Notes

Pharmacophoric Requirements for Cannabinoid Side Chains: Multiple Bond and C1′-Substituted Δ^8-Tetrahydrocannabinols

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Accumulated evidence indicates that within the cannabinoid structure the aliphatic side chain plays a pivotal role in determining cannabimimetic activity. We describe the synthesis and affinities for the CB1 and CB2 receptors of a series of novel Δ^8-THC analogues in which the side-chain pharmacophores are conformationally more defined than in the parent molecule. No analogue has the side-chain pharmacophore in a fully restricted conformation. However, our design serves to narrow down the scope of options for conformational requirements at the receptor active sites. All the analogues tested showed nanomolar or subnanomolar affinities for the receptors; 2-(6a,7,10,10a-tetrahydro-6,6,9-trimethyl-1-hydroxy-6*H*-dibenzo[*b,d*]pyranyl)-2-hexyl-1,3-dithiolane was found to possess very high affinity for both cannabinoid receptors (CB1, K_i = 0.32 nM; CB2, K_i = 0.52 nM).

Introduction

The identification of a cannabinoid receptor[1] (CB1) in mammalian brains and its further characterization as being G-protein-coupled[2] have been the topic of intense investigation leading to its cloning from rat[3] and human[4] cDNA libraries. In addition, efforts aimed at elucidating the physiological role of the cannabinoid receptor resulted in the isolation and identification of arachidonylethanolamide (anandamide) as a putative endogenous ligand.[5] More recently, a new peripheral cannabinoid receptor was cloned and expressed in macrophages in the marginal zone of the spleen (CB2).[6]

In an effort to obtain more detailed information on the regiochemical and stereochemical requirements for productive binding at the active site within a specific class of cannabimimetic (CBMM) agents, we have sought to develop novel ligands possessing high affinity and selectivity for the cannabinoid receptors. Accumulated evidence indicated that within the cannabinoid structure the aliphatic side chain plays a pivotal role in determining CBMM activity. Structural variations within this pharmacophore can result in analogues varying by up to 3 orders of magnitude in affinity for the receptor and in pharmacological potency.[7] The present paper aims at exploring, in part, the side chain's stereoelectronic requirements for activity within the classical tetrahydrocannabinol (THC) structure. More specifically and following earlier leads,[8] we have focused on the C1′ position of the side chain and explored the role of different substituents on the affinities of these analogues for the CB1 and CB2 cannabinoid receptors.

(−)-Δ^8-Tetrahydrocannabinol (Δ^8-THC) has a very similar pharmacologic profile as (−)-Δ^9-tetrahydrocannabinol (Δ^9-THC), the most active constituent of cannabis, and was chosen as the prototype in this series because of its greater chemical stability. On the basis of earlier literature reports,[7] the length of the side chain was optimized to seven carbon atoms.

In this paper we seek to extend work we recently published in which the cannabinoid side chain was partially immobilized through the introduction of multiple carbon−carbon bonds between its first two C1′ and C2′ carbons.[9] We now describe the synthesis and affinities for the CB1 and CB2 receptors of a series of novel Δ^8-THC analogues in which the side-chain pharmacophores are conformationally more defined than in the parent molecule. No analogue has the side-chain pharmacophore in a fully restricted conformation. However, our design serves to narrow down the scope of options for conformational requirements at the receptor active sites. One of the analogues synthesized, namely, 2-(6a,7,10,10a-tetrahydro-6,6,9-trimethyl-1-hydroxy-6*H*-dibenzo[*b,d*]pyranyl)-2-hexyl]-1,3-dithiolane (**5**), was found to possess very high affinity for the cannabinoid receptor.

Chemistry

Generally, the synthesis of side-chain congeners of Δ^8-THC was accomplished by the condensation of a monoterpene with an appropriately substituted resorcinol. For the synthesis of (−)-(1-heptynyl)-Δ^8-THC (**1**) (Chart 1), we followed a modification of the method described in the literature[10] and condensed 5-(1-heptynyl)resorcinol (**9**) with (+)-*cis*/*trans*-*p*-mentha-2,8-dien-1-ol[11] in the presence of *p*-toluenesulfonic acid. On the other

