Available online at www.sciencedirect.com
ScienceDirect
Bioorganic & Medicinal Chemistry Letters 17 (2007) 272–277
Bioorganic & Medicinal Chemistry Letters

Arylsulfonamides as a new class of cannabinoid CB_1 receptor ligands: Identification of a lead and initial SAR studies

N. Lambeng,[a] F. Lebon,[b] B. Christophe,[c] M. Burton,[b] M. De Ryck[d] and L. Quéré[b,*]

[a]*CNS Molecular and Cellular Biology, Centre for CNS Innovation, UCB S.A., Chemin du Foriest, B-1420 Braine L'Alleud, Belgium*
[b]*Global Chemistry, Centre for CNS Innovation, UCB S.A., Chemin du Foriest, B-1420 Braine L'Alleud, Belgium*
[c]*Non clinical development, Centre for CNS Innovation, UCB S.A., Chemin du Foriest, B-1420 Braine L'Alleud, Belgium*
[d]*Neuropharmacology R&D, Centre for CNS Innovation, UCB S.A., Chemin du Foriest, B-1420 Braine L'Alleud, Belgium*

Received 16 August 2006; revised 15 September 2006; accepted 16 September 2006
Available online 5 October 2006

Abstract—High-throughput screening of the UCB sample collection identified the piperidinyl-sulfonyl benzoic ester **1** as a novel agonist for CB_1 receptor with nanomolar affinity. We report here the pharmacological profile of compound **1** as well as preliminary biological activities in pain model. Diverse close analogs of **1** were purchased and the structure–affinity relationships among this novel class are discussed.

The endocannabinoid system appears to be involved in a rising number of pathological conditions and hence represents an exciting target for drug discovery.[1,2] The biological properties associated with the endogenous ligands (several unsaturated fatty acid derivatives) are mainly mediated by two G-protein coupled receptors, that is, CB_1 and CB_2 receptors. While CB_2 seems to be restricted to the spleen and the immune system, CB_1 is widely expressed in the central nervous system and, to a lesser extent, in the periphery (adipose tissue, gastric tract).[3] In the brain, endocannabinoids behave as retrograde signaling messengers that stimulate presynaptic CB_1 receptors on neurons. This activation results in inhibition of adenylate cyclase activity, regulation of ion channel activities and activation of the mitogen-activated protein kinase cascade.[4]

Potential therapeutic application for CB_1 agonists mainly lies in developing drugs for treating pain (chronic and acute), multiple sclerosis, tremor, anxiety/mood disorders, sleep disorders, seizures, and neuroprotection. Two products are already available on this growing (yet controversial) market, namely Marinol and Nabilone. In late 2006, a third one, Sativex, will begin phase III trials in the US for cancer patients. Other numerous cannabinoid receptor modulators have been identified including non-selective CB_1/CB_2 agonists such as WIN 55,212-2 and CP 55,940. Thus far, more than five structurally diverse sets of cannabinergic ligands have been discovered, including classical cannabinoids, non-classical cannabinoids, aminoalkylindoles, eicosanoids, and arylpyrazoles.[5]

Our interest in the discovery and development of novel therapeutics for brain disorders prompted a high-throughput screen of our chemical collection. This effort led to the discovery of compound **1** (Fig. 1) which exhibited high-affinity binding to recombinant human CB_1 with a pIC_{50} value of 9.1 ± 0.5 (Table 1). Affinity of compound **1** was 3- or 80-fold higher than CP 55,940 and WIN 55,212-2, respectively. Contrary to these two latter compounds that have a similar affinity for both cannabinoid receptors (or even greater for CB_2 for WIN 55,212-2), compound **1** exhibited a 1- to 1.5-log selectivity toward CB_2. Moreover, compound **1** was highly selective against 50 other diverse GPCRs, channels or enzymes (data not shown).

The efficacy of compound **1** was determined by measuring [^{35}S]-GTPγS binding to membranes of CHO cells expressing the human recombinant CB_1.[6] Its efficacy in increasing the binding of [^{35}S]-GTPγS was similar to reference compounds WIN 55,212-2 and CP 55,940 (Fig. 2), and thus it was characterized as a full agonist.

