1-Phenyl-3-azabicyclo[3.1.0]hexane derivatives as new ligands for sigma receptors

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Dedicated to Professor Vincenzo Tortorella in the occasion of his “Fuori Ruolo” status
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Abstract
A series of 1-phenyl-3-azabicyclo[3.1.0]hexanes were synthesized as more conformationally restricted prototypical σ ligands 3-phenylpiperidines with the aim to developing new σ ligands. Compared with 3-phenylpiperidines reported by Largent et al., binding data showed that conformational restriction was not detrimental for σ receptor affinity. Specifically, except for secondary amine 4, all racemic 1-phenyl-3-azabicyclo[3.1.0]hexane derivatives (12-19) showed moderate to high affinity for both σ₁ and σ₂ receptors. Dextrorotatory isomers with the same configuration of 3-phenylpiperidines to C-1 carbon linked to the phenyl ring showed a better affinity and selectivity for σ₁ receptors compared to the respective levorotatory isomers. Compounds (+)-14 and (+)-15 displayed very high affinity for σ₁ (Ki = 0.9 and 2.3 nM respectively) but low selectivity for receptor subtypes. Compound (+)-18 with N-phenethyl substituent embodies the highest selectivity for σ₁ receptors.

Keywords: 1-Phenyl-3-azabicyclo[3.1.0]hexane, sigma receptor, 3-phenylpiperidines

Introduction

Sigma (σ) receptors are typical binding sites interacting with several psychoactive drugs including haloperidol, benzomorphans and phencyclidine.1-5 The σ₁ subtype exhibits high affinity for (+)-benzomorphans such as (+)-pentazocine and (+)-N-allylnormetazocine (SKF-10,047) and a reduced affinity for the respective (–)-enantiomers.

Based on animal model studies, this subtype seems to be involved in cocaine induced behavioral changes, in opiate induced analgesia, steroid-induced mental disturbances and alterations in immune functions.6-8
The $\sigma_2$ subtype showed low affinity for (+)-pentazocine and (+)-SKF-10,047 and (–)-isomers did not differentiate between the two sites.

Several pharmacological studies showed that $\sigma_2$ receptors are expressed in high concentration in tumor cell lines and that they are involved in proliferation and cell viability. Thus, selective $\sigma_1$ and $\sigma_2$ ligands with agonist or antagonist properties might be potential drugs for clinical treatment of memory and learning disorders, psychoses, cocaine abuse, dyskinesia induced by classical antipsychotic therapy and cancer.

Recently, we have focused on the synthesis of substituted 1-phenyl-2-cyclopropylmethylylamines as tools capable to providing new selective $\sigma_1$ and $\sigma_2$ ligands. Taking into account these data and the synthetic opportunity to extend conformational restriction to prototypical $\sigma$ ligands 3-phenylpiperidines we synthesized a series of 1-phenyl-3-azabicyclo[3.1.0]hexanes as possible new ligands for $\sigma_1$ and $\sigma_2$ receptors. Using a different synthetic approach, 1-phenyl-3-azabicyclo[3.1.0]hexanes were previously reported by Fanshawe et al. as non-narcotic analgesic compounds and as agents for treating depression and chemical dependencies. Although some of these effects might be related to a possible interaction with $\sigma$ receptors, to date no affinity binding data have been reported about these compounds.

In these studies we report our strategy to synthesize 1-phenyl-3-azabicyclo[3.1.0]hexane derivatives and structure affinity relationships for $\sigma_1$ and $\sigma_2$ receptors.

Figure 1. 3-Phenylpiperidines and conformational restricted 1-phenyl-3-azabicyclo[3.1.0]-hexanes.

Figure 2. Retrosynthetic analysis for synthesis of 1-phenyl-3-azabicyclo[3.1.0]hexanes.
Results and Discussion

Chemistry

Racemic methyl 2-(bromomethyl)-1-phenylcyclopropanecarboxylate (±)-1 and its enantiomers (+)- and (–)-1 were synthesized according to a previously reported procedure. Treatment of 1 with NaN₃ in DMF gave azido intermediate 2 (Scheme 1). Subsequently, reduction of 2 with sodium hydrogentelluride (NaTeH) in ethanol and internal cyclization provided a good yield of lactam 3. The NaTeH was readily prepared from tellurium and NaBH₄ as reported by Barton and McCombie. Treatment of 3 with diborane in anhydrous THF gave 1-phenyl-3-azabicyclo[3.1.0]hexane 4.

The final compound 12 was prepared by alkylation of 4 with commercially available 1-bromo-3-methylbut-2-ene. Intermediates 5-11 were synthesized by treatment of commercially available amines with methyl bromoester 1. Compounds 13-20 were obtained by reduction with diborane in anhydrous THF of the respective lactams (5-11). All structures of synthesized compounds were fully consistent with ¹H-NMR and ¹³C-NMR spectral data.

Scheme 1. a: NaN₃, DMF, 30 °C, 4 h; b: Te/NaBH₄, C₂H₅OH; c: B₂H₆/THF, reflux, 12 h; d: BrCH₂CH=C(CH₃)₂/NaHCO₃/DMF, 80 °C, 12 h; e: RNH₂/CH₃OH, reflux, 6 h; f: B₂H₆/THF, reflux, 12 h.

The σ receptor affinities of racemic 1-phenyl-3-azabicyclo[3.1.0]hexanes 4 and 12-19 are reported in Table 1. These data show that secondary amine 4 did not interact significantly with σ₁ and σ₂ receptors (Ki > 10,000). However, nitrogen substitution with propyl or 2-methyl-2-butene led to compounds (13 and 12 respectively) able to interact with σ receptors. Specifically,
compound 12 showed a good affinity with a slight preference for $\sigma_1$ receptor subtypes. $N$-cyclohexyl substitution gave compound 14 with the highest affinity in the racemic series.