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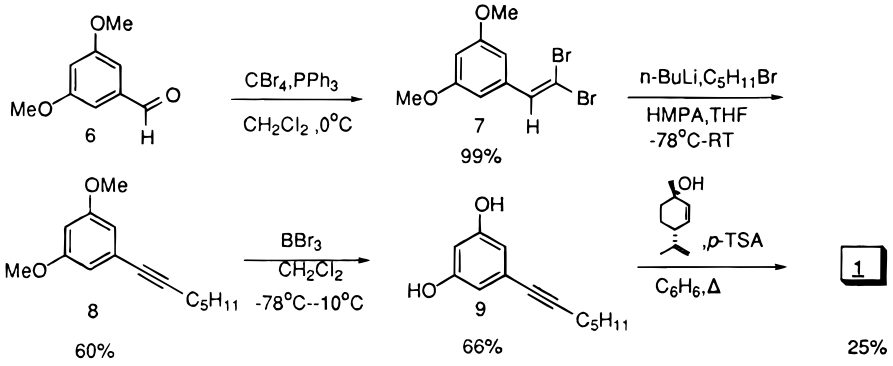
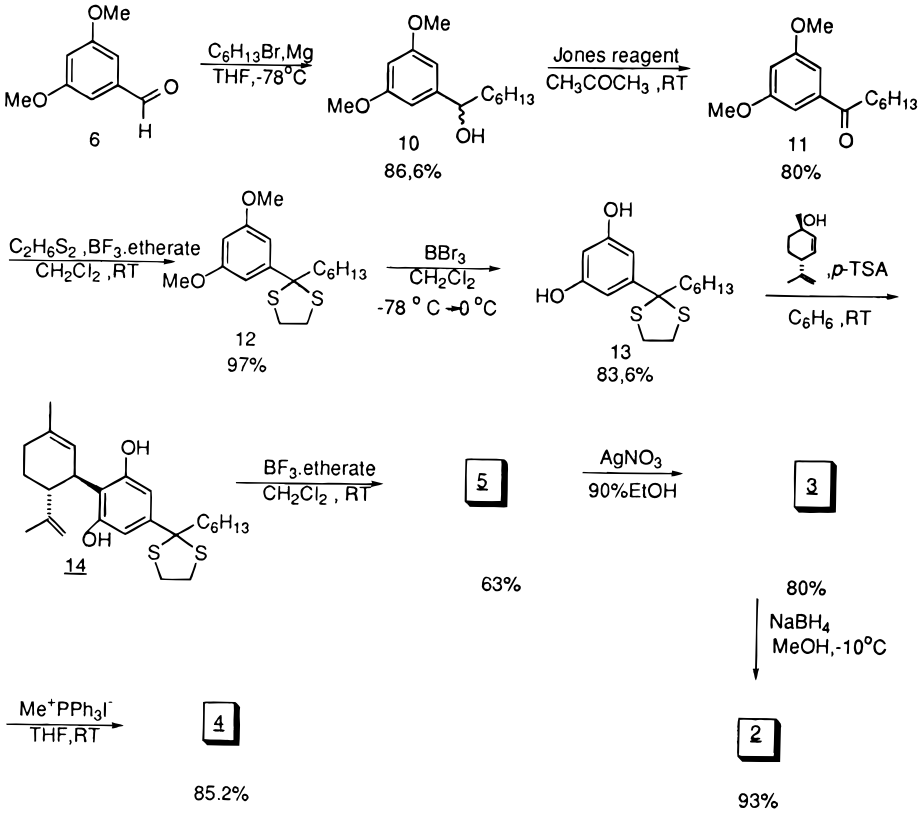
Scheme 1



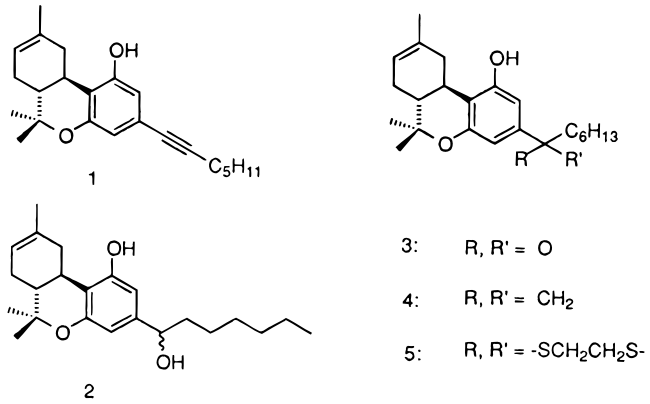
Scheme 2



hand, condensation of 2-(3,5-dihydroxyphenyl)-2-hexyl-1,3-dithiolane (**13**) with (+)-*cis*/*trans*-*p*-menta-2,8-dien-1-ol was accomplished by sequential treatment with *p*-toluenesulfonic acid and boron trifluoride etherate[12] to give 2-(6a,7,10,10a-tetrahydro-6,6,9-trimethyl-1-hydroxy-6*H*-dibenzo[*b*,*d*]pyranyl)-2-hexyl-1,3-dithiolane (**5**) (Chart 1), in a 63% overall yield.

