Available online at www.sciencedirect.com
SCIENCE DIRECT®
Journal of Molecular Graphics and Modelling 22 (2004) 221–230
Journal of Molecular Graphics and Modelling
www.elsevier.com/locate/JMGM

Molecular modeling of σ1 receptor ligands: a model of binding conformational and electrostatic considerations

Tamara M. Gund*, Jie Floyd, Dawoon Jung

Department of Chemistry, Chemical Engineering and Environmental Science, New Jersey Institute of Technology, University Heights, Newark, NJ 07102, USA

Accepted 14 August 2003

Abstract

We have performed molecular modeling studies on several σ1 specific ligands, including PD144418, spipethiane, haloperidol, pentazocine, and others to develop a pharmacophore for σ1 receptor–ligand binding, under the assumption that all the compounds interact at the same receptor binding site. The modeling studies have investigated the conformational and electrostatic properties of the ligands. Superposition of active molecules gave the coordinates of the hypothetical 5-point σ1 pharmacophore, as follows: R1 (0.85, 7.26, 0.30); R2 (5.47, 2.40, −1.51); R3 (−2.57, 4.82, −7.10); N (−0.71, 3.29, −6.40); carbon centroid (3.16, 4.83, −0.60), where R1, R2 were constructed onto the aromatic ring of each compound to represent hydrophobic interactions with the receptor; and R3 represents a hydrogen bond between the nitrogen atom and the receptor. Additional analyses were used to describe secondary binding sites to electronegative groups such as oxygen or sulfur atom. Those coordinates are (2.34, 5.08, −4.18). The model was verified by fitting other σ1 receptor ligands. This model may be used to search conformational databases for other possibly active ligands. In conjunction with rational drug design techniques the model may be useful in design and synthesis of novel σ1 ligands of high selectivity and potency. Calculations were performed using Sybyl 6.5.

Keywords: σ1 receptor; σ1 ligand; Pharmacophore; Sybyl

1. Introduction

Sigma receptor binding sites are non-opiate, non-phencyclidine, and non-dopamine receptors with a unique drug selectivity pattern and anatomical distribution [1]. The cloning and functional expression of a σ receptor extracted from guinea pig liver [2,3] and humans [4] proves its existence. σ receptors have been the focus of extensive studies because of their potentially important roles in biochemical, physiological, and behavioral processes. Potential therapeutic use has been foreseen for σ ligands in psychiatric diseases [5], in the treatment of cocaine abuse, in neuroprotection, in the treatment of schizophrenia, and in mediating the antipsychotic effects by inhibiting neurotransmitter release [6–9].

Sigma receptors have been classified into σ1 and σ2 subtypes based upon the different biochemical and pharmacological properties of structurally diverse ligands [10,11]. Recently a σ3 receptor subtype [12–14] has also been postulated. σ1, unlike the σ2 site, displays restricted stereospecificity for the (+)-isomers of benzomorphans, morphinans and other opiates. Due to the pharmacological and structural affiliations of the σ1 site with yeast and mammalian sterol C8-C7 isomerases, the σ1 binding site was proposed to be carried by a sterol isomerase-related protein involved in postsqualene cholesterol biosynthesis [15,16]. Early studies suggested that σ1 sites regulate gastrointestinal effects, inhibit both electrical and serotonin-induced guinea pig ileum contractions, and mediate the motor effects of σ ligands. Furthermore the σ1 receptor plays an important role in the facilitation of central cholinergic functions. In addition, the specific agonists for the σ1 receptor may be novel candidates as therapeutic agents [17–21].

Sigma2 sites may mediate the effects of certain σ drugs on K^+ channels. They may also be associated with calcium channels on the basis of the modulatory effects of inorganic calcium channel blockers such as Cd^{2+} on σ1 and σ2 sites. Sigma2 agents may be useful therapeutic agents for treatment of drug-induced and idiotypic motor disorders such as dystonia.

Understanding the function of the receptor subtypes and the binding mechanism of sigma ligand subtypes may lead to the development of selective and potent therapeutic agents which could be used in treating various mental, motor, and other disorders. At this time, the precise function

* Corresponding author. Tel.: +1-973-596-3669; fax: +1-973-596-3586.
E-mail address: gund@adm.njit.edu (T.M. Gund).

1093-3263/$ – see front matter © 2003 Elsevier Inc. All rights reserved.
doi:10.1016/j.jmgm.2003.08.001

of the σ receptors, and the clinical relevance of σ1 and σ2 subtypes are not clear.

Recently, many high affinity σ1 ligands have been synthesized. These include derivatives of: piperidine and piperazine [17,20,22–30]; alkylamine [18,19,23,31–33]; dextromethorphan [34]; morphan [35]; NANM (SKF10047, *N*-allyl-*N*-normetazone) [36,37]; benzoxazolone and benzothiazolone [38]; camphidine [39] and benzamide [40]. Furthermore, various SAR studies of σ1 ligands were also performed [3,25,26,31–36,41]. On the other hand, very few σ2 selective ligands are known. Examples include: azaperol [42], related BMY-14802 (4-amino-1-arylbutanols) [42], vesamicol analogues [43], alkylamine derivatives [44], trishomocubane [45], and *N*-alkylazacycloheptane derivatives [39]. In addition, even fewer studies were made on σ3 ligands [46], probably because subtype 3 warrants further investigation. Many specific radioligands for σ1 receptor subtypes were synthesized [28–30,40,47–51], but none for σ2 or σ3 receptor subtypes. [^{3}H]-(+)-Pentazocine is the most commonly used radiolabel for σ1 sites, while [^{3}H]-DTG in the presence of (+)-pentazocine for σ2 sites.

Several molecular modeling studies [52–56] to determine a pharmacophore of binding have been reported. The model published by Gund and Shukla [53] in 1991 was based on the Manallack model [56], but used a different mode of binding of haloperidol and was extended to other types of ligands that did not contain nitrogen or benzene. The model was established to fit compounds such as progesterone, (+)-pentazocine, pre084 and other compounds. Gund's model proposed several important sites for σ ligand binding. However, the pharmacophore model was developed for general σ classes of compounds before σ subtype ligands were differentiated. Now many new compounds have emerged, σ1, σ2 and σ3 receptor subtypes have been found. With many new potent σ1 ligands that are now available, we were able to determine a new pharmacophore model, which is specific for σ1 ligand binding.

