-H aromatics phenylpropylaminic substructure, $J = 8.5$ Hz), 6.38 (2H, m, 2,6-H aromatics phenylethanolaminic substructure), 6.21 (1H, m, 4-H aromatic phenylethanolaminic substructure), 5.46 (2H, s, CH_2-furazan), 4.84 (1H, m, C*H*OH), 3.48 (1H, m, C*H*CH_3), 3.25−3.04 (3H, m, C*H*$_2$NH, H(C*H*)PhO-), 2.72−2.65 (1H, m, H(C*H*)PhO-), 1.23 (3H, d, CH_3, $J = 6.3$ Hz). ^{13}C NMR (CD_3OD) δ 169.6, 160.9, 160.0, 158.6, 153.4, 149.8, 144.8, 131.6, 130.8, 116.4, 105.3, 103.3, 70.5, 60.8, 57.2, 52.7, 39.1, 16.2. Anal. ($C_{21}H_{24}N_4O_6 \cdot H_2C_2O_4$, 518.48) C, H, N.

***rac*-(2*R*)-*N*-[(2*R*)-2-(3,5-Dihydroxyphenyl)-2-hydroxyethyl]-1-{4-[(3-methylfuroxan-4-yl)methoxy]phenyl}propan-2-amonium Oxalate (36a).** White solid (40%), mp 125.5−126.9 °C; ^{1}H NMR (CD_3OD) δ 7.21 (2H, d, 2,6-H aromatics phenylpropylaminic substructure, $J = 8.7$ Hz), 6.99 (2H, d, 3,5-H aromatics phenylpropylaminic substructure, $J = 8.7$ Hz), 6.38 (2H, m, 2,6-H aromatics phenylethanolaminic substructure), 6.20 (1H, m, 4-H aromatic phenylethanolaminic substructure), 5.18 (2H, s, CH_2-furoxan), 4.88−4.85 (1H, m, C*H*OH), 3.55−3.48 (1H, m, C*H*CH_3), 3.23−3.07 (3H, m, C*H*$_2$NH, H(C*H*)PhO-), 2.75−2.67 (1H, m, H(C*H*)PhO-), 2.18 (3H, s, CH_3-furoxan), 1.20 (3H, d, CH_3, $J = 6.3$ Hz). ^{13}C NMR (CD_3OD) δ 168.0, 160.0, 158.3, 156.5, 144.9, 131.8, 131.1, 116.3, 114.2, 105.3, 103.2, 70.3, 62.2, 57.0, 52.7, 39.5, 15.5, 7.8. Anal. ($C_{21}H_{25}N_3O_6 \cdot H_2C_2O_4 \cdot 1/2H_2O$, 514.48) C, H, N.

***rac*-(2*S*)-*N*-[(2*R*)-2-(3,5-Dihydroxyphenyl)-2-hydroxyethyl]-1-{4-[(3-methylfuroxan-4-yl)methoxy]phenyl}propan-2-amonium Oxalate (36b).** White solid (40%), mp 93.5−94.1 °C; ^{1}H NMR (CD_3OD) δ 7.21 (2H, d, 2,6-H aromatics phenylpropylaminic substructure, $J = 8.7$ Hz), 6.99 (2H, d, 3,5-H aromatics phenylpropylaminic substructure, $J = 8.7$ Hz), 6.37 (2H, m, 2,6-H aromatics phenylethanolaminic substructure), 6.20 (1H, m, 4-H aromatic phenylethanolaminic substructure), 5.18 (2H, s, CH_2-furoxan), 4.85−4.81 (1H, m, C*H*OH), 3.53−3.46 (1H, m, C*H*CH_3), 3.27−3.02 (3H, m, C*H*$_2$NH, H(C*H*)PhO-), 2.74−2.67 (1H, m, H(C*H*)PhO-), 2.18 (3H, s, CH_3-furoxan), 1.23 (3H, d, CH_3, $J = 6.6$ Hz). ^{13}C NMR (CD_3OD) δ 168.4, 160.0, 158.3, 156.5, 144.8, 131.8, 131.2, 116.4, 114.2, 105.3, 103.3, 70.4, 62.3, 57.2, 52.7, 39.0, 16.3, 7.8. Anal. ($C_{21}H_{25}N_3O_6 \cdot H_2C_2O_4 \cdot 1/2H_2O$, 514.48) C, H, N.