The two resorcinols, 5-(1-heptynyl)resorcinol (**9**) (Scheme 1) and 2-(3,5-dihydroxyphenyl)-2-hexyl-1,3-dithiolane (**13**) (Scheme 2), were synthesized from a common, commercially available starting material, 3,5-dimethoxybenzaldehyde (**6**). Thus, the Wittig reaction of 3,5-dimethoxybenzaldehyde (**6**) with α,α-dibromomethylenetriphenylphosphorane[13] gave the vinylic dibromide **7** in a 99% yield. Sequential treatment of **7** with *n*-butyllithium and 1-bromopentane in the presence of hexamethylphosphoric triamide[14] (HMPA) afforded 1,3-dimethoxy-5-(1-heptynyl)benzene (**8**) in 60% yield. Subsequently, deprotection of the methoxy groups to provide 5-(1-heptynyl)resorcinol (**9**) was achieved using boron tribromide in methylene chloride at −10 °C for 16 h, resulting in a 66% yield after purification.

Chart 1



Reaction of 3,5-dimethoxybenzaldehyde with hexylmagnesium bromide occurred under the usual condi-

Table 1. Affinities (K_i) of Cannabinoid Analogues for the CB1 and CB2 Receptors (95% Confidence Limits)

	K_i (nM)[a]	
compd	CB1	CB2
1	0.65 ± 0.12	3.1 ± 0.13
2	86.4 ± 17.9	65.6 ± 18.6
3	21.7 ± 7.1	83.7 ± 19.2
4	2.17 ± 0.46	3.3 ± 0.58
5	0.32 ± 0.08	0.52 ± 0.17
14	136 ± 35	50.4 ± 14.2

[a] Affinities of the cannabinoid analogues for CB1 and CB2 were determined using rat brain (CB1) or mouse spleen (CB2) membranes and [^{3}H]CP-55,940 as previously described.[22] K_i values were obtained from three independent experiments run in duplicate and are expressed as the mean of the three values.

tions, giving 3,5-dimethoxy-1-(1′-hydroxyheptyl)resorcinol (**10**) (Scheme 2) in an 87% yield. Oxidation of **10** with Jone's reagent at room temperature resulted in the formation of ketone **11** in an 80% yield, which upon treatment with 1,2-ethanedithiol and boron trifluoride etherate in methylene chloride gave the corresponding thioketal **12** in a 97% yield. This was demethylated with boron tribromide in methylene chloride at 0 °C for 12 h to afford resorcinol **13** in 84% yield. Close analogues of **13** and **5** have been reported by Fahrenholtz.[15]

Compound **5** served as a starting point for several side-chain analogues. Thus cleavage of the 1,3-dithiolane group with silver nitrate in aqueous 90% ethanol[16] afforded the keto analogue **3**, which was subsequently converted to the corresponding alcohol **2** in 93% yield. Finally the preparation of **4** was most efficiently achieved using the Taylor protocol.[17]

Receptor Binding Studies

The abilities of **1**−**5** and **14** to displace radiolabeled CP-55,940 from purified rat forebrain synaptosomes and mouse spleen synaptosomes were determined as described in the methods section. K_i values calculated from the respective displacement curves are listed in Table 1 and serve as indicators for the affinities of these Δ^8-THC analogues for the CB1 and CB2 receptors.

Results and Discussion

The compounds included in this study are Δ^8-THC analogues in which the optimized seven-carbon side chain was modified at the C1′ position. As can be observed in Table 1, the range of K_i values of the six analogues included in this study spans over 2 orders of magnitude, indicating that structural modifications at the benzylic position of the side chain of the Δ^8-THC skeleton can have a profound effect on the affinities of these molecules for both the CB1 and CB2 receptors. Our results confirm earlier findings[9] clearly indicating that a seven-carbon side chain on the tetrahydrocannabinoid structure can lead to high-affinity analogues for both cannabinoid receptors as is exemplified by **1**, **4**, and **5**. Also in accordance with earlier structure−activity relationships (SAR),[7] the cannabidiol analogue **14** has considerably reduced afffinities.