2. Methods

The calculations in this study were carried out using the Sybyl V6.5 molecular modeling program implemented on a SGI platform. The method applied was to locate all possible low energy conformations of each molecule, and then superimpose the chosen conformers at several defined positions.

As in Manallack's model, we have also applied the methods of Lloyd and Andrews [57], who advanced the idea of a common structural model of all CNS active drugs that includes the aromatic ring and N moieties as primary binding groups. To define the primary pharmacophore for the σ binding sites by mapping the topographic arrangements of the phenyl ring, N atom, and N lone pair vector; a point R3 was placed 2.8 Å tetrahedrally from N atoms to represent an interaction between a protonated N atom and its binding site; dummy atoms were built 3.5 Å above and below a phenyl ring to represent hydrophobic bonding to a receptor. In our model, the centroid was not always necessary; sometimes we chose a carbon from a phenyl ring. An electronegative atom such as O or S is usually found in superpotent σ1 ligands between the phenyl group and nitrogen atom, and it shows the capability of influencing the binding affinity [27]; therefore, we also mapped the oxygen (or sulfur) positions with all applicable compounds as a secondary binding site. Receptor models were generated using a computerized technique that allowed the simultaneous minimization of both energy and geometric fit between chosen features.

3. Results

3.1. Choice of σ1 ligands

The choice of compounds for the σ1 subtype was based on potency, selectivity, and structural diversity. The pharmacophore for σ1 was defined using PD144418, spipethiane, haloperidol, (+)-pentazocine with relative affinity ranging from 0.08 to 5.8nm. In this study, we selected ligands shown in Scheme 1 for σ1 receptors based on their affinity and selectivity.

3.2. Generation of molecular structures and conformations

The crystal structures of pentazocine and haloperidol from the Cambridge Structural Database were used as the initial bioactive conformer. Other ligands were built using Sybyl, and then energy-minimized using the Tripos force field with a distance-dependent dielectric function and a convergence criterion of 0.005 kcal/mol of energy difference between successive iterations.

A grid search was performed on each molecule by varying torsion angles to find all low energy conformations. The torsion angles that were varied in this study have been labeled in Scheme 1. For highly flexible compounds such as PRE084, a larger interval of torsion angles was applied first to generate a database; then the range of torsion angles could be narrowed down; if necessary, a smaller 1° interval of torsion angles could be implemented for further searching. We allowed a maximum of 5000 conformers for each grid search. Structures with 10 kcal less than the global minimum were considered as possible candidates for bioactive conformations.

3.3. Superimposition of the four key molecules PD144418, spipethiane, haloperidol, (+)-pentazocine

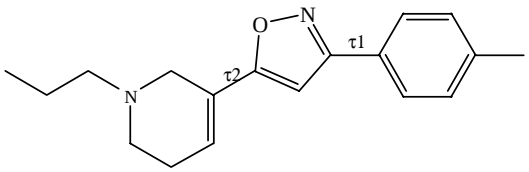
PD144418 was by far the most potent and selective σ1 ligand, and was used as the template. Spipethiane is the second most selective and also a very potent σ1 ligand. Haloperidol and (+)-pentazocine are active molecules of the butyrophenone and benzomorphan drug classes respectively.

In order to align O from PD144418 with S from spipethiane, the dihedral angle around the rotatable bond τ2 of PD144418 must be 0° or in the s-*cis* position. The energy of the minimum s-*cis* conformation is only 0.7 kcal higher than its s-*trans* global minimum conformer. Therefore, the lowest energy s-*cis* conformer was chosen as the initial bioactive structure. The global minimum energy of spipethiane was chosen from its grid search study performed on varying torsion angles 1 and 2 as shown in Scheme 1. Choice of C-center of the four molecules was done on an atom-by atom basis to achieve the best fit.

We applied the multifit command in Sybyl. This procedure minimizes the respective geometric fits of the molecules by altering the torsion angles of each molecule while minimizing the structure. Multifitting three molecules PD144418, spipethiane and haloperidol on positions R1, R2, R3, C-center, O and N gave three molecular structures with good fitting to each other. They were of higher energy but less than 10 kcal/mol from their global minimum energy. The fitting of spipethiane to PD144418 had a RMS of 0.289; fitting of haloperidol to PD144418 was 0.755. These three structures were used as the final bioconformers.

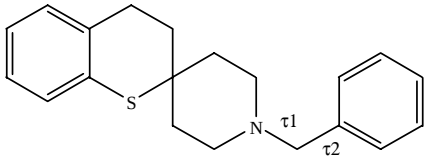
(a)

PD144418



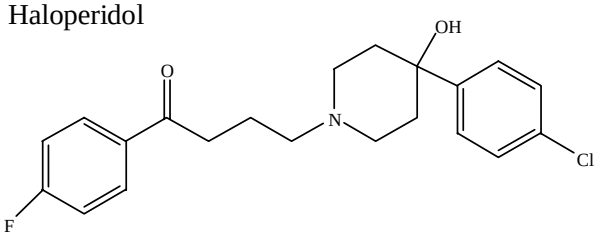
(1-propyl-5-(3-*p*-tolyl-isoxazol-5-yl)-1,2,3,6-tetrahydropyridine

Spipethiane

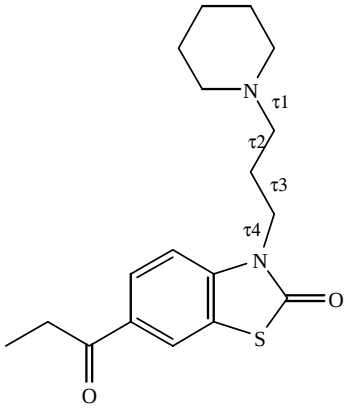


1-benzyl-3,4-dihydrospiro[2H-1-benzothiopyran-2,4'-piperidinel]

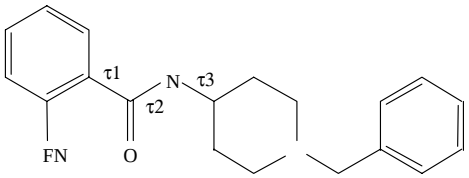
Haloperidol



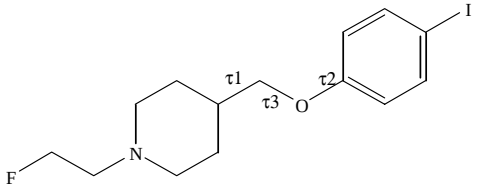
4-[4-(4-chlorophenyl)-4-hydroxy-1-piperidinyl]-1'-(4-fluorophenyl)-1-butanone