Table 1. $\sigma_1$ and $\sigma_2$ binding affinities [$K_i \pm \text{SEM (nM)}$]

<table>
<thead>
<tr>
<th>Compound</th>
<th>R</th>
<th>$[^3\text{H}]$Pentaz $\sigma_1$</th>
<th>$[^3\text{H}]$DTG $\sigma_2$</th>
</tr>
</thead>
<tbody>
<tr>
<td>(±)-4</td>
<td>$-\text{H}$</td>
<td>$&gt;10,000$</td>
<td>$&gt;10,000$</td>
</tr>
<tr>
<td>(±)-12</td>
<td>$-\text{CH}_3$</td>
<td>$12 \pm 1$</td>
<td>$24 \pm 3$</td>
</tr>
<tr>
<td>(±)-13</td>
<td>$-\text{CH}_3$</td>
<td>$84 \pm 7$</td>
<td>$336 \pm 23$</td>
</tr>
<tr>
<td>(±)-14</td>
<td>$-\text{cyclohexyl}$</td>
<td>$2.1 \pm 0.2$</td>
<td>$4.3 \pm 0.3$</td>
</tr>
<tr>
<td>(±)-15</td>
<td>$-\text{cyclohexyl}$</td>
<td>$12.4 \pm 0.8$</td>
<td>$141 \pm 4$</td>
</tr>
<tr>
<td>(±)-16</td>
<td>$-\text{cyclohexyl}$</td>
<td>$369 \pm 31$</td>
<td>$461 \pm 36$</td>
</tr>
<tr>
<td>(±)-17</td>
<td>$-\text{cyclohexyl}$</td>
<td>$261 \pm 14$</td>
<td>$514 \pm 27$</td>
</tr>
<tr>
<td>(±)-18</td>
<td>$-\text{cyclohexyl}$</td>
<td>$88.3 \pm 6$</td>
<td>$910 \pm 35$</td>
</tr>
<tr>
<td>(±)-19</td>
<td>$-\text{cyclohexyl}$</td>
<td>$191 \pm 14$</td>
<td>$103 \pm 12$</td>
</tr>
</tbody>
</table>
These results were similar to the N-substitution on 3-phenylpiperidine, octahydrobenzo[f]quinoline and cis-benzomorphan derivatives\textsuperscript{12,18} which revealed an increase in binding affinity when nitrogen of the secondary amines was substituted with bulk alkyl substituents.

Compared to 14, introduction of a methylene spacer between the nitrogen and cyclohexyl residue (15) decreased by six time the binding affinity for $\sigma_1$ receptors and to a greater extent for $\sigma_2$. The substitution of cyclohexane of 15 with more bulk adamantane (16) induced a strong reduction of $\sigma_1$ and $\sigma_2$ receptor affinity.

The benzyl derivative 17 revealed a lower $\sigma$ binding affinity compared with the respective cycloalkyl 15. Moreover, an increase of the methylenic chain in phenylethyl and phenylpropyl derivatives (18 and 19 respectively) induced different effects on affinity and selectivity. In particular, 18 showed an improved affinity and selectivity for $\sigma_1$ subtypes, whereas compound 19 displayed a slight preference for $\sigma_2$ receptors.

Considering the better binding profile of racemic compounds 14, 15, 18 and 19 we also evaluated the affinity of the respective enantiomers (Table 2). The binding affinity data showed that (+)-(1R,5S)-isomers have higher affinity for the $\sigma_1$ receptor compared with (–)-(1S,5R)-isomers. Specifically, (+)-(1R,5S)-14 displayed a slight improvement of affinity and selectivity for $\sigma_1$ receptors compared with the respective (–)-(1S,5R)-isomer which showed a little preference for $\sigma_2$ subtypes.

Enantiomers (+)-(1R,5S)- and (–)-(1S,5R)-15 increased their binding affinity for both $\sigma_1$ and $\sigma_2$ subtypes and thus with no substantial increase of selectivity with respect to the racemic mixture.

Conversely to these results, an improved selectivity for $\sigma_1$ receptors was obtained with N-phenethyl derivative (+)-(1R,5S)-18 which had the highest selectivity of the series. The respective isomer (–)-(1S,5R)-18 showing the same decrease of affinity for both $\sigma_1$ and $\sigma_2$ receptors did not provide significant results compared with racemic mixture.

The preference of (±)-19 for $\sigma_2$ receptors was not confirmed in the respective enantiomers. In particular (+)-(1R,5S)- and (–)-(1S,5R)-19 increased both $\sigma_1$ and $\sigma_2$ receptor affinity with a reversed selectivity compared to (±)-19. Moreover, the evaluation of binding affinity of N-phenethyl derivative (+)-(1R,5S)- and (–)-(1S,5R)-18 and phenylpropyl (+)-(1R,5S)- and (–)-(1S,5R)-19, provided evidence that the increase of methylenic chain spacer was more critical for $\sigma_2$ with respect to $\sigma_1$ receptors.