Introduction of a triple bond in the benzylic position restricts rotation around the C1′−C2′ bond. The resulting Δ^8-THC analogue **1** has a high affinity for CB1 as well as a 5-fold selectivity for this receptor over its subtype CB2. In a previous publication,[9] using a high-affinity 11-hydroxyhexahydrocannabinol prototype, we had observed that the 1′-alkyne analogue showed a considerable (10-fold) selectivity for CB1. The corresponding Δ^8-THC analogue **1** included in this study also showed selectivity for CB1 (5-fold). On the basis of earlier work in which 11-hydroxyhexahydrocannabinol (11-OH-HHC) analogues had optimal pharmacophoric requirements for CB1,[12,18] we had anticipated that **1** would show a lower affinity for CB1 than its corresponding 11-OH-HHC analogue. To our surprise **1** had a higher affinity with a subnanomolar K_i value. This perhaps indicates that the presence of the C1′-triple bond in the 11-OH-HHC prototype does not allow the simultaneous optimal alignment of both the 11-hydroxy and side-chain pharmacophores in the CB1 active site thus reducing this analogue's affinity for the receptor.

The introduction of a 1′-methylene group (**4**) results in a high-affinity Δ^8-THC ligand with only limited selectivity for CB1. However, this high affinity is severely reduced when the sp^2 carbon atom in **4** is substituted with an oxygen in the more polar keto analogue **3**. This analogue now has a 10-fold lower affinity for CB1 and an even lower affinity for CB2. The affinity for CB1 is further reduced with a hydroxyl group substitution at C1′. This equally populated diastereomeric mixture (**2**) shows relatively low affinity for CB1 and virtually no selectivity with respect to CB2. The results indicate a requirement for hydrophobic groups at C1′ for optimal selectivity for both CB receptors and reduced tolerance for more polar substituents. The above conclusion is strongly reinforced by our results with the 1′,1′-dithiolane analogue **5**. This compound with K_i values of 0.32 and 0.52 nM for CB1 and CB2, respectively is a cannabimimetic molecular probe with one of the highest affinities reported to date. This increase in affinity can clearly be attributed to the hydrophobic subsite for both CB1 and CB2 at the level of the benzylic side-chain carbon. Currently, we are investigating the structural requirements for interaction of this subsite through the design and synthesis of a series of new analogues. We are hopeful that the identification of the above-mentioned subsite will provide us with new opportunities for the design of very high-affinity novel analogues with selectivities for the two cannabinoid receptor subtypes.

Materials and Experimental Procedures

All reactions were carried out under scrupulously dry conditions. Organic phases were dried over Na_2SO_4 and evaporated under reduced pressure. Silica gel 60, 200−400 mesh, and ASTM, 150−230 mesh (E. Merck, Germany), were used for flash and gravity column chromatography, respectively. All compounds were demonstrated to be homogeneous by analytical TLC on precoated silica gel TLC plates (grade 60, F254, E. Merck), and chromatograms were visualized by phosphomolybdic acid staining. ^{1}H NMR spectra were recorded on a Bruker AC 300 spectrometer operating at 300 MHz and are reported in units of δ relative to internal $CHCl_3$ at 7.24 ppm. All NMR spectra were recorded in $CDCl_3$ unless otherwise stated. Analyses indicated by the symbols of the elements were carried out by the microanalytical section of the Institute of Organic and Pharmaceutical Chemistry, National Hellenic Research Foundation.

Vinylic Dibromide 7. To a solution of carbon tetrabromide (2 g, 12 mmol) in 28 mL of anhydrous methylene chloride at 0 °C was added triphenylphosphine (7.86 g, 30 mmol) portionwise so that the reaction temperature did not exceed 5 °C. The

mixture was stirred at 0 °C for an additional 30 min, and a solution of 3,5-dimethoxybenzaldehyde (2 g, 12 mmol) in 7.5 mL of dry methylene chloride was added dropwise via syringe. Stirring at 0 °C was continued until TLC showed that all of the aldehyde had reacted (3 h). At completion, methylene chloride was removed on a rotary evaporator, and the residue was purified by flash column chromatography (20% diethyl ether-petroleum ether as eluent) to afford 3.46 g (yield 99%) of dibromide **7**: ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.41 (s, 1H), 6.68 (d, J = 2.1 Hz, 2H), 6.44 (brs, 1H), 3.79 (s, 6H). Anal. ($C_{10}H_{10}Br_2O_2$) C, H.