3-(1-piperidinoethyl)-6-propylbenzothiazolin-2-one



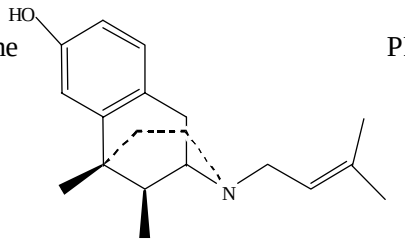
N-(N-benzylpiperidin-4-yl)-2-flurobenzamide



1-(2-fluoroethyl)-4-[(4-iodophenoxy)methyl]piperidine

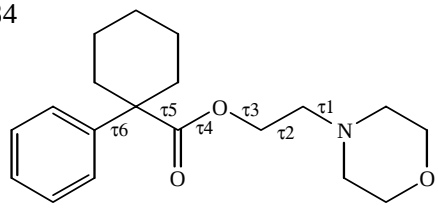
Scheme 1. σ1 Ligands examined in this study.

(+) –Pentazocine



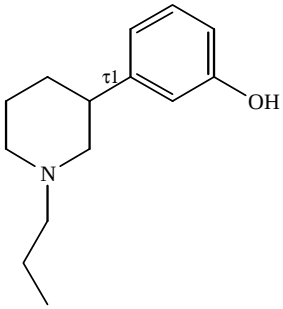
(+)-1,2,3,4,5,6-Hexahydro-6,11-dimethyl-3-(3-methyl-2-butenyl)-2,6-methano-3-Benzazocin-8-ol.

PRE084



2-(4-morpholino)ethyl-l-phenylcyclohexane-1-carboxylate

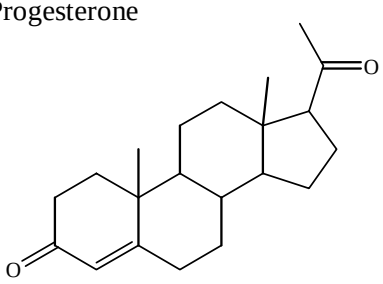
(+)3PPP



(+)-3-(3-hydroxyphenyl)-N-(1-propyl)piperidine

(b)

Progesterone



Scheme 1. (*Continued*).

Fitting the bioconformer PD144418 with various energy conformations of (+)-pentazocine gave RMS values of 0.812–1.007. The better fitting result, however, used a high-energy conformer ($\Delta E > 10$ kcal). The highest RMS value used a low energy conformation of (+)-pentazocine ($\Delta E = 0.5$). Finally to derive the best model, we compromised energy and fit, and used a conformation of (+)-pentazocine with an energy of $\Delta E = 4.1$ kcal and obtained a RMS of 0.949.

The three molecules, spipethiane, haloperidol, (+)-pentazocine were then fitted to PD144418. The final fitting conformers with sites of superposition and with the pharmacophore model are shown in Fig. 1. A 3D stereoscopic view of the superimposition of the four chosen bioconformers with the superimposed pharmacophore model is shown in Fig. 2.

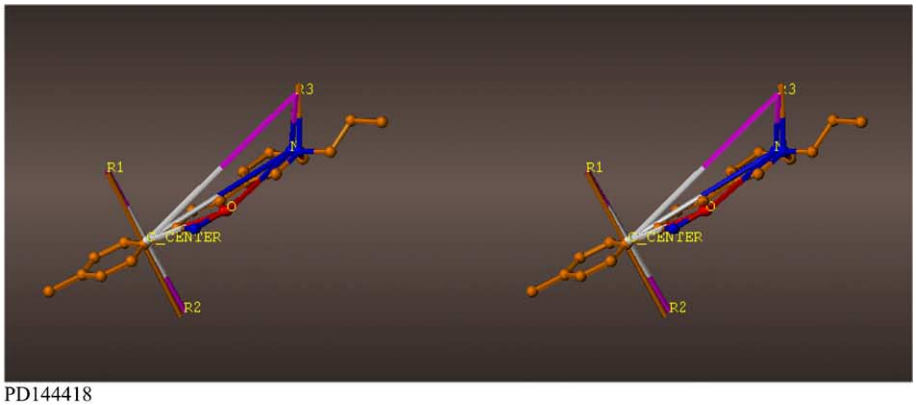
3.4. Primary pharmacophore

The receptor points of the R1, R2, R3, C-center and N atoms for molecules PD144418, spipethiane, haloperidol, (+)-pentazocine were averaged to give the following coordinates (in Å) of the primary model: R1 (0.85, 7.26, 0.30); R2 (5.47, 2.40, −1.51); R3 (−2.57, 4.81, −7.10); N (−0.71, 3.29, −6.40); Carbon center (3.16, 4.83, −0.60). The distance from the C-center to the N atom was 7.14 Å; from the C-center to the R3 was 8.66 Å; from R3 and N atom was 2.80 Å. The angles R1-C-N and C-N-R3 were 90.21°, 119.78°, respectively, and the dihedral angle R1-C-N-R3 was 12.00°.

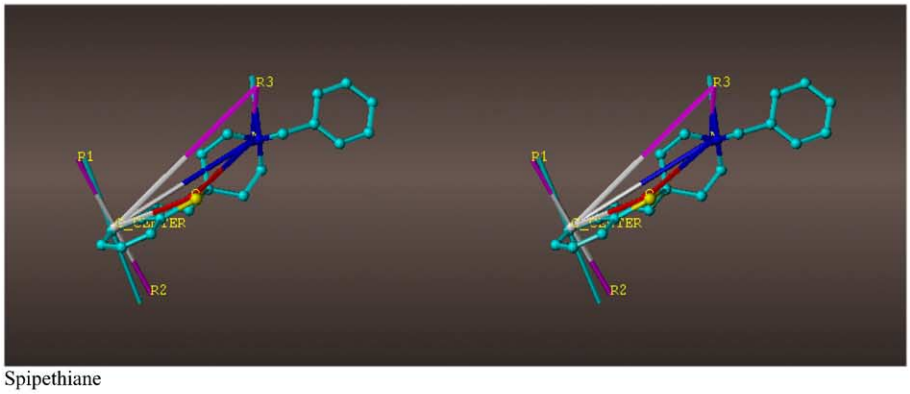
3.5. Secondary binding requirements

The O atom from PD144418, the carbonyl oxygen from haloperidol, and S atom from spipethiane were fitted and averaged to give the following coordinates (2.33, 5.08, −4.18). The distance from O to C-center was 3.68 Å, O to N was 4.17 Å, and angle of C–O–N was 130.71°.