In conclusion, in these paper we present novel 1-phenyl-3-azabicyclo[3.1.0]hexane derivatives capable of interacting with moderate to high affinity with sigma receptors. Conformational restriction of phenyl-3-azabicyclo[3.1.0]hexane derivatives compared with 3-phenylpiperidine did not produce compounds with high selectivity for $\sigma$ receptor subtypes in this series but like (+)- and (–)-3-PPP (3-phenylpropylpiperidine) only a slight preference for $\sigma_1$ receptors. However, the very high affinity of compounds (+)-(1R,5S)-14 and (+)-(1S,5R)-15 gave a good starting point to design new potential $\sigma_1$ and $\sigma_2$ selective ligands.
Table 2. \( \sigma_1 \) and \( \sigma_2 \) binding affinities \([K_i \pm SEM (nM)]\)

<table>
<thead>
<tr>
<th>Compound</th>
<th>R</th>
<th>(^{3}\text{H})\text{(+)}\text{Pentaz} \sigma_1</th>
<th>(^{3}\text{H})\text{DTG} \sigma_2</th>
</tr>
</thead>
<tbody>
<tr>
<td>(+)-(1R,5S)-14</td>
<td></td>
<td>0.91 ± 0.2</td>
<td>5.8 ± 1</td>
</tr>
<tr>
<td>(−)-(1S,5R)-14</td>
<td></td>
<td>14.6 ± 1.3</td>
<td>9.6 ± 0.9</td>
</tr>
<tr>
<td>(+)-(1R,5S)-15</td>
<td></td>
<td>2.3 ± 0.4</td>
<td>33.8 ± 1.2</td>
</tr>
<tr>
<td>(−)-(1S,5R)-15</td>
<td></td>
<td>9.3 ± 1.7</td>
<td>46.6 ± 7</td>
</tr>
<tr>
<td>(+)-(1R,5S)-18</td>
<td></td>
<td>55 ± 11</td>
<td>1135 ± 75</td>
</tr>
<tr>
<td>(−)-(1S,5R)-18</td>
<td></td>
<td>164 ± 8</td>
<td>1765 ± 94</td>
</tr>
<tr>
<td>(+)-(1R,5S)19</td>
<td></td>
<td>16 ± 0.8</td>
<td>66 ± 16</td>
</tr>
<tr>
<td>(−)-(1S,5R)-19</td>
<td></td>
<td>48 ± 1.7</td>
<td>63 ± 4</td>
</tr>
</tbody>
</table>

Experimental Section

General Procedures. Reagents for organic synthesis were purchased from Aldrich & Sigma Chemicals Co. NMR spectra were recorded on a Varian Inova 200 spectrometer with TMS as
an internal standard. Thin-layer chromatography (TLC) was performed on precoated silica gel
60 F254 aluminum sheets (Merck); with visualization under UV light and in a iodine chamber.
Melting points were determined in open capillary tubes on a Büchi melting point apparatus
and are uncorrected. Infrared spectra (IR) were obtained using a 1600 FT-IR Perkin-Elmer
spectrophotometer. All optical rotations were determined in CH3OH solution (C = 1)
employing an Optical Activity Ltd. automatic polarimeter type AA-10. Elemental analyses (C,
H, N) were done on a Carlo Erba Model 1106 elemental analyzer.

Methyl 2-(azidomethyl)-1-phenyl-cyclopropanecarboxylate (2). To a solution of methyl
2-(bromomethyl)-1-phenylcyclopropanecarboxylate (±)-1 (200 mg, 0.74 mmol) in DMF NaN3 at
0 °C was added (57.9 mg, 0.89 mmol) and the whole was stirred for 4 h. Water was added and
the resulting mixture was evaporated under reduced pressure. The residue was partitioned
between brine and AcOEt and the organic phase was dried (Na2SO4) and evaporated to give, as
an oil, 171 mg (theoretical yield) of azide 2.

The handling of NaN3 and derivatives might be dangerous. For this reason, the reaction
was repeated several time on a reduced scale in order to reduce the risk of explosion.

IR (KBr): 1718 (C=O); 1H NMR (CDCl3) δ: 1.45 (dd, 1H J = 4.6, 9.0 Hz), 1.72 (dd, 1H, J =
4.6, 7.2 Hz), 1.83-1.93 (m, 1H), 3.62 (dd, 1H, J = 10.0, 13.0 Hz), 3.65 (s, 3H), 3.68 (dd, 1H, J =
4.4, 13.0 Hz), 7.25-7.40 (m, 5H). 13C NMR (CDCl3) δ: 21.85, 33.57, 36.36, 51.95, 52.71,
125.30, 127.32, 128.65, 138.71, 172.61.

(±)-1-Phenyl-3-azabicyclo[3.1.0]hexan-2-one (3). A mixture of powdered tellurium (2.41 g,
18.58 mmol), NaBH4 (1.69 g, 44.59 mmol) and ethanol (80 ml) was heated under reflux in a
nitrogen atmosphere until the tellurium disappeared. After cooling to room temperature, a
solution of azide 2 (3.42 g, 14.86 mmol) in ether (75 ml) was added to the dark red solution of
sodium hydrogen telluride. The color turned black and after nitrogen evolution and precipitation
of metallic tellurium, the mixture was left open to air with stirring for 12 h. The mixture was
filtered through celite, heated to 60 °C and evaporated under reduced pressure to provide 1.7 g of
lactam 3, as a white solid.

(Yield 65.7%), m. p. 70-72 °C; IR (KBr): 1668 (C=O); 1H NMR (CDCl3) δ: 1.06 (dd, 1H J =
4.0, 4.6 Hz), 1.45 (dd, 1H, J = 4.6, 8.0 Hz), 2.25-2.20 (m, 1H), 3.25 (d, 1H, J = 10.4 Hz), 3.50
(dd, 1H, J = 5.6, 10.4 Hz), 7.15-7.42 (m, 5H), 7.64 (s, 1H). 13C NMR (CDCl3) δ: 18.49,
22.15, 33.20, 42.75, 126.44, 127.76, 128.22, 135.64, 178.27.