1,3-Dimethoxy-5-(1′-heptynyl)benzene (8). Dibromide **7** (2 g, 6.9 mmol) was dissolved in anhydrous THF (22 mL) and cooled to −78 °C under an argon atmosphere. A 1.6 M solution of *n*-butyllithium in hexanes (9.5 mL, 15.18 mmol) was added over 15 min, and the reaction mixture was stirred for 1 h at −78 °C. A solution of 1-bromopentane (0.95 mL, 7.59 mmol) in 15 mL of dry hexamethylphosphoramide was then added dropwise, and stirring was continued at −78 °C for 4.5 h, whereupon the reaction was quenched by the addition of a saturated solution of NH_4Cl. Workup under standard conditions followed by flash column chromatography (5% diethyl ether−petroleum ether as eluent) provided 950 mg (yield 60%) of compound **8**: ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.55 (d, J = 1.16 Hz, 2H), 6.39 (brs, 1H), 3.76 (s, 6H), 2.39 (t, J = 6.85 Hz, 2H), 1.7−1.3 (m, 6H), 0.92 (t, J = 7.2 Hz, 3H). Anal. ($C_{15}H_{20}O_2$) C, H.

5-(1′-Heptynyl)resorcinol (9). Boron tribromide (1.28 g, 5.12 mmol) was added to a solution of 1,3-dimethoxy-5-(1-heptynyl)benzene (0.54 g, 2.33 mmol) in methylene chloride (77.5 mL) at −78 °C under an argon atmosphere. Following the addition of BBr_3, the reaction temperature was gradually raised over a period of 2 h to −10 °C. Stirring was continued at that temperature until completion of the reaction was indicated by TLC (16 h). Unreacted boron tribromide was destroyed by addition of methanol. The solvent was removed in vacuo, and the residual oil was diluted with ethyl acetate. The solution was washed with saturated sodium bicarbonate (2 × 20 mL), water (2 × 10 mL), and brine (2 × 20 mL). The organic layer was dried over Na_2SO_4, filtered, and concentrated in vacuo. Purification by flash column chromatography (60% diethyl ether−petroleum ether as eluent) gave 310 mg (yield 65%) of 5-(1-heptynyl)resorcinol (9): ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.46 (d, J = 2.0 Hz, 2H), 6.29 (brs, 1H), 5.10 (brs, 2H), 2.36 (t, J = 7 Hz, 2H), 1.65−1.25 (m, 6H), 0.91 (t, J = 7 Hz, 3H). Anal. ($C_{13}H_{16}O_2$) C, H.

(−)-(1′-Heptynyl)-Δ^8-THC (1). 5-(1-Heptynyl)resorcinol (**9**) (150 mg, 0.736 mmol) was azeotroped with dry benzene (2 × 10 mL) and dissolved in dry benzene (7 mL). Dry *p*-toluenesulfonic acid was added (26 mg, 0.137 mmol) followed by the addition of (+)-*cis*/*trans*-*p*-mentha-2,8-dien-1-ol (140 mg, 0.92 mmol). The reaction mixture was heated under dry argon to 80 °C for 4 h. Upon completion, the mixture was diluted with diethyl ether (40 mL) and the ethereal solution washed successively with saturated aqueous $NaHCO_3$ (2 × 5 mL), H_2O (2 × 5 mL), and brine (2 × 10 mL) and dried over Na_2SO_4. Filtration, concentration in vacuo, and purification by flash column chromatography (10% diethyl ether−petroleum ether as eluent) afforded 62 mg of **1** (25%): ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.78 (s, 1H, OH), 6.28 (d, J = l.96 Hz, 1H), 6.09 (d, J = 1.96 Hz, 1H), 5.43 (bs, 1H), 3.2 (dd, J = 4, 12 Hz, 1H), 2.8−2.53 (m, 3H), 2.15 (m, 1H), 1.95−1.52 (m, 6H), 1.38 (s, 3H), 1.37−1.2 (bs, 6H), 1.09 (s, 3H), 0.90 (t, J = 7 Hz, 3H). Anal. ($C_{23}H_{30}O_2$) C, H.