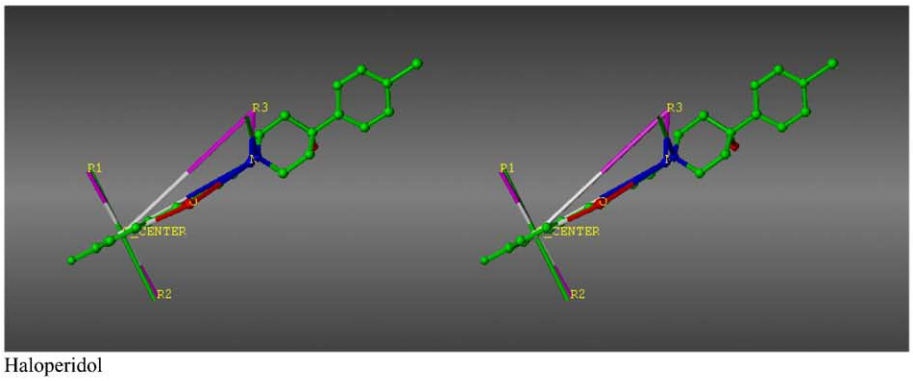
By studying many structures of σ1 ligands, we concluded that besides the primary binding regions, there could be a secondary binding region that surrounds the oxygen and sulfur atom of the molecules studied. This pharmacophore model is shown in Fig. 3.



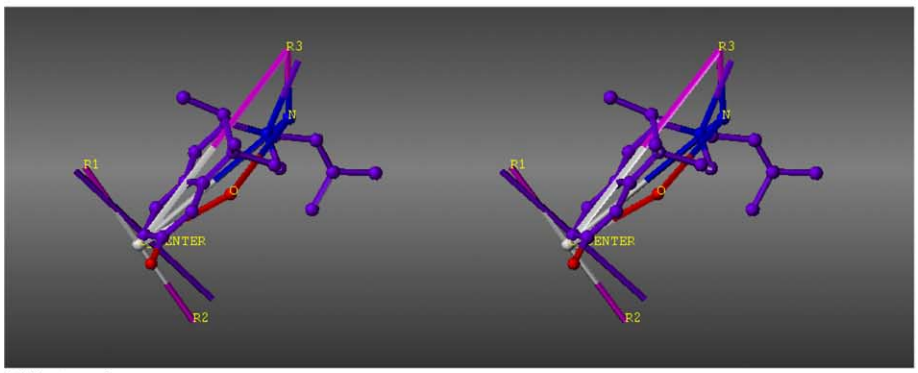
PD144418



Spipethiane



Haloperidol



(+)Pentazocine

Fig. 1. A stereoscopic view of the chosen fitting conformers of PD144418, spipethiane, (+)-haloperidol, and pentazocine. Sites of superposition are indicated by R1 to R3, C-center, nitrogen, lone pair, and oxygen or sulfur.

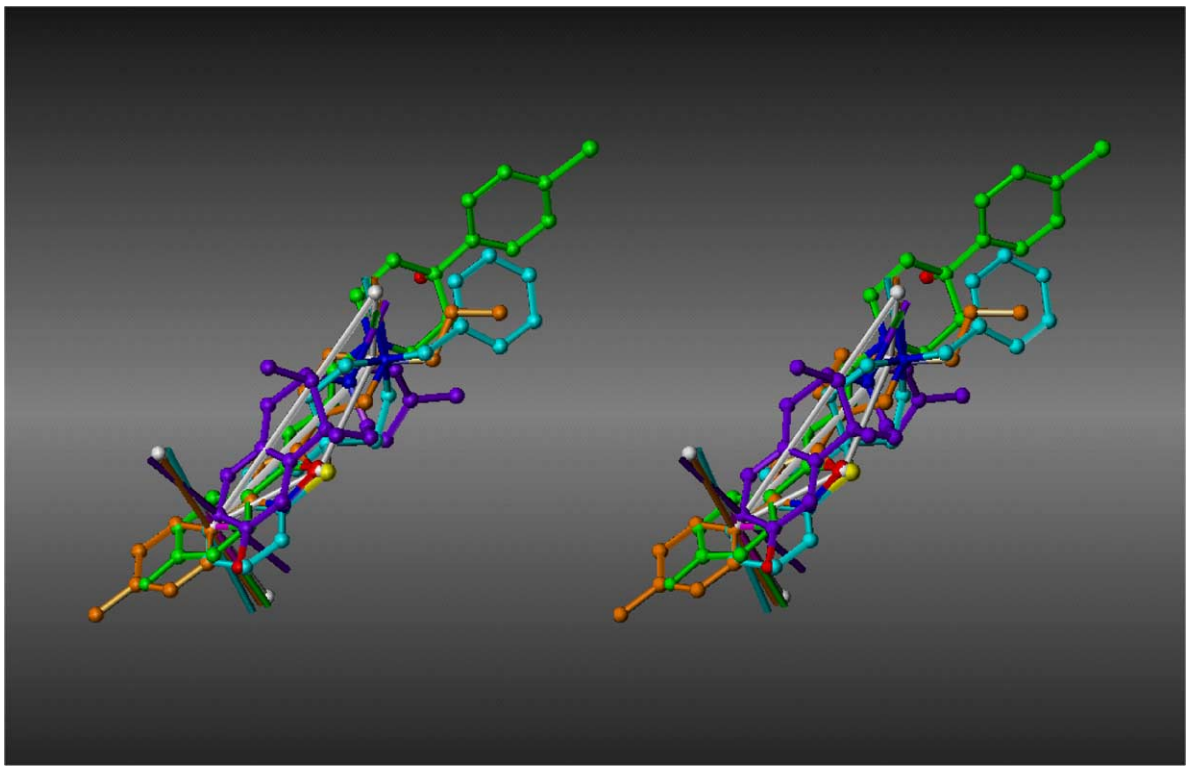
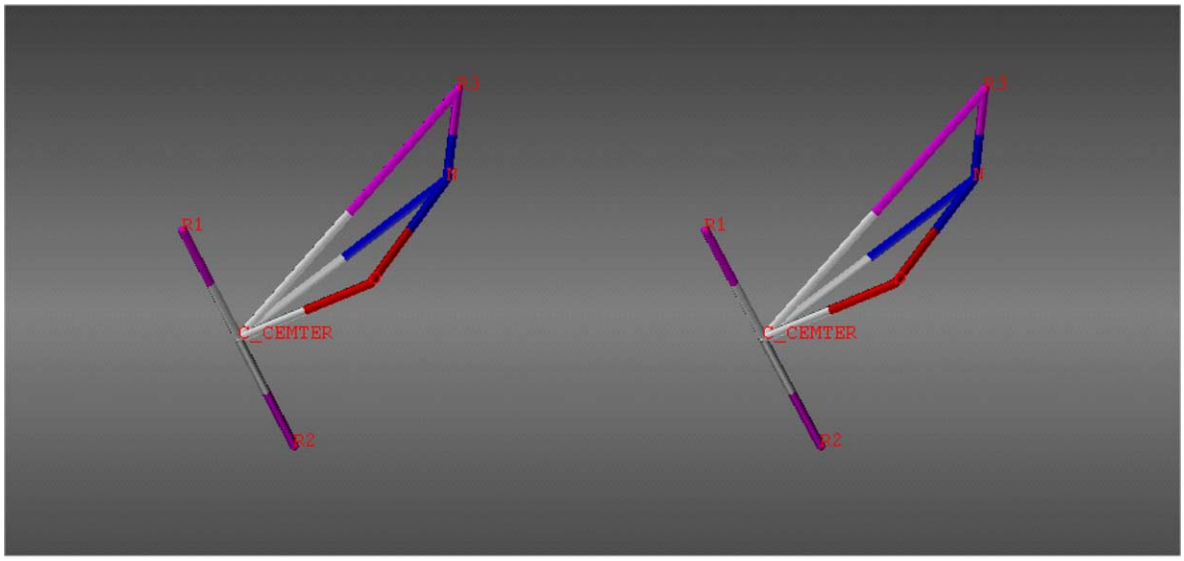