Anal. Calcd. for C11H11NO: C, 76.28; H, 6.40; N, 8.09. Found: C, 76.00; H, 6.45; N, 8.10.

(±)-1-Phenyl-3-azabicyclo[3.1.0]hexan-2-one (4). To a solution (1 M in THF) of 40 ml of diborane
cooled to 0 °C and under nitrogen atmosphere, a solution of lactam 3 (1.7 g, 9.81 mmol) in
anhydrous THF (5ml) was slowly added. The mixture was heated to reflux for 8 h and
subsequently permitted to cool to room temperature. Twelve ml of a 6 M hydrochloric acid
solution was slowly added through a dropping funnel. THF was removed by distillation at
atmospheric pressure and the mixture was basified with NaOH 2 M. The latter was extracted three times with a total of 100 ml of CHCl₃. The organic extract was dried with anhydrous Na₂SO₄ and evaporated under reduced pressure to give the crude base 4 which were purified on silica gel by flash chromatography using CHCl₃/cyclohexane/EtOH (50:48:2) as eluent. The purified compound 4 (1.4 g, yield 90%) was dissolved in ether and treated with an ether solution of oxalic acid to give the oxalate salts as a white solid. The analytically pure samples were obtained by recrystallization (EtOH/ether).

m. p. 180-182 °C; ¹H NMR (DMSO-d₆) δ: 1.08 (dd, 1H, J = 5.4, 6.0 Hz), 1.55 (dd, 1H, J = 5.0, 5.4 Hz), 2.08-2.20 (m, 1H), 3.30-3.54 (m, 3H), 3.68 (d, 1H, 11.2 Hz), 7.15-7.42 (m, 5H), 8.00 (s, broad, 3H). ¹³C NMR (DMSO-d₆) δ: 16.17, 23.85, 30.94, 47.30, 49.82, 127.04, 127.11, 129.00, 140.06, 165.45.

Anal. Calcd. for C₁₁H₁₃N: C, 82.97; H, 8.23; N, 8.80. Found: C, 82.87; H, 8.34; N, 8.85.

(±)-1-Phenyl-3-propyl-3-azabicyclo[3.1.0]hexan-2-one (5). A mixture of 2-(bromomethyl)-1-phenylcyclopropanecarboxylate (±)-1 (400 mg, 1.48 mmol), 2 ml of propylamine (1.44 g, 24.3 mmol) and 2 ml of toluene was heated at 80 °C in a sealed reaction vessel for 6 h. After the recovery of the reaction mixture, the solvent was evaporated under reduced pressure and the residue was dissolved in CHCl₃ and washed with a solution of 2 N HCl and subsequently dried over anhydrous Na₂SO₄. In vacuo evaporation of chloroform solution gave a crude product that was purified by flash chromatography using cyclohexane/ethyl acetate (8:2) as eluent.

(Yield 85 %); m. p. 73-75 °C; IR (KBr): 1671 (C=O); ¹H NMR (CDCl₃) δ: 0.88 (t, 3H, J = 7.4), 1.45 (dd, 1H, J = 6.0, 7.0 Hz), 1.38-1.70 (m, 3H), 2.06-2.19 (m, 1H), 3.25 (t, 2H, J = 7.8), 3.40 (d, 1H, J = 10.4 Hz), 7.22-7.47 (m, 5H). ¹³C NMR (CDCl₃) δ: 11.10, 16.80, 19.06, 23.68, 32.27, 50.38, 53.27, 125.42, 126.47, 128.77, 137.75, 174.27.

Anal. Calcd. for C₁₄H₁₇NO: C, 78.10; H, 7.96; N, 6.51. Found: C, 78.30; H, 8.00; N, 6.53.

The compounds 6-11 were prepared using the above procedure.

(±)-3-Cyclohexyl-1-phenyl-3-azabicyclo[3.1.0]hexan-2-one (6). (Yield 80 %); M. p. 90-92 °C; IR (KBr): 1672 (C=O); ¹H NMR (CDCl₃) δ: 1.06-1.80 (m, 10H) 2.07-2.12 (m, 3H), 3.03-3.23 (m, 1H), 3.93 (dd, 1H, J = 6.1, 10.6 Hz), 4.09 (d, 1H, J = 10.6 Hz), 7.18-7.45 (m, 5H). ¹³C NMR (CDCl₃) δ: 22.57, 23.16, 23.30, 24.32, 27.91, 28.78, 52.63, 60.31, 124.38, 126.33, 128.78, 138.24, 177.10. Anal. Calcd. for C₁₇H₂₁NO: C, 79.96; H, 8.29; N, 5.49. Found: C, 79.88; H, 8.48; N, 5.47.

(+)-(1R,5S)-3-Cyclohexyl-1-phenyl-3-azabicyclo[3.1.0]hexan-2-one (6). (Yield 75 %); M. p. 90-92 °C; [α]D²⁰ + 56.4°; IR, ¹H NMR and ¹³C NMR are identical to those of the racemate.

Anal. Calcd. for C₁₇H₂₁NO: C, 79.96; H, 8.29; N, 5.49. Found: C, 79.90; H, 8.20; N, 5.43.

(–)-(1S,5R)-3-Cyclohexyl-1-phenyl-3-azabicyclo[3.1.0]hexan-2-one (6). (Yield 78 %); M. p. 90-92 °C; [α]D²⁰ - 58.5°; IR, ¹H NMR and ¹³C NMR are identical to those of the racemate. Anal. Calcd. for C₁₇H₂₁NO: C, 79.96; H, 8.29; N, 5.49. Found: C, 80.00; H, 8.45; N, 5.50.