1-(3,5-Dimethoxyphenyl)heptan-1-ol (10). 3,5-Dimethoxybenzaldehyde (**6**) (2 g, 12 mmol) was dissolved in dry tetrahydrofuran (24 mL) under an argon atmosphere and the flask cooled to −78 °C. Hexylmagnesium bromide [prepared from 1-bromohexane (5.94 g, 36 mmol) and Mg turnings (876 mg, 36 mmol) in dry tetrahydrofuran (72 mL)] was added, and stirring continued for 4 h at −60 °C. Upon completion the reaction was quenched by the addition of saturated aqueous ammonium chloride (10 mL). The reaction mixture was extracted with ethyl acetate (3 × 50 mL), washed with saturated NH_4Cl (2 × 10 mL) and brine (20 mL), dried (Na_2SO_4), and evaporated. Purification by flash column chromatography (20% diethyl ether−petroleum ether as eluent) provided 2.6 g of compound **10** (86%): ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.49 (d, J = 2.1 Hz, 2H), 6.36 (t, J = 2.1 Hz, 1H), 4.57 (t, J = 5.7 Hz, 1H), 3.78 (s, 6H), 1.96 (brs, 1H, O*H*), 1.70 (m, 2H), 1.27 (bs, 8H), 0.87 (t, J = 6.8 Hz, 3H). Anal. ($C_{15}H_{24}O_3$) C, H.

1-(3,5-Dimethoxyphenyl)-1-heptan-1-one (11). Resorcinol **10** (2.47 g, 9.8 mmol) was dissolved in acetone (25 mL). To this cold (0 °C) solution was added a solution of Jone's reagent (14 mL; prepared from 7 g of CrO_3, 50 mL of H_2O, and 6.1 mL of concentrated H_2SO_4), and the reaction mixture was stirred at room temperature for 0.5 h. Upon completion the reaction was quenched by the addition of propan-2-ol, the mixture diluted with ethyl acetate (80 mL), and the organic phase washed with 20% aqueous sodium bisulfite (2 × 20 mL), H_2O (20 mL), and brine (25 mL), dried (Na_2SO_4), and evaporated. Purification by flash column chromatography (30% diethyl ether−petroleum ether) yielded compound **11** (1.95 g, 79.6%): ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.09 (d, J = 2.32 Hz, 2H), 6.64 (t, J = 2.32 Hz, 1H), 3.84 (s, 6H), 2.91 (t, J = 7.3 Hz, 2H), 1.71 (m, 2H), 1.33 (brs, 6H), 0.89 (t, J = 6.7 Hz, 3H). Anal. ($C_{15}H_{22}O_3$) C, H.

2-(3,5-Dimethoxyphenyl)-2-hexyl-1,3-dithiolane (12). Ketone **11** (1.9 g, 7.6 mmol) was dissolved in methylene chloride (30 mL), and 1,2-ethanedithiol (1.3 g, 13.8 mmol) and boron trifluoride etherate (0.272 mL, 2.3 mmol) were added. The solution was stirred at room temperature overnight, and at completion a saturated solution of $NaHCO_3$ (10 mL) was added. The mixture was diluted with diethyl ether; the organic layer was washed with water (15 mL) and brine (2 × 15 mL), dried (Na_2SO_4), and evaporated to afford 2.4 g of **12** (97%) sufficiently pure for the following step: ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.86 (d, J = 2.0 Hz, 2H), 6.33 (t, J = 2.0 Hz, 1H), 3.79 (s, 6H), 3.38−3.19 (m, 4H), 2.33 (t, J = 7.4 Hz, 2H), 1.21 (brs, 8H), 0.83 (t, J = 6.9 Hz, 3H). Anal. ($C_{17}H_{26}O_2S_2$) C, H.

2-(3,5-Dihydroxyphenyl)-2-hexyl-1,3-dithiolane (13). Boron tribromide (0.47 mL, 4.93 mmol) was added to a solution of **12** (0.6 g, 1.84 mmol) in methylene chloride (49 mL) at −78 °C under an argon atmosphere. Following the addition of boron tribromide, the reaction temperature was gradually raised over a period of 3 h to 0 °C. Stirring was continued at that temperature until completion of the reaction (12 h). Workup of the reaction mixture was performed as described earlier for compound **9**. Purification by flash column chromatography (50% diethyl ether−petroleum ether as eluent) gave 460 mg (84%) of compound **13**: ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.76 (d, J = 2.2 Hz, 2H), 6.21 (t, J = 2.2 Hz, 1H), 4.95 (s, 2H), 3.36−3.16 (m, 4H), 2.25 (m, 2H), 1.19 (brs, 8H), 0.82 (t, J = 7.0 Hz, 3H). Anal. ($C_{15}H_{22}O_2S_2$) C, H.