Keywords: CB1; Pain; CB1 agonist; Cannabinoids; Arylsulfonamide.
* Corresponding author. Tel.: +3223862714; fax: +3223862704; E-mail: luc.quere@ucb-group.com

0960-894X/$ - see front matter © 2006 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2006.09.049

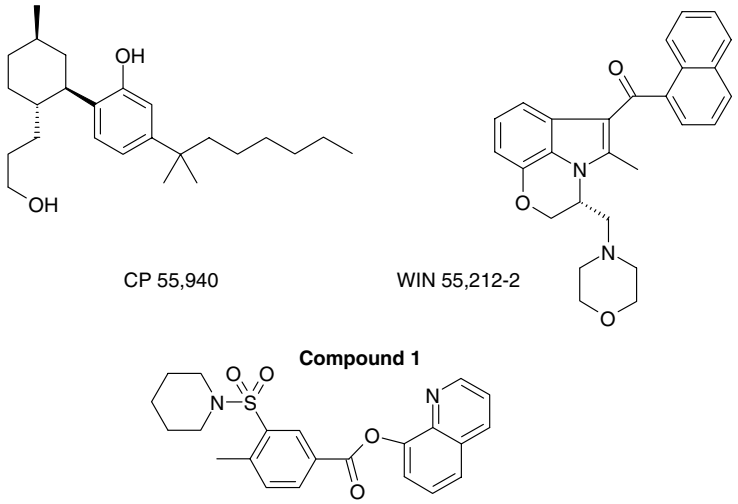


Figure 1. Molecular structures of CP 55,940, WIN 55,212-2, and compound **1**.

Table 1. Affinity and efficacy of compound **1** at human recombinant cannabinoid receptors compared to reference compounds

Compound	pIC_{50} CB_1 receptor[a]	[^{35}S]-GTPγS pEC_{50} CB1 receptor[a]	pK_i CB_2 receptor
WIN 55,212-2	7.2 ± 0.4	6.5 ± 0.4	8.7–9.4[b]
CP 55,940	8.6 ± 0.3	6.8 ± 0.4	8.7–9.3[b]
1	9.1 ± 0.5	8.0 ± 0.4	7.6

[a] Values are expressed as means ± standard deviation of at least four independent experiments.
[b] From the literature 7.

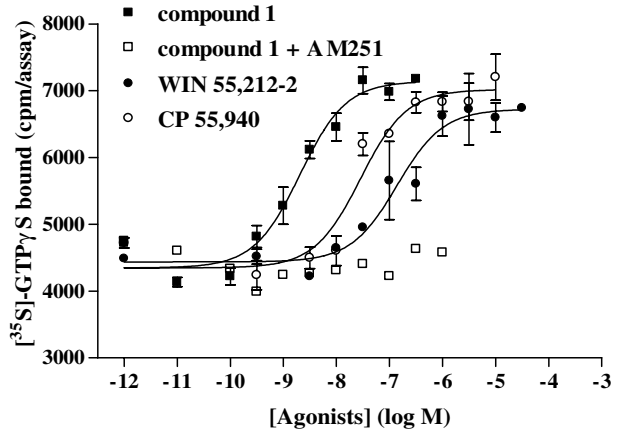


Figure 2. Activation by WIN 55,212-2 (●), CP 55,940 (○), and compound **1** in the presence (□) or not (■) of AM251 (10^{-5} M) of the binding of [^{35}S]-GTPγS to membranes of CHO cells expressing human recombinant CB_1. Data are reported as means ± SD and are representative of at least four independent experiments.

This agonist activity was totally antagonized by the CB1 antagonist AM251 at 10^{-5} M (Fig. 2).

Furthermore, compound **1** was more potent than the two reference derivatives (i.e., it had a higher pEC_{50}; see Table 1).