(±)-3-(Cyclohexylmethyl)-1-phenyl-3-azabicyclo[3.1.0]hexan-2-one (7). (Yield 87 %); M. p. 94-96 °C; IR (KBr): 1684 (C=O); ¹H NMR (CDCl₃) δ: 0.80-1.38 (m, 7H), 1.48 (dd, 1H, J = 4.6, 7.8 Hz), 1.57-1.85 (m, 5H), 2.13 (m, 1H), 3.01(dd, 1H, J = 6.8, 16.4 Hz), 3.13 (dd, 1H, J = 7.2,
16.4 Hz), 3.35 (d, 1H, J = 10.4 Hz), 3.61 (dd, 1H, J = 5.6, 10.3 Hz), 7.18-7.48 (m, 5H). 13C NMR (CDCl3) δ: 23.44, 24.33, 24.85, 27.31, 29.91, 30.42, 37.21, 53.32, 54.01, 123.15, 126.11, 129.32, 136.42, 180.22. Anal. Calcd. for C18H23NO: C, 80.26; H, 8.61; N, 5.20. Found: C, 80.00; H, 8.50; N, 5.22.

(+)-(1R,5S)-3-(Cyclohexylmethyl)-1-phenyl-3-azabicyclo[3.1.0]hexan-2-one (7). (Yield 85 %); M. p. 94-96 °C; [α]D 20 + 65.2°; IR, 1H NMR and 13C NMR are identical to those of the racemate.


(–)-(1S,5R)-3-(Cyclohexylmethyl)-1-phenyl-3-azabicyclo[3.1.0]hexan-2-one (7). (Yield 90 %); M. p. 94-96 °C; [α]D 20 – 63.3°; IR, 1H NMR and 13C NMR are identical to those of the racemate.


(±)-3-(1-Adamantylmethyl)-1-phenyl-3-azabicyclo[3.1.0]hexan-2-one (8). (Yield 51 %); M. p. 138-140 °C; IR (KBr): 1676 (C=O); 1H NMR (CDCl3) δ: 1.03 (dd, 1H, J = 5.8, 7.4 Hz) 1.45-1.95 (m, 12H), 1.97-2.05 (m, 4H), 2.10 (m, 1H), 2.80 (s, 2H), 3.51 (d, 1H, J = , 10.3 Hz) 3.73 (dd, 1H, J = 5.7, 10.3 Hz), 7.15-7.50 (m, 5H). 13C NMR (CDCl3) δ: 22.99, 23.77, 28.41, 35.33, 37.01, 39.42, 42.21, 53.95, 56.97, 122.45, 126.71, 128.42, 136.77, 182.45. Anal. Calcd. for C22H27NO: C, 82.20; H, 8.47; N, 4.36. Found: C, 82.24; H, 8.40; N, 4.30.

(±)-3-Benzyl-1-phenyl-3-azabicyclo[3.1.0]hexan-2-one (9). (Yield 92 %); M. p. 86-88 °C; IR (KBr): 1674 (C=O); 1H NMR (CDCl3) δ: 1.00 (dd, 1H J = 5.4, 8.0 Hz), 1.48 (dd, 1H J = 4.8, 5.4, Hz) 1.98-2.18 (m, 1H), 3.30 (d, 1H, J = 10.4 Hz), 3.98 (dd, 1H, J = 5.9, 10.2 Hz), 4.37 (s, 2H), 7.12-7.60 (m, 10H). 13C NMR (CDCl3) δ: 22.55, 23.98, 34.73, 51.63, 53.48, 122.53, 126.12, 127.47, 127.53, 128.32, 128.95, 136.21, 137.45, 181.22. Anal. Calcd. for C18H17NO: C, 82.10; H, 6.51; N, 5.32. Found: C, 82.04; H, 6.48; N, 5.30.

(±)-1-Phenyl-3-(2-phenylethyl)-3-azabicyclo[3.1.0]hexan-2-one (10). (Yield 90 %); M. p. 91-93 °C; IR (KBr): 1664 (C=O); 1H NMR (CDCl3) δ: 1.20 (dd, 1H J = 4.8, 7.5 Hz), 1.45 (dd, 1H J = 7.5, 7.9, Hz) 2.00-2.20 (m, 1H), 2.88 (t, 2H, J = 6.8), 3.30 (d, 1H, J = 10.4 Hz), 3.49 (d, 1H, J = 10.2 Hz), 4.20 (dd, 1H, J = 6.0, 10.3 Hz), 7.12-7.60 (m, 10H). 13C NMR (CDCl3) δ: 23.71, 24.18, 34.13, 37.74, 50.22, 54.33, 122.11, 126.42, 126.78, 128.90, 128.94, 129.88, 136.41, 138.35, 182.02. Anal. Calcd. for C19H19NO: C, 82.10; H, 6.51; N, 5.32. Found: C, 82.04; H, 6.48; N, 5.30.

(+)-(1R,5S)-1-Phenyl-3-(2-phenylethyl)-3-azabicyclo[3.1.0]hexan-2-one (10). (Yield 87 %); M. p. 91-93 °C; [α]D 20 + 59.4°; IR, 1H NMR and 13C NMR are identical to those of the racemate.

Anal. Calcd. for C19H19NO: C, 82.28; H, 6.90; N, 5.05. Found: C, 82.20; H, 6.88; N, 5.01.

(–)-(1S,5R)-1-Phenyl-3-(2-phenylethyl)-3-azabicyclo[3.1.0]hexan-2-one (10). Yield 90 %. M. p. 91-93 °C; [α]D 20 – 58.3 IR, 1H NMR and 13C NMR are identical to those of the racemate.

Anal. Calcd. for C19H19NO: C, 82.28; H, 6.90; N, 5.05. Found: C, 82.15; H, 7.00; N, 5.00.