***rac*-(2*R*)-*N*-[(2*R*)-2-(3,5-Dihydroxyphenyl)-2-hydroxyethyl]-1-{4-[(3-methylfurazan-4-yl)methoxy]phenyl}propan-2-amonium Oxalate (37a).** White solid (42%), mp 108.2−109.2 °C; ^{1}H NMR (CD_3OD) δ 7.20 (2H, d, 2,6-H aromatics phenylpropylaminic substructure, $J = 8.6$ Hz), 6.99 (2H, d, 3,5-H aromatics phenylpropylaminic substructure, $J = 8.6$ Hz), 6.38 (2H, m, 2,6-H aromatics phenylethanolaminic substructure), 6.20 (1H, m, 4-H aromatic phenylethanolaminic substructure), 5.29 (2H, s, CH_2-furazan), 4.87−4.83 (1H, m, C*H*OH), 3.54−3.47 (1H, m, C*H*CH_3), 3.22−3.07 (3H, m, C*H*$_2$NH, H(C*H*)PhO-), 2.74−2.67 (1H, m, H(C*H*)PhO-), 2.42 (3H, s, CH_3-furazan), 1.20 (3H, CH_3, d, $J = 6.3$ Hz). ^{13}C NMR (CD_3OD) δ 168.1, 160.0, 158.4, 152.8, 152.7, 144.9, 131.7, 130.9, 116.3, 105.3, 103.3, 70.3, 60.3, 57.0, 52.6, 39.5, 15.5, 8.3. Anal. ($C_{21}H_{25}N_3O_5 \cdot H_2C_2O_4 \cdot 0.7H_2O$, 502.09) C, H, N.

***rac*-(2*S*)-*N*-[(2*R*)-2-(3,5-Dihydroxyphenyl)-2-hydroxyethyl]-1-{4-[(3-methylfurazan-4-yl)methoxy]phenyl}propan-2-amonium Oxalate (37b).** White solid (42%), mp 103.6−104.3 °C; ^{1}H NMR (CD_3OD) δ 7.20 (2H, d, 2,6-H aromatics phenylpropylaminic substructure, $J = 8.6$ Hz), 7.00 (2H, d, 3,5-H aromatics phenyl-

propylaminic substructure, J = 8.6 Hz), 6.37 (2H, m, 2,6-H aromatics phenylethanolaminic substructure), 6.20 (1H, m, 4-H aromatic phenylethanolaminic substructure), 5.29 (2H, s, CH_2-furazan), 4.85−4.81 (1H, m, C*H*OH), 3.53−3.46 (1H, m, C*H*CH_3), 3.27−3.03 (3H, m, C*H*$_2$NH, H(C*H*)PhO-), 2.74−2.63 (1H, m, H(C*H*)PhO-), 2.41 (3H, s, CH_3-furazan), 1.23 (3H, d, CH_3, J = 6.6 Hz). ^{13}C NMR (CD_3OD) δ 168.6, 160.2, 158.7, 153.1, 152.9, 145.1, 132.0, 131.2, 116.6, 105.6, 103.5, 70.7, 60.5, 57.5, 53.0, 39.3, 16.5, 8.5. Anal. ($C_{21}H_{25}N_3O_5 \cdot H_2C_2O_4 \cdot 1/2H_2O$, 498.48) C, H, N.