(−)-2-(6a,7,10,10a-Tetrahydro-6,6,9-trimethyl-1-hydroxy-6*H*-dibenzo[*b*,*d*]pyranyl)-2-hexyl-1,3-dithiolane (5). To a solution of resorcinol **13** (0.46 g, 1.54 mmol) in dry benzene (14.0 mL) at 25 °C under an argon atmosphere was added (+)-*cis*/*trans*-*p*-mentha-2,8-dien-1-ol (293 mg, 1.93 mmol) followed by the addition of *p*-toluenesulfonic acid (54.5 mg, 0.29 mmol). The reaction mixture was stirred at 25 °C for 0.5 h, at which time TLC indicated the complete consumption of starting material. The reaction mixture was diluted with ether. The ethereal solution was washed with saturated $NaHCO_3$, H_2O, and brine and dried (Na_2SO_4). Evaporation of the solvent followed by flash column chromatography (10% diethyl ether−petroleum ether) afforded 670 mg of cannabidiol **14**: ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.72 (bs, 2H), 6.0 (s, 1H, O*H*), 5.55 (s, 1H), 4.9 (bs, 1H, O*H*), 4.61 (s, 1H), 4.55 (s, 1H), 3.85 (m, 1H), 3.4−3.2 (m, 5H), 2.5−2.0 (m, 6H), 1.7 (s, 3H), 1.40 (s, 3H), 1.25 (bs, 8H), 0.85 (t, J = 7 Hz, 3H). Anal. ($C_{25}H_{36}O_2S_2$) C, H.

To a solution of **14** (0.67 g, 1.55 mmol) in anhydrous dichloromethane (44 mL) was added boron trifluoride etherate (0.69 mL, 5.55 mmol) at 0 °C. Following the addition the reaction mixture was stirred at 25 °C for 4 h, at which time TLC indicated the disappearance of starting material. The

reaction was quenched by the addition of a saturated solution of $NaHCO_3$, the mixture was concentrated in vacuo and diluted with ethyl acetate, and the organic layer was washed with water (15 mL) and brine (2 × 15 mL) and dried over Na_2SO_4. Solvent evaporation and purification by flash column chromatography (10% diethyl ether−petroleum ether as eluent) afforded **5**. The overall yield from **13** was 422 mg (63%): ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.8 (d, J = 1.98 Hz, 1H, H_4), 6.6 (d, J = 1.98 Hz, 1H, H_2), 5.45 (bs, 1H, H_8), 4 75 (s, 1H, OH), 3.4−3.2 (m, 4H, -$S(CH_2)_2$S-), 3.20 (m, 1H, $H_{10\alpha}$), 2.7 (br s, 1H, H_{10a}), 2.3 (m, 2H, 2′CH_2), 2.15 (m, 1H, H_7), 2.05−1.83 (m, 3H, $H_{6\alpha}$, H_7, $H_{10\beta}$), 1.7 (s, 3H), 1.4 (s, 3H), 1.25 (brs, 8H, -CH_2-), 1.1 (s, 3H), 0.85 (t, J = 7 Hz, 3H). Anal. ($C_{25}H_{36}O_2S_2$) C, H.

(−)-1-Hydroxy-6a,7,10,10a-tetrahydro-6,6,9-trimethyl-6*H*-dibenzo[*b,d*]pyran-3-yl 1′-Hexyl Ketone (3). To a stirred solution of 1,3-dithiolane **5** (100 mg, 0.23 mmol) in 90% ethanol (4 mL) at 25 °C was added a solution of $AgNO_3$ (120 mg, 0.69 mmol) in water (0.5 mL), and the reaction mixture stirred at room temperature for 3 h. At completion the precipitate was removed and washed with ethyl acetate and the filtrate further diluted with ethyl acetate, washed with brine, and dried (Na_2SO_4). Solvent evaporation and purification of the residue by flash column chromatography (15% diethyl ether−petroleum ether as eluent) afforded 65 mg of **3** (79%): ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.18 (d, J = 1.4 Hz, 1H), 7.0 (d, J = 1.4 Hz, 1H), 6.4 (s, 1H), 5.44 (d, J = 4 Hz, 1H), 3.32 (dd, J = 4, 13 Hz, 1H), 2.9 (t, J = 7 Hz, 2H), 2.79 (m, 1H), 2.15 (m, 1H), 1.94−1.7 (m, 3H), 1.67 (s, 3H), 1.39 (s, 3H), 1.36 (bs, 8H), 1.08 (s, 3H), 0.88 (t, J = 7 Hz, 3H). Anal. ($C_{23}H_{32}O_3$) C, H.

