procedures already described: mp 167–170 °C. Anal. ($C_{12}H_{16}N_4O_3S$·HI) C, H, N.

**6-(2-Imidazolin-2-ylamino)-2*H*-1,4-benzoxazin-3(4*H*)-one Hydriodide (5).** 7-Nitro-2*H*-1,4-benzoxazin-3(4*H*)-one[4] was converted in 43% yield to 7-amino-2*H*-1,4-benzoxazin-3(4H)-one: mp 216–219 °C. The method described for **12** was used to convert this amine to **5**: yield 15%; mp 301–303 °C. Anal. ($C_{11}H_{12}N_4O_2$·HI) C, H, N. Compound **10** was prepared, by using the same methods, from 6-nitro-benzoxazinone.[4]

**1-Ethyl- and 1-Acetyl-6-(2-imidazolin-2-ylamino)indoline Hydriodide (15 and 16).** 6-Nitroindoline (Aldrich) was converted to the 1-acetyl derivative which was converted by methods already described to give **15** and **16**.

**Pharmacology.** Details of the in vitro screening procedures have been described in an earlier publication.[7]

**Activity at Peripheral $\alpha_2$-Adrenoreceptors in Vivo.** Prejunctional agonist and antagonist activities were studied in the vas deferens of pithed rats. Agonist potency was determined as the dose (μg/kg, iv) required to inhibit the electrical-evoked contractions of the vas deferens by 40% ($ED_{40}$ value). The maximal percent inhibition of the vas deferens was also noted, indicating the intrinsic activity of the test compound. Antagonist potencies were determined as the dose (as an infusion μg/kg per min) required to produce a 2-fold shift of the cumulative dose–response curve to UK-14,304[9] on the twitch response of the vas deferens. Agonist and antagonist studies were performed in separate groups of pithed rats ($n = 6$). Agonists were administered in a cumulative manner and the antagonists were continuously infused (0.03 mL/min, iv) to obtain equilibrium conditions, and a dose–response curve to UK-14,304 was constructed 10 min after the start of the infusion.

**Activity at Central $\alpha_2$-Adrenoreceptors.** Agonist activity was determined by measuring the mydriatic effects of the compounds after iv or icv administration to pentobarbitone anesthetized rats. Antagonist potency was determined by the reversal of a maximally effective mydriatic dose of guanoxabenz (300 μg/kg).[10] Compounds were administered 10 min after guanoxabenz, and the cumulative dose reversing guanoxabenz by 50% was calculated ($AD_{50}$, μg/kg).

**Acknowledgment.** We thank Diane Strachan and Brian Gadie for their assistance in the biological testing, and Pamela Parker for typing the manuscript.

**Registry No.** **1**, 87135-03-3; **1**·maleate, 87135-04-4; **2**, 4205-90-7; **4**, 70183-99-2; **4**·HCl, 103124-99-8; **5**, 120711-83-3; **5**·HI, 120711-66-2; **6**, 120711-84-4; **6**·HI, 120711-67-3; **7**, 120711-85-5; **7**·HI, 120711-68-4; **8**, 120711-86-6; **8**·HI, 120711-69-5; **9**, 120711-87-7; **9**·2HCl, 120711-70-8; **10**, 120711-88-8; **10**·HI, 120711-71-9; **11**, 120711-89-9; **11**·HI, 120711-72-0; **12**, 120711-90-2; **12**·HI, 120711-73-1; **13**, 120711-91-3; **13**·HI, 120711-74-2; **14**, 120711-92-4; **14**·HI, 120711-75-3; **15**, 120771-32-6; **15**·HI, 120711-76-4; **16**, 120771-33-7; **16**·HI, 120711-77-5; 6-nitro-1,4-benzoxazine, 120711-78-6; 3,4-dihydro-6-nitro-1,4-benzoxazine-4-carbaldehyde, 120711-79-7; 3,4-dihydro-6-amino-1,4-benzoxazine-4-carbaldehyde, 120711-80-0; 2-(methylthio)-2-imidazoline hydriodide, 5464-11-9; 3,4-dihydro-7-nitro-1,4-benzoxazine, 120711-81-1; 3,4-dihydro-4-methyl-6-nitro-1,4-benzoxazine, 120711-82-2; 3,4-dihydro-4-methyl-7-nitro-1,4-benzoxazine, 120711-93-5; 4-acetyl-3,4-dihydro-7-nitro-1,4-benzoxazine, 120711-94-6; 4-ethyl-3,4-dihydro-7-nitro-1,4-benzoxazine, 120711-95-7; 7-amino-4-ethyl-3,4-dihydro-1,4-benzoxazine, 105297-44-7; 4-acetyl-3,4-dihydro-6-nitro-1,4-benzoxazine, 120711-96-8; 4-acetyl-3,4-dihydro-6-amino-1,4-benzoxazine, 120711-97-9; 4-benzoyl-3,4-dihydro-7-nitro-1,4-benzoxazine, 120711-98-0; 4-benzyl-3,4-dihydro-7-nitro-1,4-benzoxazine, 120711-99-1; 7-amino-4-benzyl-3,4-dihydro-1,4-benzoxazine, 120712-00-7; 1-acetyl-2-imidazolidinone, 5391-39-9; 1-acetyl-2-[(4-benzyl-3,4-dihydro-1,4-benzoxazin-7-yl)imino]imidazolidine, 120712-01-8; 3,4-dihydro-4-mesyl-6-nitro-1,4-benzoxazine, 120712-02-9; 7-nitro-2*H*-1,4-benzoxazin-3(4*H*)-one, 81721-86-0; 7-amino-2*H*-1,4-benzoxazin-3(4*H*)-one, 26215-14-5; 6-nitrobenzoxazinone, 81721-87-1; 6-nitroindoline, 19727-83-4; 1-acetyl-6-nitroindoline, 22949-08-2.