In terms of tissular activity, cannabinoid CB_1 agonists inhibit the amplitude of the twitch contractions electrically induced on the isolated mouse vas deferens via pre-junctional inhibition of neurotransmitter release from peripheral autonomic nerve terminal.[8] In this paced isolated mouse vas deferens model, compound **1** behaved as a full agonist equipotent to WIN 55,212-2 (Fig. 3). The calculated pD_2 values are reported in Table 2. Compound **1** induced a sub-maximal inhibition (87% at 10^{-5} M) of the amplitude of this twitch contraction.

The agonist activity induced with WIN 55,212-2 or compound **1** was competitively antagonized by various CB_1 antagonists such as SR 141716A (calculated pA_2 values of 7.63 ± 0.24 (n = 11) and 7.67 ± 0.86 (n = 5) using

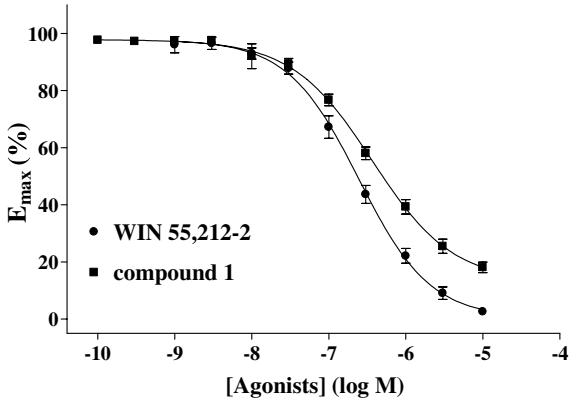


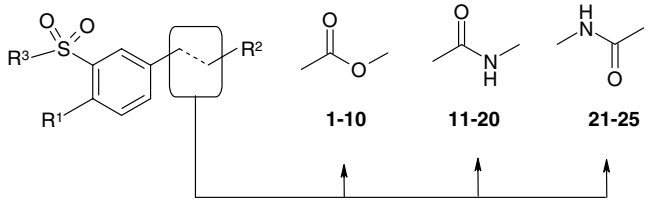
Figure 3. Concentration–response curves to WIN 55,212-2 and compound **1** elicited on the paced isolated mouse vas deferens. Data are reported as means ± SEM (n = 14–15).

Table 2. pD_2 and E_{max} calculated for compound **1**

Compound	Agonism	
	pD_2	Intrinsic activity versus WIN 55,212-2
WIN 55, 212-2	6.64 ± 0.27 (15)	100
1	6.42 ± 0.22 (14)	87.27 ± 7.85 (14)

Values are expressed as means ± SD of 14–15 independent experiments. pD_2 values calculated according to Van Rossum et al.[8]

Table 3. Affinity and efficacy of compound **1** and its derivatives at human recombinant cannabinoid CB_1 receptor



