Brief Articles

α_2-Adrenoreceptors Profile Modulation. 3.[1] (*R*)-(+)-*m*-Nitrobiphenyline, a New Efficient and α_{2C}-Subtype Selective Agonist

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To assess the stereochemical requirements for efficient α_{2C}-adrenoreceptor activation, the enantiomeric forms of *m*-nitrobiphenyline [(±)-**5**] were prepared and tested on cells expressing the human α_2-adrenoreceptor subtypes. The importance of chirality was confirmed, since the enantiomer (*R*)-(+)-**5** was much more efficient than (*S*)-(−)-**5** in producing α_{2C}-activation. Surprising reversal of enantioselectivity was observed with respect to structurally similar biphenyline [(±)-**1**] whose (*S*)-(−)-form proved the preferred α_{2C}-configuration.

Introduction

In a recent study,[1] we demonstrated that the presence of correctly oriented functions with positive electronic effect (+σ) in portion X of the biphenyline (**1**)[2] (Table 1) is an important factor for significant α_2-adrenoreceptor (α_2-AR) subtype selectivity. In fact, the dipole originating from functions such as pyridine nitrogen in position 3 (**2**) or such as −OH (**3**), −F (**4**), and $-NO_2$ (**5**) in the meta position (Table 1) allowed the formation of a hydrogen bond with a corresponding receptor residue. This resulted in efficient and preferential α_{2C}-subtype activation. Homology modeling and docking studies undertaken with **2** supported the experimental data and highlighted the crucial role of a bond between the pyridine nitrogen of **2** and the −NH indole ring of Trp 6.48 (according to the Ballesteros and Weinstein numbering),[3] which was favorably oriented in the α_{2C}-subtype only. The interesting α_{2C}-selectivity obtained and the well-known stereospecificity of nonsubtype selective α_2-agonists, such as α-methylnoradrenaline,[4] lofexidine,[5] medetomidine,[6] and its methylnaphthyl analogue,[7] prompted us to investigate the stereochemical requirements for the best α_{2C}-activation. Because of its particularly selective α_{2C}-subtype activation, as shown in Table 1, **5**,[1] from now on named *m*-nitrobiphenyline, was selected and its novel enantiomeric forms (*R*)-(+)-**5** and (*S*)-(−)-**5** were prepared and tested using Chinese hamster ovary (CHO) cells expressing human α_2-AR subtypes. Biphenyline (**1**) and its enantiomeric forms (*R*)-(+)-**1** and (*S*)-(−)-**1**, already prepared and partially studied[2] but never tested for their α_2-subtype selectivity, UK 14304, dexmedetomidine, and (−)-noradrenaline (NA) were also included in some assays for useful comparison.

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Chemistry

The preparation of the two enantiomers (*R*)-(+)-**5** and (*S*)-(−)-**5** is reported in Scheme 1. The (*R*)-(−)- and (*S*)-(+)-2-(3′-nitrobiphenyl-2-yloxy)propionic acid methyl esters [(*R*)-(−)-**6** and (*S*)-(+)-**6**, respectively] were obtained by the Mitsunobu reaction from the 3′-nitrobiphenyl-2-ol[8] and methyl (*S*)-(−)- and (*R*)-(+)-lactates, respectively, with inversion of configuration.[9] The reaction of (*R*)-(−)-**6** and (*S*)-(+)-**6** with ethylenediamine in the presence of $Al(CH_3)_3$ yielded the imidazolines (*R*)-(+)-**5** and (*S*)-(−)-**5** whose enantiomeric purity was <98%. In fact, the ^{1}H NMR spectra of their corresponding diastereomeric hydrogen dibenzoyl-D-tartrate salts displayed a double doublet at δ 1.51 and δ 1.50, respectively, for the methyl group of the OCHCH_3 moiety. Therefore, the two (*R*)-(+)-**5** and (*S*)-(−)-**5** forms were subsequently resolved by fractional crystallization of the hydrogen dibenzoyl-D- and hydrogen dibenzoyl-L-tartrate salts, respectively. Finally, HPLC and ^{1}H NMR spectroscopy of their corresponding diastereomeric ureidic derivatives **7a** and **7b**, obtained by reaction with (*R*)-(+)-α-methylbenzyl isocyanate, highlighted an enantiomeric purity of >98%.