# The Importance of the Orientation of the C9 Substituent to Cannabinoid Activity

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We have found a correlation between cannabinoid psychopharmacological activity and the orientation of the C9 substituent in one class of cannabinoid derivatives. We report here a study of the active cannabinoids $\Delta^9$-tetrahydrocannabinol ($\Delta^9$-THC), $\Delta^8$-tetrahydrocannabinol ($\Delta^8$-THC), and 11β-hexahydrocannabinol (11β-HHC); the minimally active cannabinoid 11α-hexahydrocannabinol (11α-HHC); and the inactive cannabinoids $\Delta^7$-tetrahydrocannabinol ($\Delta^7$-THC) and $\Delta^{9,11}$-tetrahydrocannabinol ($\Delta^{9,11}$-THC). Our working hypothesis is that there are two components of cannabinoid structure which confer upon these compounds reactivity characteristics crucial to activity: the directionality of the lone pairs of electrons of the phenyl group hydroxyl oxygen and the orientation of the carbocyclic ring relative to this oxygen. The structures of these six molecules were optimized by using the method of molecular mechanics as encoded in the MMP2(85) program. Other possible minimum-energy conformations of the carbocyclic ring were calculated by driving one torsion angle in this ring by use of the dihedral driver option in MMP2(85). The rotational energy behavior of the phenyl group hydroxyl in each molecule was studied also by using the dihedral driver option in MMP2(85). We found that the carbocyclic ring in 11α-HHC can exist in either a chair or a twist conformation. The carbocyclic ring in $\Delta^9$-THC, in $\Delta^8$-THC, and in $\Delta^7$-THC was found to exist only in a half-chair conformation, while the carbocyclic ring in 11β-HHC and in $\Delta^{9,11}$-THC was found to exist only in a chair form. The results of the rotational energy profiles indicated that the minimum-energy positions of the phenyl group hydroxyls are nearly identical in all molecules. These molecules, then, were found to differ only in the conformation of the carbocyclic ring in each. This conformation, in turn, determines the orientation of this ring and its C9 substituent relative to the oxygen of the phenyl group hydroxyl. In order to assess the orientation of the carbocyclic ring with respect to the phenyl group hydroxyl oxygen in each optimized structure, the following nonbonded torsion angles were measured: C10–C10a–C1–O, C8–C7–C1–O, C11–C9–C1–O, and C9–Q–C1–O (where Q is a dummy atom placed midway between C8 and C10). A correlation was found between activity and the C11–C9–C1–O angle, an angle that measures the orientation of the C11 methyl group (i.e., the C9 substituent) relative to the oxygen of the phenyl group hydroxyl. C11–C9–C1–O was found to be negative for all active cannabinoids. As C11–C9–C1–O becomes positive, activity is significantly reduced or abolished. These results seem to indicate that there is a critical area near the top of the carbocyclic ring which must not be blocked. Such findings argue strongly for a steric requirement at the site of action of these compounds.

Cannabinoids are the group of $C_{21}$ compounds that are typical of and present in *Cannabis sativa*, their carboxylic acids, analogues, and transformation products.[1] Since Mechoulam et al. reported (−)-*trans*-$\Delta^9$-tetrahydro-

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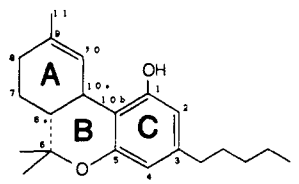
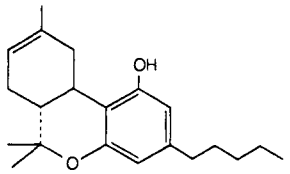
Chart I




1

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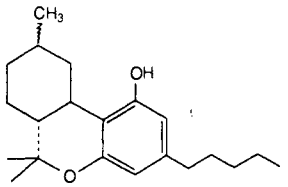
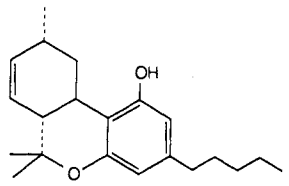
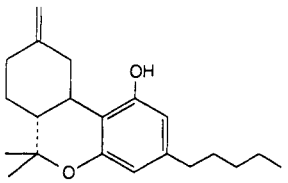



