Pergamon

Bioorganic & Medicinal Chemistry Letters, Vol. 6, No. 2, pp. 189-194, 1996

0960-894X/96 $15.00 + 0.00

0960-894X(95)00573-0

STRUCTURE ACTIVITY RELATIONSHIPS OF TETRAHYDROCANNABINOL ANALOGUES ON HUMAN CANNABINOID RECEPTORS.

Yves Gareau,* Claude Dufresne, Michel Gallant, Chantal Rochette, Nicole Sawyer, Deborah M. Slipetz, Nathalie Tremblay, Philip K. Weech, Kathleen M. Metters and Marc Labelle.

Merck Frosst Centre for Therapeutic Research P.O. Box 1005, Pointe-Claire-Dorval, Québec, Canada, H9R 4P8.

Abstract: A series of Δ^8-tetrahydrocannabinol (THC) and biphenylic derivatives were prepared and their binding affinity for both human cannabinoid receptors hCB_1 and hCB_2 evaluated.

Δ^9-Tetrahydrocannabinol, an active component of marijuana, has been used for thousands of years in the treatment of a variety of ailments.[1] It possesses a wide range of physiological effects such as analgetic, appetite stimulant, anti-inflammatory, anti-convulsive, anti-emetic, immunosupressive and intraocular pressure lowering.[1,2] In spite of this therapeutic interest, its use has been limited by its psychotropic effects.[2] The search for analogues of medicinal value without the psychotropic effects has therefore received much attention.[3]

With the recent discovery of a second peripheral cannabinoid receptor,[4] it has been proposed that the psychotropic effects of cannabinoids may be mediated by the receptor expressed in the brain (CB_1), while some of the other beneficial properties may be associated with the peripheral receptor (CB_2). Given the structural differences between CB_1 and CB_2, which have both been cloned and expressed,[4,5] it should be possible to prepare selective hCB_2 ligands that might be valuable in the treatment of certain diseases. We now report our results on the SAR of some THC analogues.

All compounds[6] were evaluated in binding studies using the displacement of radiolabelled [^{3}H] (-) CP-55940[7] to determine the ligand potencies on hCB_2 and hCB_1. The compounds in this study were Δ^8-THC analogues containing a dimethylheptyl (DMH) chain, known to demonstrate increased potency[8] relative to the pentyl chain of Δ^9-THC in the classical CB_1 linked pharmacological assays as well as at the CB_1 receptor.

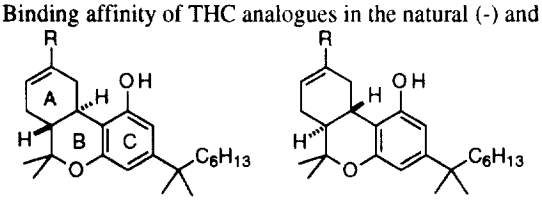
We paid special attention to the critical issue of the optical purity of cannabinoid ligands.[9] This turns out to be very important with some extremely potent ligands. Indeed, if such a ligand is present as a contaminant in its enantiomer, the K_i value obtained will be erroneous and directly linked to the optical purity of the batch under study. We were able to synthetize starting material and products at >97% optical purity using Brown's optical enrichment procedure on (+) and (-)-pinene.[10]

Table 1 shows the effect of chirality on receptor binding affinity of two pairs of enantiomers of THC analogues prepared according to reported procedure from (+) and (-)-pinene.[11] In both cases, the (-) enantiomer exhibited stronger binding than the (+) enantiomer for both the hCB_1 and hCB_2 receptor by at least 45 fold, consistent with previous reports. Unfortunately, the desired selectivity for hCB_2 over hCB_1 was found to be poor in both the (-) and in the (+) series.