An alternative synthesis of (*R*)-(+)-**5** with enantiomeric purity of >98% was performed starting from (*S*)-2-[(*p*-tolylsulfonyl)-oxy]propionamide.[10] Displacement of the *p*-tolylsulfonyl group by 3′-nitrobiphenyl-2-ol in 2-butanone in presence of CH_3OK according to the expected complete inversion of configuration[5] yielded (*R*)-(−)-**8**. The subsequent reactions do not involve the stereogenic center, and therefore, its configuration was retained.[5] Dehydration of the amide (*R*)-(−)-**8** with trifluoroacetic anhydride yielded nitrile (*R*)-(+)-**9**, which was first transformed into the imidic acid methyl ester hydrocloride and then cyclized with ethylenediamine in EtOH to give (*R*)-(+)-**5**. The unequivocal attribution of absolute configurations (*R*)-(+)-**5** and (*S*)-(−)-**5** was performed through single-crystal high-resolution X-ray diffraction analysis of the hydrogen dibenzoyl-L-tartrate salt of (−)-**5** (Figure 1).

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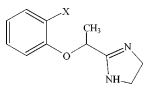
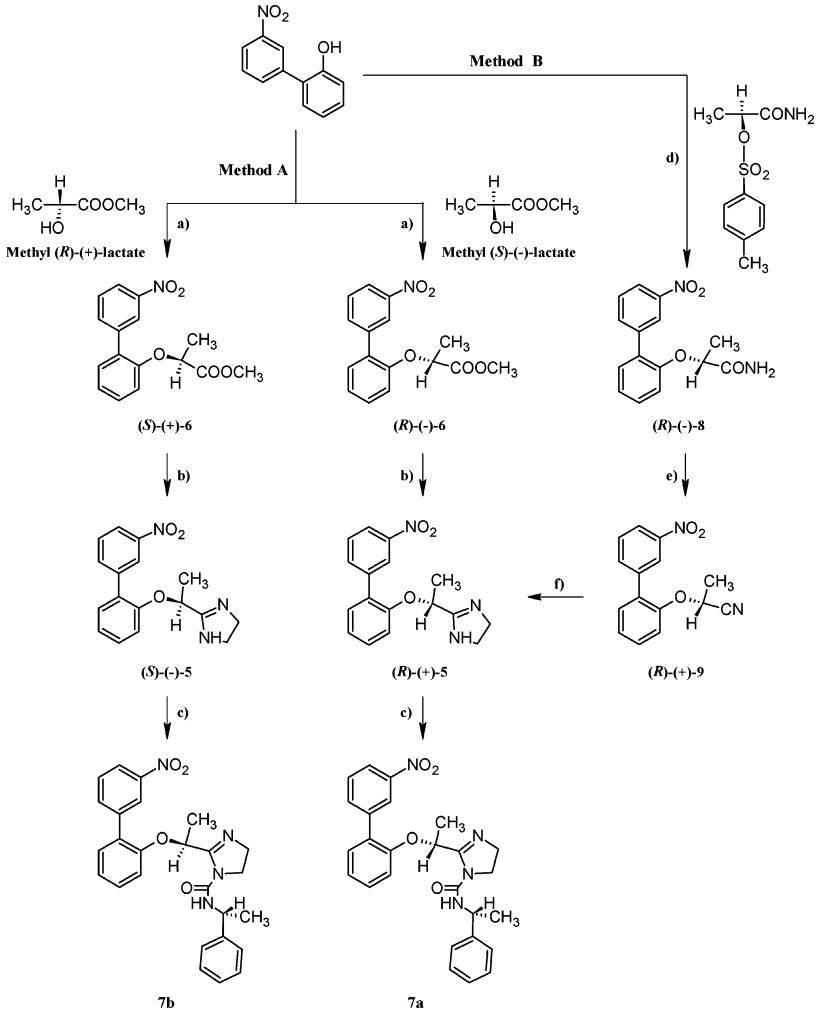
Table 1. Affinity (pK_i [a]), Potency (pEC_{50} [b]), and Intrinsic Activity (ia [b]) on Human α_2-AR Subtypes