(±)-1-Phenyl-3-(3-phenylpropyl)-3-azabicyclo[3.1.0]hexan-2-one (11). Yield 90 %; M. p. 100-102 °C; IR (KBr): 1683 (C=O); 1H NMR (CDCl3) δ: 1.35 (dd, 1H J = 4.5, 7.3 Hz), 1.53 (dd, 1H J = 7.3, 7.9, Hz) 1.80-2.10 (m, 3H), 2.70 (t, 2H, J = 6.9), 3.35 (t, 2H, J = 7.2), 3.51 (d, 1H, J = 10.6 Hz), 4.16 (dd, 1H, J = 5.8, 10.4 Hz), 7.05-7.45 (m, 10H). 13C NMR (CDCl3) δ: 2.32,
23.90, 27.72, 33.11, 36.24, 47.12, 51.11, 121.31, 126.14, 126.44, 128.11, 128.45, 129.96, 138.88, 139.90, 181.27. Calcd. for C_{20}H_{21}NO: C, 82.44; H, 7.26; N, 4.81. Found: C, 82.37; H, 7.28; N, 4.85.

(+)-(1R,5S)-1-Phenyl-3-(3-phenylpropyl)-3-azabicyclo[3.1.0]hexan-2-one (11). (Yield 87 %); M. p. 100-102 °C; [α]_D^{20} + 57.7 °; IR, 1H NMR and 13C NMR are identical to those of the racemate. Calcd. for C_{20}H_{21}NO: C, 82.44; H, 7.26; N, 4.81. Found: C, 82.37; H, 7.28; N, 4.85.

(–)-(1S,5R)-1-Phenyl-3-(3-phenylpropyl)-3-azabicyclo[3.1.0]hexan-2-one (11). (Yield 85 %); M. p. 100-102 °C; [α]_D^{20} – 56.4°; IR, 1H NMR and 13C NMR are identical to those of the racemate. Calcd. for C_{20}H_{21}NO: C, 82.44; H, 7.26; N, 4.81. Found: C, 82.48; H, 7.35; N, 4.80.

(+)-3-(3-Methylbut-2-enyl)-1-phenyl-3-azabicyclo[3.1.0]hexane (12). A mixture of (+)-1-phenyl-3-azabicyclo[3.1.0]hexane (4) (184 mg, 0.76 mmol), 4-bromo-2-methyl-2-butene (0.132 ml, 1.14 mmol), and NaHCO_3 (128 mg, 1.52 mmol) in dry DMF (15 ml) was stirred and heated to 80 °C for 6 h. The solvent was then removed under reduced pressure and the residue was extracted with CHCl_3 and water. The organic layers were dried over anhydrous Na_2SO_4 and after evaporation of the solvent the crude product was purified by flash column chromatography using CHCl_3/cyclohexane/EtOH (5:4:1) as eluent. The free base (103 mg, yield 60%) was dissolved in diethyl ether and treated with a solution of oxalic acid dihydrate in diethyl ether to give the oxalate salt as a white solid. The analytically pure sample was obtained by crystallization from methanol/diethyl ether.

M. p. 175-178 °C; 1H NMR (DMSO-d_6) δ: 1.04 (dd, 1H, J = 4.9, 8.0 Hz), 1.40 (dd, 1H, J = 3.8, 4.9 Hz), 1.70 (s, 3H), 1.75 (s, 3H, 2.09-2.18 (m, 1H), 3.20-3.90 (m, 6H), 4.90 (s, broad, 2H), 5.29 (t, 1H, J = 7.4), 7.12-7.42 (m, 5H). 13C NMR (DMSO-d_6) δ: 16.25, 18.03, 23.24, 25.67, 30.06, 51.48, 54.10, 56.71, 115.42, 126.41, 126.49, 128.43, 139.82, 140.37, 164.38. Anal. Calcd. for C_{18}H_{23}NO_4: C, 68.12; H, 7.30; N, 4.41. Found: C, 68.03; H, 7.55; N, 4.45.

Compounds 13-19 were synthesized using the same procedure reported for compound 4.

(±)-1-Phenyl-3-propyl-3-azabicyclo[3.1.0]hexane (13). (Yield 95 %); M. p. 124-26 °C; 1H NMR (DMSO-d_6) δ: 0.90 (t, 3H, J = 7.6), 1.04 (dd, 1H J = 8.1, 5.7 Hz), 1.42 (dd, 1H J = 4.8, 5.7 Hz), 1.55-1.70 (m, 2H), 2.00-2.20 (m, 1H), 3.03 (t, 2H, J = 8.6), 3.36 (dd, 1H, J = 4.0, 11.0 Hz), 3.45 (d, 1H, J = 10.8 Hz), 3.63 (d, 1H, J = 10.8 Hz), 3.91 (d, 1H, J = 10.8 Hz), 6.18 (s broad, 2H) 7.18-7.47 (m, 5H). 13C NMR (DMSO-d_6) δ: 11.08, 16.52, 18.98, 30.18, 54.95, 56.08, 57.38, 126.42, 126.48, 128.43, 139.83, 164.49. Anal. Calcd. for C_{16}H_{21}NO_4: C, 65.96; H, 7.27; N, 4.81. Found: C, 65.88; H, 7.45; N, 4.79.