A phenolic oxygen and a lipophilic chain are believed to be necessary for classical cannabinoid activity.[12] Table 2 shows the effect of modifications at the C-1 position on the affinity (K_i) at both receptors while keeping the DMH chain unaltered. For instance, transformation of the phenol **1** to the methyl ether **5**[13] resulted in a 40

fold loss in potency on hCB_2. On the other hand this loss in potency resulted in an increase in selectivity to 793 fold by virtue of the very weak binding on the hCB_1 receptor, as reported by Mechoulam *et al.*[14] An important effect on hCB_1 was also observed when the methoxy group of **5** was replaced by a hydrogen atom to give **7**.[15] The binding affinity increased from 15.9 μM to 0.25 μM, still 300 fold less potent than phenol **1**. Such a large effect on affinity caused by a small structural change may indicate that a region of the hCB_1 receptor was involved in interactions such as hydrogen bonding and/or steric effects with the ligand. Interestingly, the K_i of hCB_2 was not affected by this change. The phosphate **6**[16] was not very active, particularly on hCB_1. A K_i value of 0.44 μM was obtained on CB_2 and >20 μM for CB_1. In this particular case, polarity and steric factors certainly played an important role. We concluded from the results of Table 2 that simple modification of the phenol to a methoxy group gave a less potent but selective CB_2 cannabinoid.

Table 1: Binding affinity of THC analogues in the natural (-) and the unnatural (+) series.

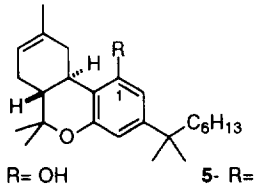


(-)-**1** R= Me (+)-**2**
(-)-**3** R= CHO (+)-**4**

	K_i (hCB_2, nM)[a]	K_i (hCB_1, nM)[a]	hCB_1/hCB_2
1-	0.49 ± 0.18	0.83 ± 0.13	1.7
2-	28.8 ± 6.4	38.8 ± 6.1	1.3
3-	2.61 ± 0.36	2.24 ± 0.05	0.9
4-	118.6 ± 30.8	97.0 ± 6.1	0.8

a) Values are mean ± S.E.M. or individual determination.

Table 2: Binding affinity of THC analogues bearing various groups at C-1.



1- R= OH **5**- R= OCH_3
6- R= $OP(O)(OC_2H_5)_2$ **7**- R= H

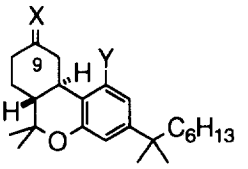
	K_i (hCB_2, nM)[a]	K_i (hCB_1, nM)[a]	hCB_1/hCB_2
1-	0.49 ± 0.18	0.83 ± 0.13	1.7
5-	20.0 ± 12.4	15850 ± 2960	793
6-	441.7 ± 74.8	>20 000, >5000	>45
7-	20.8 ± 11.2	249.7 ± 31.0	12

a) Values are mean ± S.E.M. or individual determination.

Nabilone **8**[12,17] and related compounds were prepared and the binding data are shown in Table 3. Nabilone differs structurally from Δ^9-THC and **1** by the presence of a carbonyl group at C-9. Compound **8** was very potent, but not selective with a $K_i \approx 2$ nM for both receptors. In fact, **8** is very similar to **3** with respect to binding affinity on both receptors, in spite of the carbonyl being moved one carbon atom away. The methylene analogue **10**[13] had similar potency for both receptors. On the other hand, transformation of the phenolic function of **10** to a methyl ether (**11**)[13] resulted in a loss of potency on hCB_1, leading to a high degree of selectivity (>1000), of the same order of magnitude as observed in the case of **5**. Compounds **5** and **11** actually only differ by the position of the double bond on the A ring. We concluded from this, that the position of the double bond was not critical to activity at hCB_2 in this series.

The methylation of nabilone to the ether **9**[17] resulted in a small increase in selectivity, but in a 72 fold loss in binding affinity for hCB_2 and a 284 fold loss on hCB_1. In the series of cannabinoids of Table 3, compound **11** stands out as a potent and selective hCB_2 binder.