		α_{2A}			α_{2B}			α_{2C}		
compd	X	pK_i	pEC_{50}	ia	pK_i	pEC_{50}	ia	pK_i	pEC_{50}	ia
(±)-**1**[c] biphenyline	$-C_6H_5$	7.32 ± 0.08	6.94 ± 0.06	0.70	6.30 ± 0.07	6.19 ± 0.01	0.50	6.70 ± 0.04	7.24 ± 0.01	0.80
(*R*)-(+)-**1**	$-C_6H_5$	6.73 ± 0.04	4.54 ± 0.07	0.40	5.90 ± 0.03	4.00.±0.05	0.4	6.01 ± 0.05	6.95.±0.08	0.70
(*S*)-(−)-**1**	$-C_6H_5$	7.04 ± 0.08	7.13 ± 0.03	0.80	6.23 ± 0.08	6.16 ± 0.10	0.65	6.52 ± 0.09	7.73 ± 0.14	0.90
(±)-**2**[c]	−3-pyridyl	6.86 ± 0.05	6.50 ± 0.09	0.50	6.03 ± 0.04	6.00 ± 0.09	0.50	7.19 ± 0.04	7.30 ± 0.16	1.00
(±)-**3**[c]	$-m$-OH$-C_6H_4$	7.56 ± 0.02	NA		6.45 ± 0.01	5.28 ± 0.17	0.50	7.75 ± 0.08	7.33 ± 0.15	1.15
(±)-**4**[c]	$-m$-F$-C_6H_4$	7.83 ± 0.03	6.98 ± 0.09	0.58	6.57 ± 0.07	NA		7.77 ± 0.04	7.22 ± 0.16	1.15
(±)-**5**[c] *m*-nitrobiphenyline	$-m$-$NO_2-C_6H_4$	7.22 ± 0.03	NA		6.18 ± 0.02	NA		6.63 ± 0.08	7.00 ± 0.09	0.60
(*R*)-(+)-**5**	$-m$-$NO_2-C_6H_4$	7.09 ± 0.08	NA[d]		6.13 ± 0.07	NA[d]		6.59 ± 0.09	8.00 ± 0.04	0.80
(*S*)-(−)-**5**	$-m$-$NO_2-C_6H_4$	7.14 ± 0.06	NA[d]		5.80 ± 0.06	NA[d]		6.27 ± 0.08	6.13 ± 0.01	0.45
(−)-noradrenaline			6.43 ± 0.17	1.00		7.21 ± 0.25	1.00		6.10 ± 0.05	1.00

[a] pK_i values were calculated from [^{3}H]RX 821002 inhibition experiments carried out in the absence of Mg^{2+} on membrane preparations from CHO cells expressing individually each human α_2-AR subtype (α_{2A}, α_{2B}, α_{2C}).[11] [b] pEC_{50} and intrinsic activity (ia) values were determined by applying the cytosensor microphysiometry system to the same cell models. Intrinsic activity of the tested compounds is expressed as the fraction of that of the full agonist (−)-noradrenaline taken as equal to 1. Compounds exhibiting ia of <0.3 were considered not active (NA). [c] Reference 1. [d] Antagonist profile (see text).

Scheme 1[a]



[a] Reagents: (a) DEAD, PPh_3, dry THF; (b) $Al(CH_3)_3$, $NH_2CH_2CH_2NH_2$, dry toluene, Δ; (c) *R*-(+)-α-methylbenzyl isocyanate; (d) CH_3OK, 2-butanone; (e) trifluoroacetic anhydride/dioxane; (f) HCl_g/MeOH, $NH_2CH_2CH_2NH_2$, absolute EtOH.

Results and Discussion

From data reported in Table 1, in agreement with what was previously observed for other agonists,[7] the pK_i values indicated that each couple of enantiomers displayed comparable affinity for a given receptor subtype. Moreover, (±)-**1** and (±)-**5** and their corresponding enantiomers bound α_2-subtypes with an identical rank order, their affinity for the α_{2C}-subtype being higher than that for the α_{2B}-subtype but lower than that for α_{2A}-subtype. Nevertheless, the crucial role of chirality was highlighted by pEC_{50} values determined through microphysiometry instrument.[1] In fact, (*R*)-(+)-**5**, endowed with the highest α_{2C}-agonist potency (pEC_{50} = 8.00; intrinsic activity ia = 0.80) proved 74-fold more active and 2-fold more efficient than (*S*)-(−)-**5** (pEC_{50} = 6.13; ia = 0.45). Moreover, (*R*)-(+)-**5** and

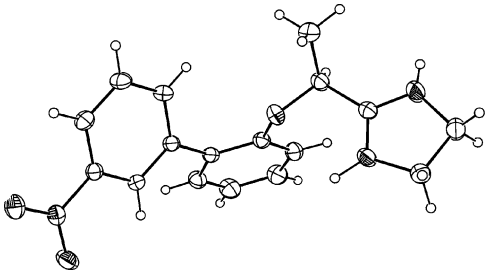


Figure 1. View of the (−)-**5** cation in the X-ray structure of the hydrogen dibenzoyl-L-tartrate salt, with 30% probability ellipsoids.