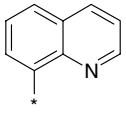
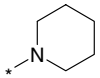
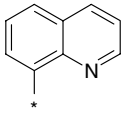
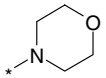
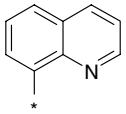
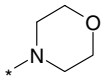
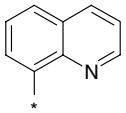
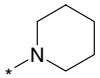
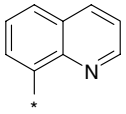
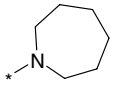
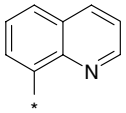
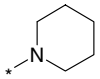
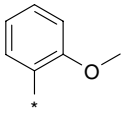
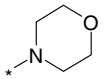
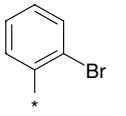
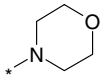
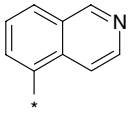
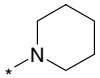
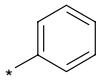
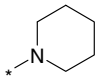
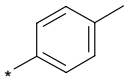
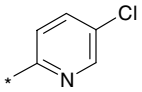
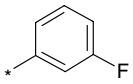
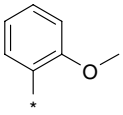
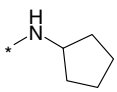
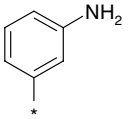
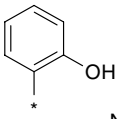
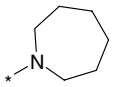
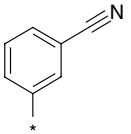
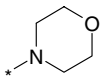
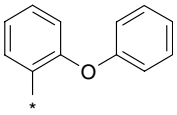
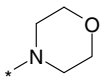
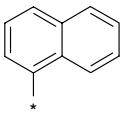
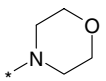
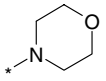
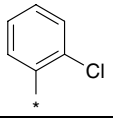
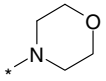
Compound	R^1	R^2	R^3	pIC_{50} CB_1 receptor	[^{35}S]-GTPγS pEC_{50} CB_1 receptor
1	Me			9.1	8.0
2	Me			8.9	7.9
3	OMe			7.4	6.4
4	H			7.8	7.3
6	H			7.3	6.7
7	H			6.8	6.2
8	Me	H		<5	<5
9	Me			5.5	7.2
10	Me			7.5	6.4
11	Me			5.9	5.2
12	Me			5.5	<5

Table 3 (*continued*)

Compound	R^1	R^2	R^3	pIC_{50} CB_1 receptor	[^{35}S]-GTPγS pEC_{50} CB_1 receptor
13	Me			5.1	<5
14	Me			5.5	<5
15	Me			5.9	<5
16	Me			5.2	<5
17	H			<5	NT
18	H			5.3	<5
19	H			5.3	<5
20	Cl			6.7	5.9
21	Me			5.8	5.3
22	Me			5.7	<5
23	Cl			6.4	6.1
24	Cl			5.4	6.2
25	Cl			5.8	<5

NT, not tested.

WIN 55,212-2 or compound **1** as agonist, respectively) or AM 251 (calculated pA_2 values of 7.86 ± 0.31 ($n = 12$) and 7.45 ± 0.44 ($n = 11$) using WIN 55,212-2 or compound **1** as agonist, respectively). Conversely, 10^{-5} M JTE907 (CB_2 antagonist) was unable to modify the concentration–response curve elicited with WIN 55,212-2 or compound **1** on the paced isolated mouse vas deferens.

Several close analogs to compound **1** were selected and purchased from commercial databases to derive a preliminary SAR around this compound (Table 3). All compounds were found to be less active than compound **1**. In the series containing an ester linker (**1**–**10**), replacement of the *N*-piperidine ring in R^3 with a *N*-morpholine (**2**) is well tolerated. Compounds **6** and **7** indicate that increasing or decreasing the size of the *N*-piperidine ring drastically reduces the activity toward the CB_1 receptor. Removal of the Me group in R^1 or replacement with a methoxy group is also detrimental to the activity. Removal of the quinoline ring in R^2, or introduction of a methoxy-phenyl group further reduces the affinity (compounds **8** and **9**), whilst reintroduction of a hydrophobic group like a bromophenyl group (compound **10**) partially restores binding to the CB_1 receptor. Replacement of the ester moiety with an amide leads to a dramatic loss of potency in most cases (**11**–**25**). Unfortunately, for the sake of comparison, the exact amide analog of compound **1** was not found in external libraries.

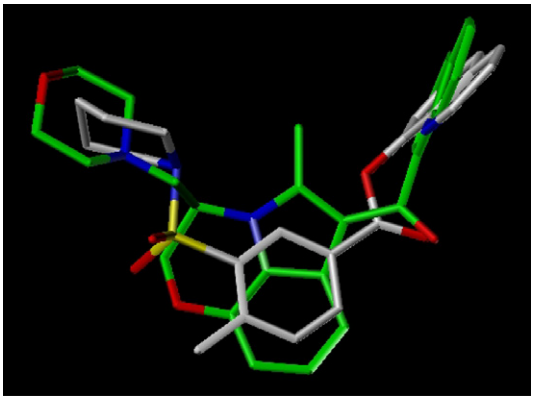


Figure 4. Superimposition of WIN 55,212-2 and compound **1** low energy conformations (Sybyl7.1). Carbon atoms of WIN 55,212-2 are colored in green.