Fig. 2. A stereoscopic view of the superimposition of chosen conformers of PD144418 (orange), spipethiane (cyan), haloperidol (green) and pentazocine (purple), with the pharmacophore model (white); nitrogens are blue; oxygens are red; and sulfur is yellow.

3.6. Electrostatic potential surfaces

Net atomic charges were calculated using AM1 from MOPAC. The net charges were then used to calculate the electrostatic isopotentials, and the potentials were graphically coded according to the magnitude of the potential where the highest positive value indicates the most repulsive interaction with a positively charged probe. The net charges



Distances: C_Center - N: 7.14Å; C_Center - R3: 8.66Å; O - N: 4.17 Å
Angles: R1-C_Center-N: 90.21º; C-N-R3: 119.78º ; O-C_Center: 3.68Å; C-O-N: 130.71º
Torsion: R1-C–Center-N-R3: 12.00º

Fig. 3. A stereoscopic view of the sigma1 pharmacophore receptor model detailing the position of receptor points R1, R2, R3, and a site for the electronegative atom.

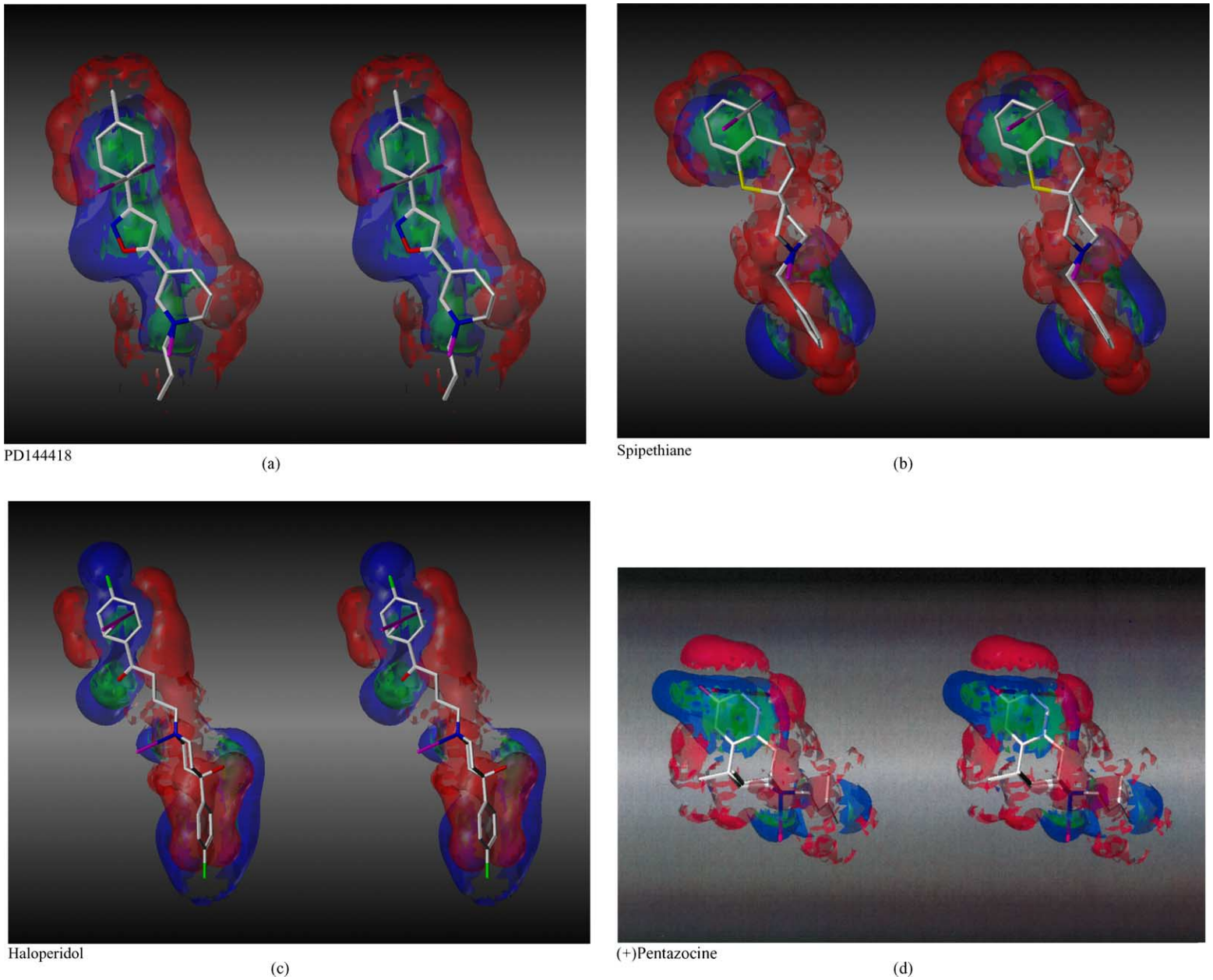


Fig. 4. Isopotential contour surfaces for PD144418, spipethiane, haloperidol and (+)-pentazocine. The contour levels are as follow: red +10 kcal; blue −4 kcal; green −15 kcal, the grid spacing is set as 0.4, the contour planes are in all planes.