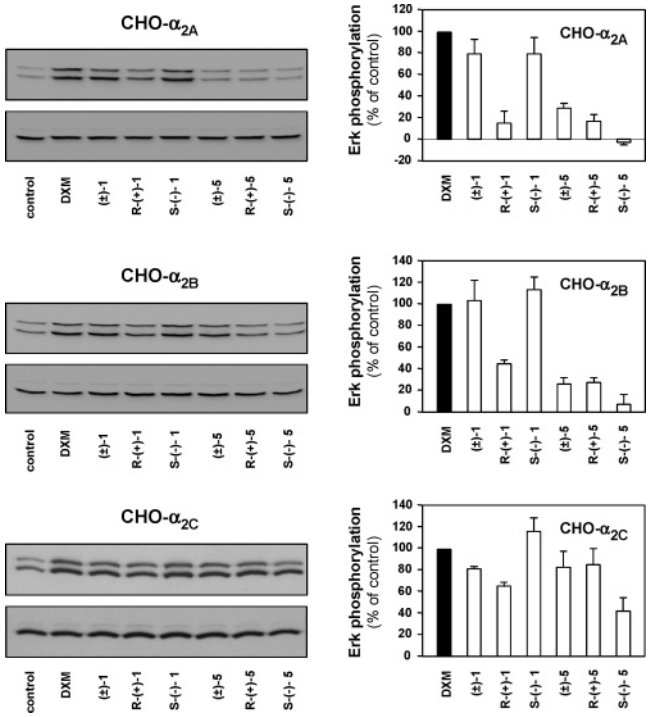


Figure 2. Effects of compounds on Erk phosphorylation. CHO cells expressing α_{2A}- (upper panels), α_{2B}- (middle panels), or α_{2C}-subtype (lower panels) were placed overnight in serum-free culture medium and then treated for 10 min with 10^{-6} M dexmedetomidine (DXM, taken as a positive control), (±)-**1**, (*R*)-(+)-**1**, (*S*)-(−)-**1**, (±)-**5**, (*R*)-(+)-**5**, and (*S*)-(−)-**5**. Phosphorylated Erk (P-Erk) and total Erk (Erk) were revealed by Western blotting with specific antibodies (left-hand panels). The effect of the different compounds on the extent of Erk phosphorylation in each CHO-α_2 cell line was quantified by densitometric analysis of the blots. Results are expressed as percent increase relative to dexmedetomidine (right-hand panels). Reported data are the mean ± SEM from four independent experiments.

(*S*)-(−)-**5** behaved as antagonists at α_{2A}- and α_{2B}-subtypes (pK_b α_{2A} of 7.11 and 7.35, respectively; pK_b α_{2B} of 6.07 and 5.93, respectively). A surprising reversal of α_{2C}-enantioselectivity was observed because the eutomers of *m*-nitrobiphenyline [(±)-**5**] and biphenyline [(±)-**1**], two structurally similar ligands, were (*R*)-(+)-**5** and (*S*)-(−)-**1**, respectively. Moreover, (*S*)-(−)-**1** proved considerably more active than its (*R*)-optical antipode at α_{2A}- and α_{2B}-subtypes. The higher potency shown by (*S*)-(−)-**1** confirmed a previous observation of ours.[2] In order to further delineate their agonist efficacy, (±)-**1**, (±)-**5**, and their corresponding enantiomeric forms were tested for their capacity to cause receptor-mediated Erk phosphorylation (Figure 2). Dexmedetomidine was used as reference agonist in these assays. The stimulation of the three receptor subtypes with (±)-**1** led to Erk activation up to an extent fairly similar to that for dexmedetomidine. Of note, whatever the considered subtype, the (*S*)-(−)-**1** was more efficient than (*R*)-(+)-**1**. Consistent with