(−)-(1-Hydroxy-6a,7,10,10a-tetrahydro-6,6,9-trimethyl-6*H*-dibenzo[*b,d*]pyran-3-yl)-α-(1′-hexyl)methanol (2). 3 (100 mg, 0.282 mmol) was dissolved in methanol (1.4 mL), and the mixture cooled to −10 °C. Sodium borohydride (8.6 mg, 0.23 mmol) was added in one portion, and stirring continued at −10 °C for 2 h, whereupon the reaction was quenched by the addition of a saturated solution of ammonium chloride. The mixture was concentrated in vacuo to remove most of the methanol. The residue was extracted with ethyl acetate (3 × 5 mL) and the organic phase washed with water (5 mL) and brine (5 mL) and then dried (Na_2SO_4). Filtration, evaporation, and purification by flash column chromatography (30% diethyl ether−petroleum ether as eluent) yielded 93.5 mg of **2** (93%): ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.4−6.33 (m, 2H), 5.42 (s, 1H), 4.5 (m, 1H), 3.26 (dd, 1H), 2.77 (m, 1H), 2.13 (m, 1H), 1.9−1.8 (m, 5H), 1.7 (s, 3H), 1.38 (s, 3H), 1.26 (bs, 8H), 1.09 (s, 3H), 0.86 (t, J = 7 Hz, 3H). Anal. ($C_{23}H_{34}O_3$) C, H.

(−)-1-(Δ^8-THC)-1-heptene (4). To a suspension of methyl triphenylphosphonium iodide (626 mg, 1.55 mmol) in anhydrous tetrahydrofuran (8.5 mL) was added potassium bis-(trimethylsilyl)amide (303 mg, 1.52 mmol). The mixture was stirred at room temperature for 0.5 h to ensure complete formation of the yellow ylid. A solution of **3** (110 mg, 0.31 mmol) in dry tetrahydrofuran (0.62 mL) was then added dropwise, and the reaction mixture was stirred for 2 h at room temperature. Upon completion, a saturated aqueous solution of ammonium chloride was added and the aqueous layer extracted with diethyl ether. The combined organic layers were washed with water and brine, dried (Na_2SO_4), and evaporated to dryness. Flash chromatography (15% diethyl ether−petroleum ether as eluent) yielded 93 mg of **4** (85%): ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.5 (d, J = 1.5 Hz, 1H), 6.34 (d, J = 1.5 Hz, 1H), 5.43 (d, J = 3.8 Hz, 1H), 5.21 (s, 1H), 4.96 (s, 1H), 4.87 (bs, 1H, O*H*), 3.23 (dd, J = 4.14 Hz, 1H), 2.73 (m, 1H), 2.41 (t, J = 7 Hz, 2H), 2.15 (m, 1H), 1.93−1.81 (m, 3H), 1.7 (s, 3H), 1.41 (s, 3H), 1.34 (bs, 8H), 1.11 (s, 3H), 0.86 (t, J = 7 Hz, 3H). Anal. ($C_{24}H_{34}O_2$) C, H.

Radioligand Binding Assay. Forebrain synaptosomal membranes were prepared from frozen rat brains by the method of Dodd et. al.[19] and were used to assess the affinities of the novel analogues for the CB1 binding sites, while affinities for the CB2 sites were measured using a membrane preparation from frozen mouse spleen using a similar procedure.[20] The displacement of specifically tritiated CP-55,940 from these membranes was used to determine the K_{50} values for the test compounds. The assay was conducted in a 96-well microfilter plate. The samples were filtered using a Packard Filtermate 96 and Whatman GF/C filter plates, and 0.5% BSA was incorporated into the wash buffer. Radioactivity was detected using MicroScint 20 scintillation cocktail added to the dried filter plates and was counted using a Packard Instruments Top Count. Data were collected from three independent experiments between 100% and 0% specific binding for [^{3}H]CP-55,940, determined using 0 and 100 nM CP-55,940. The normalized data from three independent experiments were combined and analyzed using a four-parameter logistic equation to yield IC_{50} values which were converted to K_i values using the assumptions of Cheng and Prussoff.[21]

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