were used to generate isopotential contour surfaces. The contour levels were as follows: red +10 kcal, blue −4 kcal, green −15 kcal, and the grid space was set at 0.4.

Electrostatic isopotential contour surfaces for PD144418, spipethiane, haloperidol, and (+)-pentazocine were derived and are shown in Fig. 4. There is consistency with the position of charge for the ligands. Low potentials were found around the oxygen atoms (or sulfur), nitrogen and their lone pairs, and around the hydrophobic regions. In fact, all receptor points were located in low electrostatic potential surface areas.

4. Discussion

Aided by data from radio receptor and previous pharmacological studies, our research has applied computer graphics in conjunction with energy, and degree of fit to develop a pharmacophore model for binding to σ1 receptor sites. Our model takes into account primary and secondary binding sites. For high potency, a compound must not only 'fit' the respective primary sites, but its secondary electronegative binding groups should also be in favorable locations.

Table 1
ΔE, RMS distance, potency, selectivity of several σ1 ligands

Compounds	ΔE (kcal/mol)	RMS	(σ_1 potency)/ (σ_2 potency)	σ_1 potency (nM)	σ_2 potency (nM)	Reference
PD144418[a]	2.38	0.308	5.8×10^{-5}	0.08	1377	[17]
Spipethiane[a]	5.42	0.422	0.0012	0.5 ± 0.02	416 ± 43	[27]
Haloperidol[a]	3.60	0.547	22	1.2 ± 0.20	26 ± 5.4	[17]
3-(1-Piperidinoethyl)-6-propylbenzothiazolin-2-one[c]	3.03	0.593	0.034	0.6 ± 0.3	18.1 ± 0.2	[38]
N-(*N*-Benzylpiperidin-4-yl)-2-flurobenzamide[c]	3.87	0.627	0.0083	3.39	406	[49]
1-(2-Fluoroethyl)-4-[(4-Iodophenoxy)methyl] piperidine[c]	1.06	0.810	0.0082	0.84	102	[29]
(+)-Pentazocine[a]	4.10	0.737	0.0046	5.8 ± 1.0	1253 ± 519	[17]
PRE084[c]	1.42	0.781		44[b]	NA	[58]
(+)3PPP[c]	1.93	0.54	7	23.7 ± 3.8	176.3 ± 23	[3]
Progesterone[c]	0.00	0.93		268	NA	[4]

ΔE: energy above global minimum, RMS distances compared with the σ1 model, potencies (in nM), the ratio of potency of σ1 over σ2. [^{3}H]-DTG in the presence of (+)-pentazocine to label σ2 sites, [^{3}H]-(+)-pentazocine as radioligand to label σ1 sites, unless explained in the above table otherwise.

[a] Bioactive ligands used to derive the pharmacophore model.

[b] Used [^{3}H]-(+)-SKF-10047 as radioligands.

[c] Other molecules used to test the pharmacophore model.

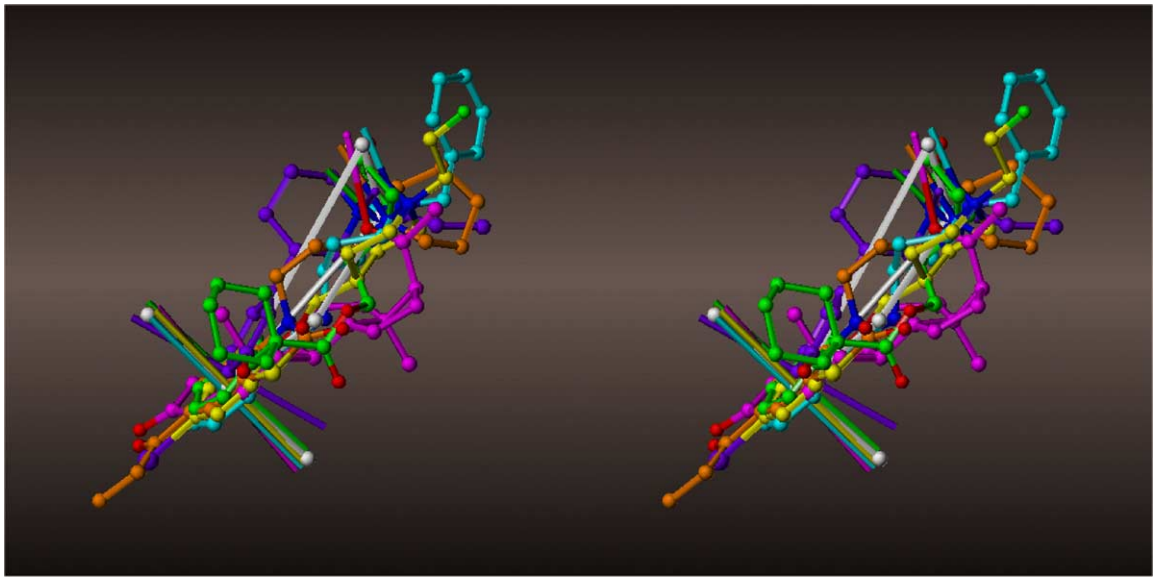


Fig. 5. A stereo view of the superimposition of chosen bioactive conformers of 3-(1-piperidinoethyl)-6-propylbenzothiazolin-2-one (orange), *N*-(*N*-benzylpiperidin-4-yl)-2-flurobenzamide (cyan), 1-(2-fluoroethyl)-4-[(4-iodophenoxy)methyl]piperidine (yellow), (+)3PPP (purple), PRE084 (green) and progesterone (magenta) with the pharmacophore model (white); nitrogens are blue; and oxygens are red.