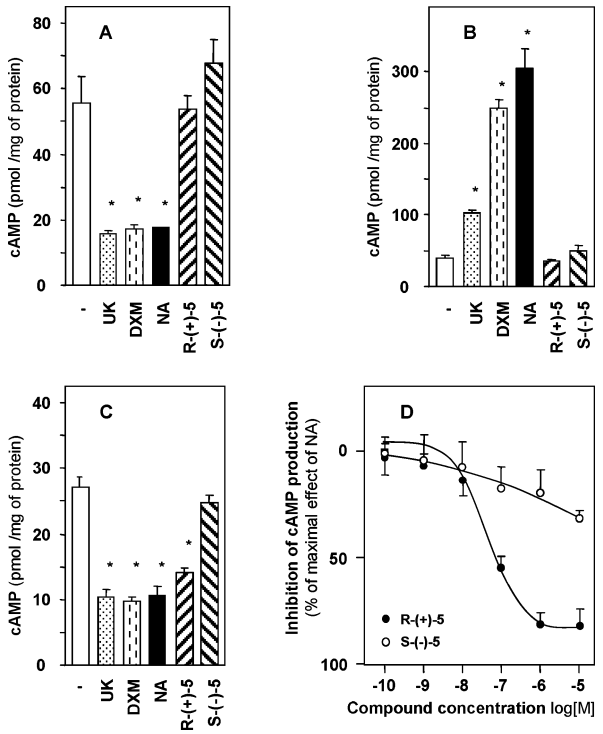
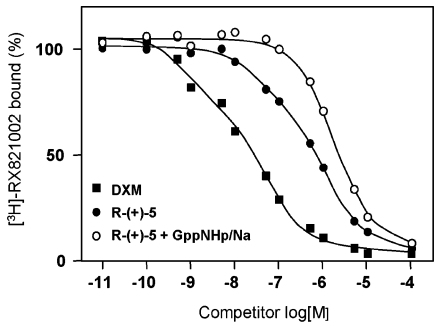


Figure 3. Effects of compounds on forskolin-induced cAMP accumulation. CHO cells expressing α_{2A}- (A), α_{2B}- (B), or α_{2C}-subtype (C) were incubated in Hepes-buffered DMEM containing 10 μM forskolin alone or in combination with 10 μM UK 14304 (UK), dexmedetomidine (DXM), (−)-noradrenaline (NA), (*R*)-(+)-**5**, or (*S*)-(−)-**5**, and cAMP was determined by RIA. Results are expressed in pmol/mg cellular protein. Reported data are the mean ± SEM from six measurements. (D) CHO cells expressing the α_{2C}-adrenoceptor were incubated in Hepes-buffered DMEM containing 10 μM forskolin and increasing concentrations of (*R*)-(+)-**5** or (*S*)-(−)-**5**. Inhibitory effect is expressed as percent of maximal effect of (−)-noradrenaline. Reported data are the mean ± SEM from three independent experiments.

results obtained with microphysiometry instrument, the racemic (±)-**5** had only marginal incidence on the Erk phosphorylation in cells expressing α_{2A}- or α_{2B}-AR but behaved as a full agonist on those expressing α_{2C}-subtype. As for (±)-**1**, the two enantiomers of (±)-**5** exhibited different efficacy, and in agreement with already noticed reversal of enantioselectivity, (*R*)-(+)-**5** was significantly more active than (*S*)-(−)-**5**.

The ability of (*R*)-(+)-**5** and (*S*)-(−)-**5** to modulate cAMP production was tested for each receptor subtype on forskolin-stimulated cells (Figure 3). The three reference compounds that were used in these experiments [UK 14304, dexmedetomidine, and (−)-noradrenaline] efficiently inhibited the forskolin-induced cAMP production in CHO expressing α_{2A}- or α_{2C}-subtype but significantly increased the cAMP level in CHO expressing the α_{2B}-subtype. Neither (*R*)-(+)-**5** nor (*S*)-(−)-**5** induced any significant modification of intracellular cAMP level in CHO expressing the α_{2A}- or α_{2B}-subtype. On the other hand, (*R*)-(+)-**5** but not (*S*)-(−)-**5** caused a significant decrease of cAMP accumulation in CHO expressing α_{2C}-subtype. In agreement with Erk-phosphorylation experiments, the inhibitory effect of (*R*)-(+)-**5** was slightly weaker than those of the full agonists and represented 80−85% of that of (−)-noradrenaline. The potency of the two enantiomers of (±)-**5** to inhibit cAMP production was examined by analyzing dose response curves (Figure 3). The weak effects of (*S*)-(−)-**5** precluded reliable estimation of its EC_{50} value. The EC_{50} of (*R*)-(+)-**5** was found to be 38 nM. This value is in agreement with results obtained with microphysiometry instrument; it also fit with the observa-