(±)-3-Cyclohexyl-1-phenyl-3-azabicyclo[3.1.0]hexane (14). (Yield 93 %); M. p. 124-26 °C; 1H NMR (DMSO-d_6) δ: 1.03-1.90 (m, 10H), 1.97-2.14 (m, 3H), 3.00-3.25 (m, 1H), 3.43 (m, 3H), 4.01 (d, 1H, J = 10.8 Hz), 5.16 (s broad, 4H), 7.20-7.42 (m, 5H). 13C NMR (DMSO-d_6) δ: 22.99, 24.25, 24.59, 28.50, 28.55, 29.81, 33.30, 55.98, 64.46, 126.49, 126.52, 128.41, 139.64, 164.44. Anal. Calcd. for C_{16}H_{25}NO_4: C, 68.86; H, 7.60; N, 4.23. Found: C, 68.56; H, 7.63; N, 4.20.
(±)-(1R,5S)-3-Cyclohexyl-1-phenyl-3-azabicyclo[3.1.0]hexane (14). (Yield 95 %); M. p. 123-25°C; [α]D20 + 58.2°; 1H NMR and 13C NMR are identical to those of the racemate. Anal. Calcd. for C19H25NO4: C, 68.86; H, 7.60; N, 4.23. Found: C, 68.55; H, 7.58; N, 4.20.

(–)-(1S,5R)-3-Cyclohexyl-1-phenyl-3-azabicyclo[3.1.0]hexane (14). (Yield 97 %); M. p. 124-26°C; [α]D20 – 60.4°; 1H NMR and 13C NMR are identical to those of the racemate. Anal. Calcd. for C19H25NO4: C, 68.86; H, 7.60; N, 4.23. Found: C, 68.58; H, 7.65; N, 4.25.

(±)-3-(Cyclohexylmethyl)-1-phenyl-3-azabicyclo[3.1.0]hexane (15). (Yield 58 %); M. p. 140-43 °C; 1H NMR (DMSO-d6) δ: 0.78-1.38 (m, 7H), 1.49 (d, 1H, J = 5.0, 5.4 Hz), 1.55-1.89 (m, 5H), 2.09 (m, 1H), 2.91 (d, 2H, J = 6.0 Hz), 3.21-3.43 (m, 2H), 3.60 (d, 1H, J = 10.4 Hz), 3.88 (d, 1H, J = 10.4 Hz), 5.87 (s broad, 4H), 7.18-7.48 (m, 5H). 13C NMR (CDCl3) δ: 16.90, 23.44, 25.16, 25.20, 25.68, 30.23, 30.51, 30.59, 34.52, 55.71, 58.07, 61.07, 126.40, 126.41, 128.38, 140.16, 164.10. Anal. Calcd. for C20H27NO4: C, 69.54; H, 7.88; N, 4.05. Found: C, 69.48; H, 7.66; N, 4.04.

(+)-(1R,5S)-3-(Cyclohexylmethyl)-1-phenyl-3-azabicyclo[3.1.0]hexane (15). (Yield 53 %); M. p. 140-43 °C; [α]D20 + 67.3°; 1H NMR and 13C NMR are identical to those of the racemate. Anal. Calcd. for C20H27NO4: C, 69.54; H, 7.88; N, 4.05. Found: C, 69.45; H, 7.66; N, 4.02.

(–)-(1S,5R)-3-(Cyclohexylmethyl)-1-phenyl-3-azabicyclo[3.1.0]hexane (15). (Yield 55 %); M. p. 140-43 °C; [α]D20 – 64.4°; 1H NMR and 13C NMR are identical to those of the racemate. Anal. Calcd. for C20H27NO4: C, 69.54; H, 7.88; N, 4.05. Found: C, 69.43; H, 7.55; N, 4.03.

(±)-3-(1-Adamantylmethyl)-1-phenyl-3-azabicyclo[3.1.0]hexane (16). (Yield 83 %); M. p. 240-42 °C; 1H NMR (DMSO-d6) δ: 1.01 (dd, 1H, J = 5.0, 7.6 Hz), 1.50-1.90 (m, 12H), 1.95-2.23 (m, 5H), 3.00 (d, 2H, J = 4.6 Hz) 3.42-3.70 (m, 2H), 3.85 (dd, 1H, J = 4.6, 10.0 Hz), 4.20 (dd, 1H, J = 4.8, 10.0 Hz), 7.15-7.48 (m, 5H), 9.7 (s broad, 1H). 13C NMR (DMSO-d6) δ: 16.40, 23.21, 27.50, 30.61, 33.02, 35.85, 58.43, 60.60, 68.22, 126.64, 126.66, 128.44, 139.45. Anal. Calcd. for C24H31NO4: C, 72.52; H, 7.86; N, 3.52. Found: C, 72.30; H, 7.58; N, 3.50.

(±)-3-Benzyl-1-phenyl-3-azabicyclo[3.1.0]hexane (17). (Yield 93 %); M. p. 169-72 °C; 1H NMR (DMSO-d6) δ: 0.95 (dd, 1H, J = 5.2, 8.2 Hz), 1.45 (t, 1H, J = 5.0, Hz) 1.95-2.15 (m, 1H), 3.23 (dd, 1H, J = 3.8, 7.2 Hz), 3.25 (m, 2H), 3.58 (d, 1H, J = 10.0 Hz) 4.13 (s, 2H), 4.58 (s broad, 2H) 7.15-7.58 (m, 10H). 13C NMR (DMSO-d6) δ: 16.93,23.61, 30.21, 54.69, 57.42, 58.00, 126.30, 128.38, 128.56, 129.89, 134.18, 140.57, 163.47. Anal. Calcd. for C20H21NO4: C, 70.78; H, 6.24; N, 4.13. Found: C, 70.70; H, 6.28; N, 4.15.