Table 3: Binding affinity of THC analogues in the nabilone and exo-methylene series.



8, 9, 10, 11

	X	Y	K_i (hCB_2, nM)[a]	K_i (hCB_1, nM)[a]	hCB_1/hCB_2
8-	O	OH	1.84 ± 0.42	2.19 ± 0.89	1.2
9-	O	OCH_3	132.2 ± 44.3	621 ± 215	4.7
10-	CH_2	OH	0.58 ± 0.30	1.82 ± 0.11	3.1
11-	CH_2	OCH_3	19.4 ± 3.8	> 20 000, > 20 000	>1000

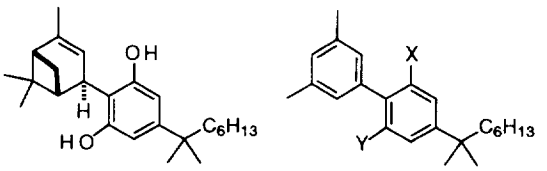
a) Values are mean ± S.E.M. or individual determination.

The radiolabelled CP-55940 used to determine the ligand potencies, is devoid of the rigid tricyclic ring system[18] common to the THC family, and we decided to test some non-tricyclic analogues. Compound **12**[19] used to prepare the tricyclic ring system, was found to be particularly potent at hCB_2 with a K_i of 41 nM (9 fold selective, Table 4). Replacement of the terpenyl moiety of **12** by a planar template that better approximates the A ring, such as a 3,5-dimethylphenyl group, greatly simplifies the molecule as well as the synthesis of potential analogues. This change resulted in stronger binding of **13** *vis-à-vis* **12** to both receptors and the net result was an improved selectivity for hCB_2. In order to evaluate the effectiveness of the phenol as hydrogen bonding donor, **13** was converted to dimethyl ether **16.** This modification led to a significant loss of the binding affinity on hCB_2 (ca. 200 fold), with a K_i of 433nM. Considering the structural simplicity of **16**, such binding is still

noteworthy. For the hCB_1 receptor, this modification resulted in an inactive compound.

The selectivity was lost by replacement of one OH group by a hydrogen atom (**14**). Mainly due to improved CB_1 binding. Removal of both OH groups yielded hydrocarbon **15**, which was essentially inactive at both receptors.

Table 4: Binding affinity of bicyclic and biphenylic analogues.



12 **13,14,15,16**

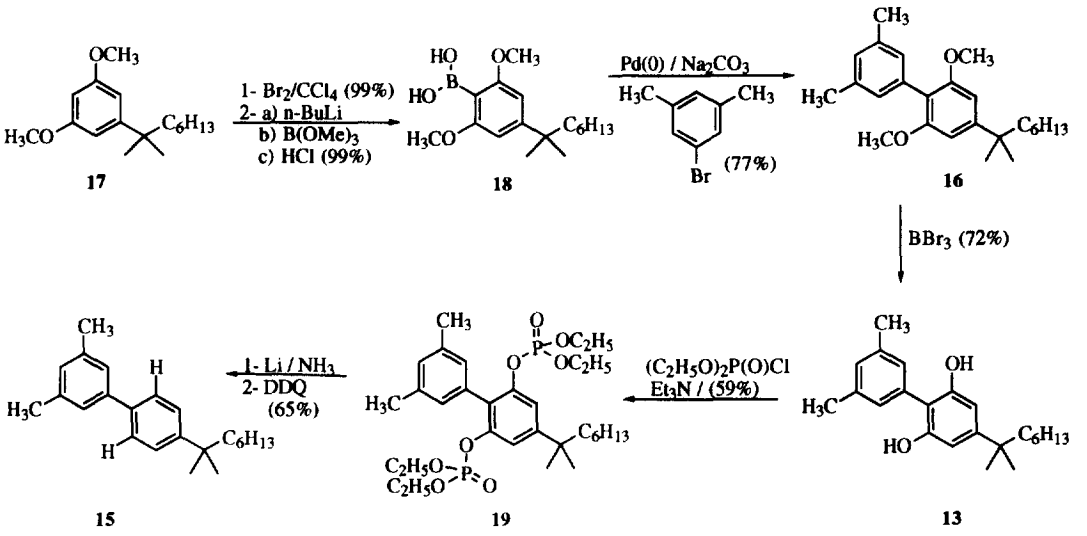
	X	Y	K_i (hCB_2, nM)[a]	K_i (hCB_1, nM)[a]	hCB_1/hCB_2
12-	-	-	41.2 ± 14.1	350.3 ± 41.4	9.0
13-	OH	OH	2.00 ± 1.08	79.1 ± 12.5	40
14-	OH	H	4.14 ± 2.51	12.5 ± 1.8	3.0
15-	H	H	2138 ± 1317	> 30 000, > 30 000	>14
16-	OCH_3	OCH_3	433 ± 203	> 20 000, > 20 000	>46