Competitor	- GppNHp/Na		+ GppNHp/Na
	pK_{iH}	pK_{iL}	pK_i
dexmedetomidine	10.04 ± 0.3	8.26 ± 0.07	7.51 ± 0.17
(*R*)-(+)-**5**	8.21 ± 0.21	6.42 ± 0.10	6.44 ± 0.05

Figure 4. Effect of GppNHp/NaCl on (*R*)-(+)-**5** binding to α_{2C}-subtype coupled to G-protein. Membranes from CHO cells expressing α_{2C}-subtype were incubated in Tris-Mg^{2+} buffer with 6 nM [^{3}H]RX 821002 and increasing concentrations of dexmedetomidine (square) or (*R*)-(+)-**5** (circle). Displacement experiments with (*R*)-(+)-**5** were carried out in the absence (dark symbols) or presence (open symbols) of 10 μM GppNHp plus 100 mM NaCl. Results are expressed as percent of maximum specific binding determined in the absence of competitor. $pK_i = -\log K_i$. K_i values were obtained from computer-assisted analysis. Reported data are the mean ± SEM from three separate experiments.

tion that 10 nM (*R*)-(+)-**5** caused a clear increase of Erk phosphorylation in CHO expressing the α_{2C}-AR subtype (data not shown). Since (*R*)-(+)-**5** behaved as an almost full agonist of α_{2C}-subtype, its binding parameters were re-examined in experimental conditions allowing receptor coupling to the G-protein (i.e., in binding buffer containing 5 mM Mg^{2+}). Inhibition curves and pK_i values derived from computer-assisted analysis of the data from these experiments are presented in Figure 4. As expected, displacement of [^{3}H]RX 821002 binding to α_{2C}-subtype by dexmedetomidine gave an inhibition curve with a Hill coefficient value significantly lower than 1 and fitted a two-site component model well. The pK_i values of dexmedetomidine for the high (pK_{iH}) and low (pK_{iL}) affinity sites (G-protein-coupled and G-protein-uncoupled receptors, respectively) were found to be 10.04 and 8.26. Although with lower potency than dexmedetomidine, (*R*)-(+)-**5** also yielded a two-component inhibition curve whose pK_{iH} and pK_{iL} were 8.21 and 6.42, respectively. To verify that these two values truly reflected binding of (*R*)-(+)-**5** to coupled and uncoupled α_{2C}-subtype, another series of inhibition experiments was performed in the presence of GppNHp/Na. As shown in Figure 4, the addition of 10 μM GppNHp and 100 mM NaCl into the incubation buffer resulted in a rightward shift and in an increase in the slope of the inhibition curve, reflecting the conversion of the whole receptor population into a low affinity state. The pK_i of (*R*)-(+)-**5** was found to be 6.44 in these conditions, and such a value matches well that obtained in experiments carried out in absence of Mg^{2+}.

In conclusion, the present study demonstrated the important role played by chirality in α_{2C}-AR activation, since the good α_{2C}-agonist properties of racemic *m*-nitrobiphenyline (±)-**5** and biphenyiline (±)-**1** were mainly produced, or in some cases even enhanced, by the enantiomeric form (*R*)-(+)-**5** or (*S*)-(−)-**1**, respectively. The surprising reversal of enantioselectivity, observed for these similar compounds, suggests that minor structural modifications on ligands belonging to the same series can produce changes in their binding mode.[12] In particular, we highlighted the efficient and α_{2C}-selective agonism of (*R*)-(+)-*m*-nitrobiphenyline, which might therefore be considered a promising new tool for characterizing the functions mediated by the α_{2C}-subtype, which still remains partially enigmatic.