This model agrees with our previous model and with Manallack's model [43] that a hydrophobic site represented by a phenyl group, a hydrophilic site represented by the nitrogen and its lone pair were required for primary binding. The new model also agrees with our previous model [30] for the important binding groups, however, the position of receptor points in this model had to be altered to fit the new structures. We also added a secondary binding site that includes electronegative atoms

In view of the variety of compounds possessing high affinity for the σ1 site, we examined several structurally unrelated ligands in our model, shown in Table 1. A good fit is indicated by low values for both the RMS distance (<0.8 Å) and potential energy (<10 kcal/mol above the global minimum). The searches were conducted from the database generated by grid search to obtain a structure with a good fit.

These highly potent molecules displayed a good fit with the model. The fitting conformers of some of these compounds are shown in Fig. 5. The better fit of the σ1 ligands correlates with stronger potency at the σ1 binding site, as demonstrated in Table 1. PD144418 fits the pharmacophore model the best, it is the most potent σ1 ligand; while pentazocine doesn't fit the model as well, and its potency is lower, progesterone has the lowest potency and fits the pharmacophore model least well. Future drug design strategies for potent σ1 ligands should concentrate on developing a three-dimensional quantitative structure-activity relationship (3D-QSAR) model, which uses diverse structures of σ1 ligands. Currently, we are conducting a study on a large set of σ1 ligands with various structures by implementing the comparative molecular field analysis (CoMFA) method to correlate biological activities with three-dimensional structural properties described by a steric and an electrostatic molecular field. On the other hand, σ2 ligands seem to bind in a very different way, the work on σ2 sites will be reported shortly.

5. Conclusions

We have derived a pharmacophore model for binding to the σ1 receptor using several σ1 specific ligands. The model is based upon the assumption that all the compounds interact at the same binding site at the receptor level. A compromise of fit and low energy was achieved. The important parameters were the fit at the phenyl moieties, nitrogen and its lone pair, oxygen (or sulfur) as the secondary binding site. The model was verified by fitting a test set of σ1 active molecules as shown in Fig. 5.

This receptor pharmacophore model should be useful for finding new compounds that could have potential σ1 activity and in predicting the activity and selectivity of σ1 ligands. There are many available conformational databases that could be searched using this pharmacophore model to identify possible candidates for testing. The model could be useful also for the design of novel ligands which could be synthesized and shown to bind to the σ1 receptor.

References

[1] J.M. Walker, W.D. Bowen, F.O. Walker, R.R. Matsumoto, B. deCosta, K.C. Rice, Pharmacol. Rev. 42 (1990) 355.
[2] M. Hanner, F.F. Moebius, A. Flandorfer, H.-G. Knaus, J. Striessnig, E. Kempner, H. Glossmann, Proc. Natl. Acad. Sci. U.S.A. 93 (1996) 8072.
[3] O. Jbilo, H. Vidal, R. Paul, N. De Nys, M. Bensaid, S. Silve, P. Carayon, D. Davi, S. Galiègue, B. Bourrié, J.-C. Cuillemot, P. Ferrara, G. Loison, J.-P. Maffrand, G. Le Fur, P. Casellas, J. Biol. Chem. 272 (43) (1997) 27107.
[4] R. Kekuda, P.D. Prasad, Y.-J. Fei, F.H. Leibach, V. Ganapathy, Biochem. Biophys. Res. Commun. 229 (1996) 553.
[5] J. Maj, Z. Rogoz, G. Skuza, H. Mazela, Eur. J. Pharmacol. 315 (1996) 235.
[6] S.-I. Ogawa, S. Okuyama, H. Araki, S. Otomo, Eur. J. Pharmacol. 263 (1994) 9.
[7] S. Okuyama, Y. Imagawa, S.-I. Ogawa, H. Araki, A. Ajima, M. Tanaka, M. Muramatsu, A. Nakazato, K. Yamaguchi, M. Yoshida, S. Otomo, Life Sci. 53 (18) (1993) 285.
[8] S. Okuyama, A. Nakazato, CNS Drug Rev. 2 (2) (1996) 226.
[9] P.D. Shepard, H. Lehmann, S.T. Connelly, Eur. J. Pharmacol. 265 (1994) 141.
[10] W.D. Bowen, S.B. Hellewell, K.A. McGarry, Eur. J. Pharmacol. 163 (2/3) (1989) 309.
[11] E.W. Karbon, K. Naper, M.J. Pontecorvo, Eur. J. Pharmacol. 193 (1991) 21.
[12] R.G. Booth, S.D. Wyrick, R.J. Baldessarini, N.S. Kula, A.M. Myers, R.B. Mailman, Mol. Pharmacol. 44 (1993) 1232.
[13] R.G. Booth, S.D. Wyrick, Med. Chem. Res. 4 (1994) 225.
[14] D.L. DeHaven-Hudkins, L.C. Fleissner, Y.F.-R. Fleissner, Eur. J. Pharmacol. 227 (1992) 371.
[15] F.F. Moebius, R.J. Reiter, M. Hanner, H. Glossmann, Br. J. Pharmacol. 121 (1997) 1.
[16] F.F. Moebius, J. Striessnig, H. Glossmann, Trends Pharmacol. Sci. 18 (1997) 67.
[17] H.C. Akunne, S.Z. Whetzel, J.N. Wiley, A.E. Corbin, F.W. Ninteman, H. Tecle, Y. Pei, T.A. Pugsley, T.G. Heffner, Neuropharmacology 36 (1) (1997) 51.
[18] S. Chaki, M. Tanaka, M. Muramatsu, S. Otomo, Eur. J. Pharmacol. 251 (1994) R1.
[19] R.R. Matsumoto, W.D. Bowen, B.R. De Costa, Eur. J. Pharmacol. 280 (3) (1995) 301.
[20] K. Matsuno, M. Nakazawa, K. Okamoto, Y. Kawashima, S. Mita, Eur. J. Pharmacol. 306 (1996) 271.
[21] T. Senda, K. Matsuno, K. Okamoto, T. Kobayashi, K. Nakata, S. Mita, Eur. J. Pharmacol. 315 (1996) 1.
[22] F. Beradi, G. Giudice, R. Perrone, V. Tortorella, S. Govoni, L. Lucchi, J. Med. Chem. 39 (1996) 4255.
[23] B.R. de Costa, X.-S. He, J.T.M. Linders, C. Dominguez, Z.Q. Gu, W. Williams, W.D. Bowen, J. Med. Chem. 36 (1993) 2311.
[24] P.J. Gilligan, G.A. Cain, T.E. Christos, J. Med. Chem. 35 (23) (1992) 4344.
[25] R.L. Hudkins, R.B. Mailman, D.L. DeHaven-Hudkins, J. Med. Chem. 37 (1994) 1964.
[26] S. Mantegani, E. Brambilla, P. Cremonesi, C. Cacci, M.G. Fornaretto, N. Carfagna, M. Colombo, R.A. McArthur, M. Varasi, Bioorg. Med. Chem. Lett. 7 (12) (1997) 1525.
[27] W. Quaglia, M. Giannella, A. Piergentili, M. Pigini, L. Brasili, R.D. Toro, L. Rossetti, S. Spampinato, C. Melchiorre, J. Med. Chem. 41 (1998) 1557.
[28] R.N. Waterhous, K. Mardon, J.C. O'Brien, Nuclear Med. Biol. 24 (1997) 45.
[29] R.N. Waterhouse, K. Mardon, K.M. Giles, T.L. Collier, J. Med. Chem. 40 (1997) 1657.
[30] R.N. Waterhouse, T.L. Collier, Nuclear Med. Biol. 24 (1997) 127.
[31] F. Berardi, N.A. Colabufo, L. Lucchi, J. Med. Chem. 39 (1996) 176.