The most potent compounds in the amide series are compounds **20** and **23**. These compounds have a chlorine in place of the Me group. Due to the expected gain in stability with the amide linker versus the ester linkage, those two derivatives may represent interesting hits for optimizing the original scaffold.

The literature provides substantial information on the stereo electronic requirements for CB_1 receptor recognition and activation.[9] The most remarkable features comprise two hydrophobic binding pockets (one of limited depth and one larger) and a hydrogen bonding site for the phenolic hydroxyl group present on the classical cannabinoids. Compound **1** does not bear any hydrogen donor element and thus, from a structural point of view, seems to belong to the non-classical cannabinoid family typified by WIN 55,212-2. Moreover, by superimposing compound **1** and WIN 55,212-2, we found a good steric overlap and a reasonably close matching of pharmacophoric elements like hydrogen bond acceptor areas and π-electron-rich areas (Fig. 4).

In vivo activity. Intraperitoneal administration of compound **1** moderately reduced nociception in both early and late phases of the mouse formalin test (Fig. 5). In either case, the lowest active dose was 30 mg/kg. Anti-hyperalgesic effects within active dose range (30–180 mg/kg) did not statistically differ from each other (no dose-dependency) and hovered around 50% and 60% for the early and late phases, respectively. It is conceivable that suboptimal drug metabolism is responsible for this activity pattern. Interestingly, only at the highest dose (180 mg/kg ip) did compound **1** induce hypokinesia with reduced postural tone. However, animals remained normothermic and showed no clear signs of catalepsy. In the late phase of the mouse

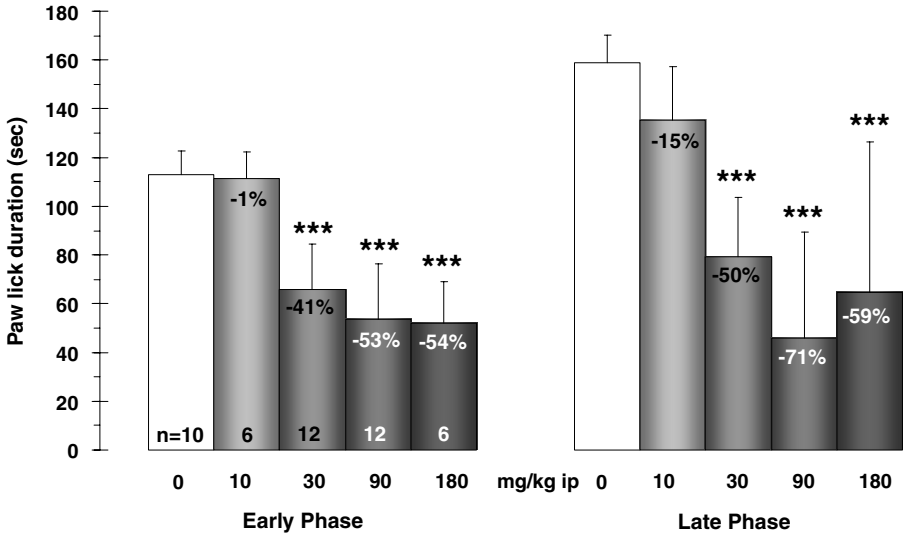


Figure 5. Mouse formalin test (male NMRI mice, 25–30 g; 1.5% formalin, 25 μL, hindpaw, intraplantar). Effects of compound **1** (−30 min, ip) on duration of paw licking response (s) in early (0–5 min) and late (10–30 min) phases of nociception. Dunnett's test: $^{***}p < 0.001$.

formalin test, the CB_1 receptor agonist WIN 55,212-2 showed an ED_{50} value of 0.21 mg/kg (4.0×10^{-7} mol/kg), but hypokinesia and hypotonia were present from 0.3 mg/kg onwards.