Experimental Section

(*R*)-2-[1-(3′-Nitrobiphenyl-2-yloxy)ethyl]-4,5-dihydro-1*H*-imidazole [(*R*)-(+)-5]. Method A. A solution of ethylenediamine (1.29 mL, 19.30 mmol) in dry toluene (6.7 mL) was added dropwise to a mechanically stirred solution of 2 M trimethylaluminum (9.6 mL, 19.30 mmol) in dry toluene (16 mL) at 0 °C under nitrogen atmosphere. After 1 h, the solution was cooled to 0 °C and a solution of (*R*)-(−)-**6** (2.87 g, 9.64 mmol) in dry toluene (11.5 mL) was added dropwise. The mixture was heated to 70 °C for 3 h, cooled to 0 °C, and quenched cautiously with MeOH (4.5 mL), followed by H_2O (1 mL). After addition of $CHCl_3$ (35 mL) and filtration, the organic layer was extracted with 2 N HCl. The aqueous layer, made basic with 10% NaOH, was extracted with $CHCl_3$. Removal of dried solvent gave the free base (*R*)-(+)-**5**, which was purified by flash chromatography using cyclohexane/AcOEt/MeOH/33% NH_4OH (6:3:1:0.1) (1.92 g, 64% yield). The enantiomeric purity, determined by ^{1}H NMR (CD_3OD) on its corresponding diastereomeric hydrogen dibenzoyl-D-tartrate salt, was about 86%. After dry EtOH fractional crystallization of this salt, the enantiomeric purity of (*R*)-(+)-**5**, determined by HPLC and ^{1}H NMR on the corresponding ureidic derivative **7a**, was >98% (80% yield). (*R*)-(+)-**5** oxalate salt: $[\alpha]_{20}^{D}$ +24.57 (*c* 1, MeOH); mp 209−210 °C; ^{1}H NMR (DMSO) δ 1.53 (d, 3, CH_3), 3.88 (s, 4, NCH_2CH_2N), 5.38 (q, 1, CH), 7.04−7.47 (m, 8, ArH), 9.41 (br s, 1, NH, exchangeable with D_2O). Anal. ($C_{17}H_{17}N_3O_3 \cdot H_2C_2O_4$) C, H, N.

Similarly, (*S*)-(−)-**5** was obtained from ester (*S*)-(+)-**6** and further purification by fractional crystallization of its hydrogen dibenzoyl-L-tartrate salt (51% yield). (*S*)-(−)-**5** oxalate salt: $[\alpha]_{20}^{D}$ −23.62 (*c* 1, MeOH); mp 209−210 °C. Anal. ($C_{17}H_{17}N_3O_3 \cdot H_2C_2O_4$) C, H, N.

Method B. HCl was bubbled through a stirred and cooled (0 °C) solution of (*R*)-(+)-**9** (0.77 g, 2.88 mmol) and MeOH (0.24 mL, 5.76 mmol) in dry $CHCl_3$ (5 mL) for 45 min. After 12 h at 0 °C, dry ether was added to the mixture to give the intermediate imidate, which was filtered. This solid (0.168 g, 0.50 mmol) was added to a cooled (0 °C) and stirred solution of ethylenediamine (0.041 mL, 0.62 mmol) in absolute EtOH (2.4 mL). After 1 h, concentrated HCl (a few drops) in absolute EtOH (1.2 mL) was added to the mixture, which was stored overnight in the refrigerator. It was then diluted with absolute EtOH (2 mL) and heated at 75 °C for 5 h. After the mixture was cooled, the solid was collected and discarded and the filtrate was concentrated and filtered again. The filtrate was evaporated to dryness to give a residue, which was purified by flash chromatography using cyclohexane/AcOEt/MeOH/33% NH_4OH (6:3:1:0.1) as eluent to give 0.27 g (30% yield) of (*R*)-(+)-**5**: enantiomeric purity, >98%.

(*R*)-2-(3′-Nitrobiphenyl-2-yloxy)propionic Acid Methyl Ester [(*R*)-(−)-6]. A solution of DEAD (0.99 g, 4.91 mmol) in dry THF was added dropwise to a mixture of methyl (*S*)-(−)-lactate (0.42 g, 4.043 mmol), 3′-nitrobiphenyl-2-ol (0.87 g, 4.043 mmol), and triphenylphosphine (1.06 g, 4.043 mmol) in THF. The mixture was stirred at room temperature under nitrogen atmosphere overnight. The THF was evaporated and a diethyl ether/hexane mixture was added to precipitate triphenylphosphine oxide, which was filtered off. Removal of the solvent gave a residue that was purified by flash chromatography using cyclohexane/AcOEt (95:5) (1.026 g, 83% yield): $[\alpha]_{20}^{D}$ −20.78 (*c* 1, MeOH); ^{1}H NMR ($CDCl_3$) δ 1.53 (d, 3, $CHCH_3$), 3.72 (s, 3, OCH_3), 4.82 (q, 1, CH), 6.82−8.52 (m, 8, ArH).