[32] B.R. De Costa, C. Dominquez, X.-S. He, J. Med. Chem. 35 (23) (1992) 4334.
[33] R.A. Glennon, S.Y. Ablordeppey, A.M. Ismaiel, M.B. El-Ashmawy, J.B. Fischer, K.B. Howie, J. Med. Chem. 37 (1994) 1214.
[34] A.H. Newman, J.H. Shah, S. Izenwasser, B. Heller, M. Mattson, F.C. Tortella, Med. Chem. Res. 6 (1996) 102.
[35] C.M. Bertha, M.V. Mattson, K.C. Rice, J. Med. Chem. 37 (19) (1994) 3163.
[36] F.I. Carroll, P. Abraham, K. Parham, X. Bai, X. Zhang, G.A. Brine, S.W. Mascarella, B.R. Martin, E.L. May, C. Sauss, L.Di. Paolo, P. Wallace, J.M. Walker, W.D. Bowen, J. Med. Chem. 35 (1992) 2812.
[37] G. Ronsisvalle, A. Marrazzo, O. Prezzavento, L. Pasquinucci, F. Vittorio, V. Pittala, M.S. Pappalardo, S. Cacciaguerra, S. Spampinato, J. Med. Chem. 41 (1998) 1574.
[38] H. Ucar, S. Cacciaguerra, S. Spampinato, K. Van derpoorten, M. Isa, M. Kanyonyo, J.H. Poupaert, Eur. J. Pharmacol. 355 (1997) 267.
[39] A. Yamashita, N. Takahashi, D. Mochizuki, R. tsujita, S. Yamada, H. Kawakubo, Y. Suzuki, H. Watanabe, Bioorg. Med. Chem. Lett. 7 (17) (1997) 2303.
[40] C.S. Dence, C.S. John, W.D. Bowen, M.J. Welch, Nuclear Med. Biol. 24 (1997) 333.
[41] M. Kassiou, V.H. Nguyen, R. Knott, M.J. Christie, T.W. Hambley, Bioorg. Med. Chem. Lett. 6 (6) (1996) 595.
[42] J.C. Jaen, B.W. Caprathe, T.A. Pugsley, L.D. Wise, H.J. Akunne, Med. Chem. 36 (1993) 3929.
[43] S.M.N. Efange, R.H. Mach, C.R. Smith, A.B. Khare, C. Foulon, S.K. Akella, S.R. Childer, S.M. Parsons, Biochem. Pharmacol. 49 (6) (1995) 791.
[44] Y. Zhang, W. Williams, W.D. Bowen, C. R. J. Med. Chem. (1996) 39, 3564.
[45] V.H. Nguyen, M. Kassiou, G.A.R. Johnston, M.J. Christie, Eur. J. Pharmacol. 311 (1996) 233.
[46] S.D. Wyrick, R.G. Booth, A.M. Myers, C.E. Owens, E.C. Bucholtz, P.C. Hooper, N.S. Kula, R.J. Baldessarini, R.B. Mailman, J. Med. Chem. 38 (1995) 3857.
[47] C.-C. Chien, F.I. Carroll, G.P. Brown, Y.-X. Pan, W. Bowen, G.W. Pasternak, Eur. J. Pharmacol. 321 (1997) 361.
[48] T.L. Collier, J.C. O'Brien, R.N. Waterhous, J. Labell. Compd. Radiopharm. XXXVIII (9) (1996) 785.
[49] C.-Y. Shiue, G.G. Shiue, S.X. Zhang, S. Wilder, J.H. Greenberg, F. Benard, J.A. Wortman, A.A. Alavi, Nuclear Med. Biol. 24 (1997) 671.
[50] H. Ujike, K. Akiyama, S. Kuroda, Neuropharmacol. Neurotoxicol. 7 (5) (1996) 1057.
[51] R.N. Waterhous, T.L. Collier, J.C. O'Brien, J. Labell. Compd. Radiopharm. XXXVIII (3) (1996) 215.
[52] S.Y. Ablordeppey, M.B. El-Ashmawy, R.A. Glennon, Med. Chem. Res. 1 (1992) 425.
[53] T.M. Gund, K. Shukla, J. Mathematical Chem. 8 (1991) 309.
[54] B.L. Largent, H. Wikstrom, A.L. Gundlach, S.H. Snyder, Mol. Pharmacol. 32 (1987) 722.
[55] D.T. Manallack, P.M. Beart, Eur. J. Pharmacol. 144 (1987) 231.
[56] D.T. Manallack, M.G. Wong, M. Costa, P.R. Andrews, P.M. Beart, Mol. Pharmacol. 34 (1988) 863.
[57] E.J. Lloyd, P.R. Andrews, J. Med. Chem. 29 (1986) 453.
[58] T.P. Su, X.Z. Wu, E.J. Cone, K. Shukla, T.M. Gund, A.L. Dodge, D.W. Parish, Multiple Σ And PCP Receptor Ligands, Proceedings of the Third Joint French–US Seminar, CNRS-NSF, September 15–19, 1991.