a) Values are mean ± S.E.M. or individual determination.

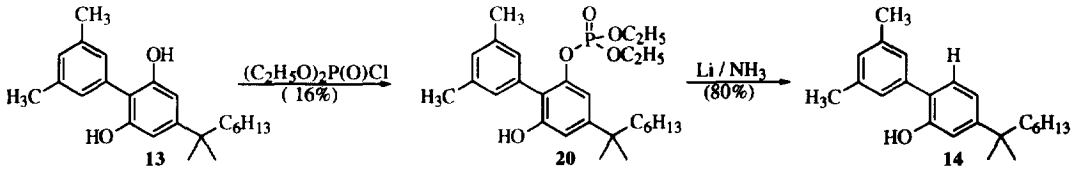
Compounds (**13-16**) were prepared according to Schemes 1 and 2. The bis-ether **17** was brominated[20] and converted to the boronic acid **18** via a metal-halogen exchange reaction with n-butyllithium. A Suzuki coupling reaction with 5-bromo-*m*-xylene gave **16**[21] in an overall yield of 75%. Demethylation[11a] of the bis-ether was performed with BBr_3 at 0° C to furnish **13** in 72% yield. Compound **15**[11a] was obtained by the formation of diphosphonate **19** followed by reduction with Li/NH_3 and reoxidation of the aromatic ring with DDQ (because of partial overreduction) in 38% yield for the 3 steps. Finally, compound **14** was prepared in a manner similar to that used for the synthesis of **15**, by formation of monophosphonate **20** in 16% yield and reduction with Li/NH_3 in 80% yield.

In conclusion, we have shown that in the naturally occuring Δ^8-THC stereochemical series, analogues such as **5** and **11** are very selective for the hCB_2 receptor regardless of the position of the exo or endo double bond. It was preferable not to have the free OH group at the C-1 position to obtain hCB_2 selective compounds. When the hydroxyl group (**1**) was replaced by a methoxy group (**5**), significant binding was retained at the hCB_2 receptor while the selectivity toward hCB_2 increased to 1000 fold. We have demonstrated, with non-cannabinoid type structures like **12-14** that the tricyclic moiety is not necessary to reach nM affinity, and that such compounds may have high selectivity.

Scheme 1



Scheme 2



References:

1. Mestel, R. *New Scientist*, **1993**, *139*, 21.
2. Dewey, W.L. *Pharmacol. Rev.*, **1986**, *38*, 151.
3. Razdan, R.K. *Pharmacol. Rev.*, **1986**, *38*, 75; Devane, W.A.; Breuer, A.; Sheskin, T.; Järbe, T.U.C.; Eisen, M.S.; Mechoulam, R. *J. Med. Chem.*, **1992**, *35*, 2065.
4. Munro, S.; Thomas, K. L.; Abu-Shaar, M. *Nature*, **1993**, *365*, 61.
5. Matsuda, L.A.; Lolait, S.J.; Brownstein, M.J.; Young, A.C.; Bonner, T.I. *Nature*, **1990**, *346*, 561.
6. Satisfactory analytical data for all compounds have been obtained.
7. Johnson, M.R.; Melvin Jr, L.S.; **1983** U.S. Patent 4 371 720.
8. Reggio, P.H.; Panu, A.M.; Miles, S. *J. Med. Chem.*, **1993**, *36*, 1761; Adams, R.; Harfenst, M.; Lowe, S. *J. Amer. Chem. Soc.* **1949**, *71*, 1624.
9. Martin, B.R.; Balster, R.L.; Razdan, R.K.; Harris, L.S.; Dewey, W.l. *Life Sciences*; **1981**, *29*, 565; Mechoulam, R.; Lander, N.; Breuer, A.; Zahalka, J. *Tetrahedron: Asymmetry*, **1990**, *1*, 315.

10. Brown, H.C.; Joshi, N.N. *J. Org. Chem.*, **1988**, *53*, 4059. Both enantiomers are now commercially available at 97% ee.
11. (a) Dominianni, S.J.; Ryan, C.W.; DeArmitt, C.W. *J. Org. Chem.*, **1977**, *42*, 344. (b) Cooper, M.A.; Salmon, J.R.; Whittaker, D.; Scheidegger, U. *J. Chem. Soc. (B)*, **1967**, 1259. (c) Fallis, A.G. *Can. J. Chem.*, **1975**, 1657; (d) Burstein, S.H.; Audette, C.A.; Breuer, A.; Devane, W.A.; Colodner, S.; Doyle, S.A.; Mechoulam, R. *J. Med. Chem.*, **1992**, *35*, 3135 (e) The same procedure was used for the preparation of the DMH as for the preparation of the pentyl chain: Mechoulam, R.; Braun, P.; Gaoni, Y. *J. Amer. Chem. Soc.*, **1972**, *94*, 6159;.
12. Stark, P.; Archer, R.A. *Pharmacologist*, **1975**, *17*, 210.
13. We used the procedure described for a similar compound by: Wildes, J.W.; Martin, N.H.; Pitt, C.G.; Wall, M.E. *J. Org. Chem.*, **1971**, *36*, 721.
14. Edery, H.; Grunfeld, Y.; Ben-Zvi, Z.; Mechoulam, R. *Ann. N.Y. Acad. Sci.* **1971**, *191*, 40; Reggio, P.H.; Seltzman, H.H.; Compton, D.R.; Prescott, Jr, W.R.; Martin, B.R. *Mol. Pharmacol.* **1990**, *38*, 854.
15. Compound **7** was prepared by treatment of **6** with Li/NH_3 according to ref. 10a.
16. Compound **6** was prepared by treatment of **1** with diethyl chlorophosphonate according to ref. 10a.
17. Huffman, J.W.; Joyner, H.H.; Lee, M.D.; Jordan, R.D.; Pennington, W.T. *J. Org. Chem.*, **1991**, *56*, 2081.
18. CP-55940 is a non-tricyclic ring system. Melvin, L.S.; Milne, G.M.; Johnson, M.R.; Subramaniam, B.; Wilken, G.H.; Howlett, A.C. *Mol. Pharmacol.* **1994**, *44*, 1008.
19. Mechoulam, R.; Lander, N.; Dikstein, S.; Shalita, B.; **1981** *Canadian Patent*, 1 114 828.
20. Srebnik, M.; Mechoulam, R.; Yona, I. *J. Chem. Soc. Perkin Trans. 1*, **1987**, 1423.
21. Fu, J.; Snieckus, V. *Tetrahedron Lett.*, **1990**, *31*, 1665. Alo, B.I.; Kandil, A.; Patil, P.A.; Sharp, M.J.; Siddiqui, M.A.; Snieckus, V. *J. Org. Chem.*, **1991**, *56*, 3763.

(*Received in USA* 27 *October* 1995; *accepted* 7 *December* 1995)