Similarly, (*S*)-(+)-**6** was obtained starting from methyl (*R*)-(+)-lactate (85% yield): $[\alpha]_{20}^{D}$ +19.51 (*c* 1, MeOH).

(*R*)-2-[1-(3′-Nitrobiphenyl-2-yloxy)ethyl]-4,5-dihydroimidazole-1-carboxylic Acid [(*R*)-1-Phenylethyl]amide (7a). (*R*)-(+)-α-Methylbenzyl isocyanate (0.14 g, 0.95 mmol) was added to a solution of (*R*)-(+)-**5** (0.30 g, 0.95 mmol) in dry CH_2Cl_2. After 4

h at room temperature, the solvent was removed and the residue was purified by flash chromatography using cyclohexane/AcOEt/33% NH_4OH (5:5:0.1). ^1H NMR ($CDCl_3$) δ 1.39 (d, 3, $CH_3CHC{=}N$), 1.51 (d, 3, CH_3CHN), 3.82 (m, 4, NCH_2CH_2N), 5.01 (m, 1, CHN), 5.28 (d, 1, NH, exchangeable with D_2O), 5.98 (q, 1, $CHC{=}N$), 6.95−8.44 (m, 13, ArH). Anal. ($C_{26}H_{26}N_4O_4$) C, H, N.

Similarly, **7b** was obtained from (*S*)-(−)-**5.** ^1H NMR ($CDCl_3$) δ 1.40 (d, 3, $CH_3CHC{=}N$), 1.53 (d, 3, CH_3CHN), 3.82 (m, 4, NCH_2CH_2N), 5.01 (m, 1, CHN), 5.28 (d, 1, NH, exchangeable with D_2O), 5.98 (q, 1, $CHC{=}N$), 6.95−8.44 (m, 13, ArH). Anal. ($C_{26}H_{26}N_4O_4$) C, H, N.

(*R*)-2-(3′-Nitrobiphenyl-2-yloxy)propionamide [(*R*)-(−)-8]. A solution of 3′-nitrobiphenyl-2-ol (1.5 g, 7 mmol) in 2-butanone (5 mL) was added to a solution of potassium methoxide (1.0 g, 14 mmol) in 2-butanone (4 mL) and stirred for 1 h at 40 °C. (*S*)-2-[(*p*-Tolylsulfonyl)oxy]propionamide (3.9 g, 16 mmol) in 2-butanone (2 mL) was added dropwise, and the mixture was stirred at 70 °C for 24 h. The mixture was cooled and filtered. The solution was diluted with 5% Na_2CO_3 solution and extracted with EtOAc. Removal of dried solvent gave (*R*)-(−)-**8** (1.48 g, 70% yield): mp 167−168 °C, $[\alpha]_{20}^{D}$ −16.49 (*c* 1, MeOH); ^1H NMR ($CDCl_3$) δ 1.58 (d, 3, CH_3), 4.88 (q, 1, CH), 6.31 (br s, 1, NH, exchangeable with D_2O), 6.93−8.43 (m, 8, ArH). Anal. ($C_{15}H_{14}N_2O_4$) C, H, N.

(*R*)-2-(3′-Nitrobiphenyl-2-yloxy)propionitrile [(*R*)-(+)-9]. To a stirred solution of (*R*)-(−)-**8** (1.0 g, 3.49 mmol) in dry dioxane and pyridine (0.55 g, 7 mmol), trifluoroacetic anhydride (0.78 g, 3.7 mmol) was added dropwise at room temperature. The mixture was stirred for 5 h, and AcOEt was added. The organic layer, washed with H_2O and dried over Na_2SO_4, was evaporated and the residue was purified by flash chromatography using cyclohexane/AcOEt (8:2) as eluent to give (*R*)-(+)-**9** (0.66 g, yield 70%): $[\alpha]_{20}^{D}$ +38.06 (*c* 1, MeOH); ^1H NMR ($CDCl_3$) δ 1.72 (d, 3, CH_3), 4.83 (q, 1, CH), 7.20−8.43 (m, 8, ArH).

Supporting Information Available: Chemical methodology, biological experiments, crystallographic data for (−)-**5** as the hydrogen dibenzoyl-L-tartrate salt, and elemental analysis results. This material is available free of charge via the Internet at http://pubs.acs.org.

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