Conclusions. In summary, we report the discovery of a novel class of CB_1 agonists, with compound **1** active in the low nanomolar range, behaving as a full agonist and more potent, in vitro, than current reference compounds. This derivative is highly selective for the CB receptors and has been shown to be biologically active in the mouse formalin test, without inducing any side effects associated with classical cannabinoid agonists. However, the modest in vivo activity observed in this model contrasts with the high in vitro potency and suggests that compound **1** displays suboptimal PK properties. None of the amide analogs that we purchased were found to be as active as compound **1**, but some interesting hits were nevertheless unraveled.

Acknowledgments

The authors thank B. Carlier and M. Grossmann for technical expertise as well as Dr. C. Genicot, Dr. F. Moureau, and Dr. C. Audouin for their careful revision of the manuscript.

References and notes

1. Lambert, D. M.; Fowler, C. J. *J. Med. Chem.* **2005**, *48*, 5059.
2. Piomelli, D. *Curr. Opin. Invest. Drugs* **2005**, *6*, 672.
3. Herkenham, M. In *Cannabinoid Receptors*; Pertwee, R. G., Ed.; Academic Press: New York, 1995; p 145.
4. (a) Matsuda, L. A.; Lolait, S. J.; Brownstein, M. J.; Young, A. C.; Bonner, T. I. *Nature* **1990**, *346*, 561; (b) Gerard, C. M.; Mollereau, C.; Vassart, G.; Parmentier, M. *Biochem. J.* **1991**, *279*, 129; (c) Mackie, K.; Lai, Y.; Westenbroek, R.; Mitchell, R. *J. Neurosci.* **1995**, *15*, 6552; (d) Bouaboula, M.; Poinot-Chazel, C.; Bourrie, B.; Canat, X.; Calandra, B.; Rinaldi-Carmona, M.; Le Fur, G.; Casellas, P. *Biochem. J.* **1995**, *312*, 637.
5. Pertwee, R. G. *Curr. Med. Chem.* **1999**, *6*, 635.
6. The [^{35}S]-GTPγS binding assay was carried out in 96-well microplates in 50 mM Tris–HCl, pH 7.4, containing 50 mM NaCl, 10 mM $MgCl_2$, 5 μM GDP, 0.5% BSA, 10 μg/ml saponin, 20 μg protein, and 0.25 nM [^{35}S]-GTPγS. Non-specific binding was determined in the presence of 100 μM GppNHp. Incubations were performed for 90 min at 25 °C and terminated by rapid vacuum filtration onto Unifilter plate using the Unifilter system and 4 washes with 50 mM Tris, pH 7.4, containing 0.05% BSA. Unifilter plates were then dried for 1 h at 60 °C before adding 30 μL/well of Microscint-20, and radioactivity counted using a TopCount reader.
7. (a) Mauler, F.; Mittendorf, J.; Horvath, E.; De Vry, J. *J. Pharmacol. Exp. Ther.* **2002**, *302*, 359; (b) MacLennan, S. J.; Reynen, P. H.; Kwan, J.; Bonhaus, D. W. *Br. J. Pharmacol.* **1998**, *124*, 619.
8. (a) Rinaldi-Carmona, M.; Barth, F.; Heaulme, M.; Shire, D.; Calandra, B.; Congy, C.; Martinez, S.; Maruani, J.; Neliat, G.; Caput, D.; Ferrara, P.; Soubrie, P.; Breliere, J.-C.; Le Fur, G. *FEBS Lett.* **1994**, *350*, 240; (b) Lay, L.; Angus, J. A.; Wright, C. E. *Eur. J. Pharmacol.* **2000**, *391*, 151; (c) Van Rossum, J. M.; Hurkmans, J. A. T. M.; Wolters, C. J. J. *Arch. Int. Pharmacodyn. Ther.* **1963**, *143*, 299.
9. Reggio, P. H. *Curr. Pharm. Des.* **2003**, *9*, 1607.