Amperometric Detection of NO Release from Derivatives 15a, 34a, and 36a. The membrane-covered tip of the measuring electrode was inserted into a solution containing phosphate buffer (50 mM/pH 7.4). The solution was constantly mixed by a magnetic stirrer and kept at 37 ± 0.5 °C in a closed glass vial. The current was recorded for 15 min to allow for baseline stabilization, and then a solution of the appropriate compound **15a** (1 μM), **34a** (500 μM), or **36a** (500 μM) in 100 μL of DMSO (1% v/v in the final solution) was added via a gastight syringe. Consecutive additions of sodium ascorbate (1 mM) in HPLC-grade water (50 μL) and glutathione (3 mM) in phosphate buffer (100 μL) were performed via a gastight syringe. The final volume of the tested mixture was 10 mL. Change in the current was recorded as a function of time, and data were elaborated with a MacLab System PowerLab. Experiments were run at least in triplicate after appropriate calibration of the electrode with $NaNO_2$.[25]

Functional Studies. Tracheas were isolated from male guinea pigs weighing 200−250 g, while aortae were isolated from male Wistar rats weighing 200−250 g. The animals, treated humanely in accordance with recognized guidelines on experimentation, were anaesthetized with CO_2 and killed by decapitation. As few animals as possible were used. The purposes and the protocols of our studies have been approved by the Ministero della Salute, Rome, Italy. The tissues were mounted in organ baths containing 30 mL of Krebs-bicarbonate buffer of the following composition (mM): NaCl, 111.2; KCl, 5.0; $CaCl_2$, 2.5; $MgSO_4$, 1.2; KH_2PO_4, 1.0; $NaHCO_3$, 12; glucose, 11.1. The solution was maintained at 37 °C and continuously gassed with 95% O_2−5% CO_2 (pH 7.4).

Trachea Preparation. The tracheal tube was quickly excised, cleaned free of excess tissues, and then was cut into single rings. Four to five rings were joined together by thread to form a chain and mounted in organ baths. After 2 h equilibration period, 1 μM carbachol was added to the organ bath to induce a spasm of the trachea rings, and when a constant level was reached, cumulative concentration−response curves to compounds **15a**, **15b**, **16a**, **16b**, **34a**−**37a**, and **34b**−**37b** and the references were determined. Effects of 10 μM ODQ[16] and 100 μM propranolol were evaluated in separate series of experiments in which they were added 40 min before the start of the curve. EC_{50} values are the mean of 4−8 determinations. Responses were recorded by an isometric transducer connected to the MacLab System PowerLab.

Aorta Preparation. The aorta endothelium was removed and the vessels were helically cut: three strips were obtained from each aorta. The tissues were mounted under 1.0 g of tension in organ baths. The aortic strips were allowed to equilibrate for 2 h and then contracted with 1 μM l-phenylephrine. When the response to the agonist reached a plateau, cumulative concentrations of the vasodilating agent were added. Effects of 10 μM ODQ on relaxation were evaluated in a separate series of experiments in which it was added 5 min before the contraction. EC_{50} values are the mean of 4−8 experiments. Responses were recorded by an isometric transducer connected to the MacLab System PowerLab.

Antioxidant Activity. Hepatic microsomal membranes from male Wistar rats (200−250 g) were prepared by differential centrifugation (8000 × g, 20 min; 120 000 × g, 1 h) in a HEPES/sucrose buffer (10 mM/250 mM, pH 7.4) and stored at −80 °C. Incubation was performed at 37 °C in a Tris-HCl/KCl buffer (100 mM/150 mM, pH 7.4) containing microsomal membranes (2 mg prot/mL), ascorbic acid (100 μM), and DMSO solutions of the tested compounds. The addition of DMSO alone (maximal amount 5%) did not change significantly the extent of peroxidation in the control experiments. Lipid peroxidation was initiated by adding 2.5 μM $FeSO_4$. Aliquots were taken from the incubation mixture at 5, 15, and 30 min and treated with 10% w/v trichloroacetic acid (TCA). Lipid peroxidation was assessed by spectrophotometric (543 nm) determination of the TBARS, consisting mainly of malondialdehyde (MDA), and TBARS concentrations (expressed in nmol/mg protein) were obtained by interpolation with a MDA standard curve. The antioxidant activity of compounds, tested as hydrochloride salts, was evaluated as the percentage of inhibition of TBARS production with respect to control samples, using the plateau values obtained after 30 min of incubation. IC_{50} values were calculated by nonlinear regression analysis.

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Supporting Information Available: Tables of elemental analyses of all new compounds. This material is available free of charge via the Internet at http://pubs.acs.org.

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