(±)-1-Phenyl-3-(2-phenylethyl)-3-azabicyclo[3.1.0]hexane (18). (Yield 68 %); M. p. 215-18 °C; 1H NMR (DMSO-d6) δ: 1.05 (dd, 1H J = 5.2, 8.0 Hz), 1.42 (dd, 1H J = 4.9, 5.2, Hz) 2.05-2.23 (m, 1H), 2.94 (dd, 2H, J = 5.2, 10.6), 3.27 (t, 2H, J = 9.0), 3.10-3.50 (m, 2H), 3.62 (d, 1H, J = 10.8 Hz), 3.90 (d, 1H, J = 10.6 Hz), 4.49 (s broad, 2H), 7.15-7.50 (m, 10H). 13C NMR (DMSO-d6) δ: 16.48, 23.26, 30.01, 31.89, 55.06, 55.52, 57.50, 126.46, 126.60, 126.65, 128.43, 128.57, 128.68, 137.76, 139.96, 164.30. Anal. Calcd. for C21H23NO4: C, 71.37; H, 6.56; N, 3.96. Found: C, 71.07; H, 6.58; N, 3.93.
(+)-(1R,5S)-1-Phenyl-3-(2-phenylethyl)-3-azabicyclo[3.1.0]hexane (18). (Yield 48 %); M. p. 215-18 °C; [α]D20 = +60.4°; 1H NMR and 13C NMR are identical to those of the racemate. Anal. Calcd. for C21H23NO4: C, 71.37; H, 6.56; N, 3.96. Found: C, 71.12; H, 6.51; N, 3.91.

(−)-(1S,5R)-1-Phenyl-3-(2-phenylethyl)-3-azabicyclo[3.1.0]hexane (18). (Yield 54 %); M. p. 215-18 °C; [α]D20 = +59.2°; 1H NMR and 13C NMR are identical to those of the racemate. Anal. Calcd. for C21H23NO4: C, 71.37; H, 6.56; N, 3.96. Found: C, 70.99; H, 6.48; N, 3.90.

(±)-1-Phenyl-3-(3-phenylpropyl)-3-azabicyclo[3.1.0]hexane (19). (Yield 88 %); M. p. 159-62 °C; 1H NMR (DMSO-d6) δ: 1.09 (dd, 1H, J = 5.2, 8.2 Hz), 1.40 (dd, 1H, J = 4.9, 5.2, Hz) 1.80-2.03 (m, 2H), 2.04-2.12 (m, 1H) 2.59 (t, 2H, J = 7.4 Hz), 3.08 (t, 2H, J = 7.8), 3.36 (dd, 1H, J = 3.2, 10.4 Hz), 3.40 (d, 1H, J = 10.6 Hz), 3.60 (d, 1H, J = 10.8 Hz), 3.90 (d, 1H, J = 10.4 Hz), 5.25 (s broad, 2H) 7.09-7.48 (m, 10H). 13C NMR (CDCl3) δ: 16.45, 23.23, 27.16, 30.0, 32.18, 54.03, 55.0, 57.40, 126.06, 126.41, 126.44, 127.67, 128.28, 139.83, 140.73, 164.22. Anal. Calcd. for C22H25NO4: C, 71.91; H, 6.86; N, 3.81. Found: C, 72.00; H, 6.97; N, 3.85.

(+)-(1R,5S)-1-Phenyl-3-(3-phenylpropyl)-3-azabicyclo[3.1.0]hexane (19). (Yield 90 %); M. p. 159-62 °C; [α]D20 = +60.9°; 1H NMR and 13C NMR are identical to those of the racemate. Anal. Calcd. for C22H25NO4: C, 71.91; H, 6.86; N, 3.81. Found: C, 72.13; H, 6.98; N, 3.85.

(−)-(1S,5R)-1-Phenyl-3-(3-phenylpropyl)-3-azabicyclo[3.1.0]hexane (19). (Yield 90 %); M. p. 100-102 °C; [α]D20 = +59.5°; 1H NMR and 13C NMR are identical to those of the racemate. Anal. Calcd. for C22H25NO4: C, 71.91; H, 6.86; N, 3.81. Found: C, 72.10; H, 6.95; N, 3.83.

Radiology and binding assays

σ1-Site binding assays were carried out on guinea pig brain membranes as previously reported. 19 Briefly, each tube containing 500 µg of membrane protein was incubated for 150 min at 37 °C with [3H]-(+)-pentazocine (3 nM) in 50 mM Tris-HCl pH 7.4. Non-specific binding was determined using 10 µM haloperidol. The final volume of the assay samples was 1.0 mL. After incubation the samples were filtered through a Schleicher & Schnell GF 6 glass fiber filter which had been pre-soaked for 1 h in a 0.5% poly(ethyleneimine) solution. Filters were washed twice with 4 ml of ice-cold buffer before transfer to scintillation vials.

σ2-Site binding assays were carried out on guinea pig brain membranes, prepared as previously described by Mach et al. 20 The membranes were incubated with [3H]DTG [1,3-di-(2-tolyl)-guanidine] (3 nM) in the presence of 400 nM (+)-SKF10,047 (to block binding to σ1 sites). The final volume of the assay sample was 0.5 mL. Incubations proceeded for 2 h at room temperature in 50 mM Tris-HCl, pH 8.0. Non-specific binding was evaluated in the presence of 5 µM DTG. Each assay was terminated by the addition of ice-cold 10 mM Tris-HCl pH 8.0, followed by filtration through a poly(ethyleneimine) (0.5% w/v) treated GF 6 glass fiber filter which were washed twice with 4 ml of ice-cold buffer before transfer to scintillation vials. The Ki values were calculated using the EBDA/LIGAND program 21 purchased from Elsevier/Biosoft.
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References

3. Hellewell, S. B.; Bowen, W. D. Brain Res. 1990, 527, 244.