3a, C-11 methyl group β, equatorial

b, C-11 methyl group α, axial




4




5

cannabinol ($\Delta^9$-THC; 1; see Chart I) to be the major psychopharmacologically active constituent of cannabis,[2,3] many cannabinoid analogues have been synthesized and tested. Structure–activity relationships (SARs) in the cannabinoids have been generated for effects on prostaglandin synthesis[4] and for analgesic activity,[5] but principally for psychopharmacological activity.[6] Little is yet known, however, about the molecular basis of action of these compounds. Cannabinoids are thought by some to interact with a specific receptor in the central nervous system.[7] Others hold that cannabinoids exert their effect by interactions with membranes in one of several ways: by fluidizing them in a manner similar to the Meyer–Overton explanation of anesthesia,[8] by altering their composition,[9] or by stereoselectively interacting with the phospholipid bilayer.[10]

In our work, we seek to reveal elements of molecular reactivity that are related to activity. The information provided by SAR studies of cannabinoid psychopharmacological activity is an excellent starting point for our work. We use the methods of theoretical chemistry to calculate the molecular properties generated by the entire structure of each cannabinoid. These molecular properties are presumed to be directly responsible for the molecular interactions at the site of action. Our ultimate aim is to shed more light on the basis of action of these compounds at the molecular level.

Our previous studies of the molecular determinants for cannabinoid activity have led us to hypothesize that there are two aspects of the structure of $\Delta^9$-THC (1) that confer reactivity characteristics upon the molecule which are crucial to activity. These components are the orientation of the lone pairs of electrons of the phenyl group hydroxyl oxygen and the orientation of the carbocyclic ring (ring A) relative to this oxygen.[11] Because we believe that two aspects are important, our work on the molecular determinants for cannabinoid psychopharmacological activity has taken two directions. In the first, we have studied the position of the phenyl group hydroxyl in both active and inactive cannabinoids, all of which possess very similar carbocyclic ring orientations.[12] Our original study of the template molecule, $\Delta^9$-THC (1), revealed that there are two minimum-energy conformations for the phenyl group hydroxyl in 1. In conformation I (C2–C1–O–H = −1°), the phenolic hydrogen points away from the carbocyclic ring; consequently, the lone pairs of electrons of the oxygen point toward this ring. In conformation II (C2–C1–O–H = 155°), the phenolic hydrogen points toward the carbocyclic ring; consequently, the lone pairs of electrons of the oxygen point away from this ring.[11] Comparisons with other cannabinoids led us to hypothesize that conformation II is the more relevant conformation at the site of action.[12,13] We are currently synthesizing compounds that will test this part of our working hypothesis.[13]

This paper deals with the second part of our working hypothesis. Here we investigate the effect that a change in orientation of the carbocyclic ring (ring A) relative to the phenyl group hydroxyl oxygen has upon activity. To this end, we have carefully chosen six cannabinoids to study. The following criterica were used in the selection of subject molecules: (1) All cannabinoids must have been assayed in the same pharmacological test. (2) All cannabinoids must differ from $\Delta^9$-THC (1) only in their carbocyclic ring. Thus, subject molecules must have pentyl side chains, have no additional substitution on rings B and C, possess ring B conformations identical with that of ring B in 1, and possess phenyl group hydroxyl conformations identical with those of 1. (3) The variations in ring A must be such that the electrostatic potential generated in this region be similar to that of 1.[11] Molecules that possess functional group substitutions in ring A such as hydroxyl, carboxyl, ester, aldehyde, or amino groups were not considered for this study, because the addition of these functionalities would introduce two variables, conformational changes and electrostatic potential changes.

Using the above criteria, we chose five cannabinoids in addition to (−)-$\Delta^9$-THC (1). These five cannabinoids are the active cannabinoids $\Delta^8$-THC (**2**),[14] 11$\beta$-HHC (C11 methyl group equatorial; **3a**),[15] the minimally active com-

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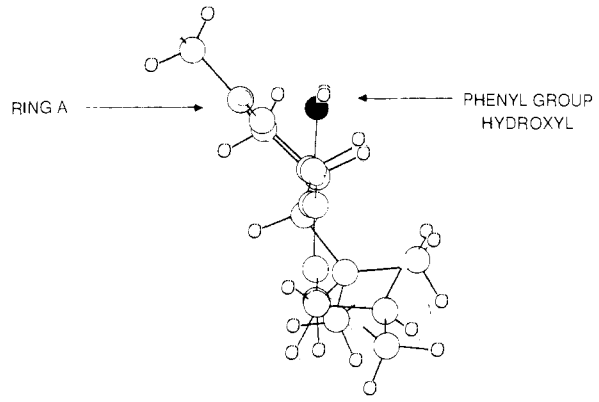


**Figure 1.** Conformation of (−)-$\Delta^9$-THC (1) (with propyl side chain) as determined by MMP2(85): the perspective of the carbocyclic ring (ring A) is viewed in the direction parallel to the vector from C2 to C10b. Here the oxygen of the phenyl group hydroxyl is shown as a blackened circle.

pound 11$\alpha$-HHC (C11 methyl group axial; **3b**),[15] and the inactive compounds $\Delta^7$-THC (**4**)[16] and $\Delta^{9,11}$-THC (**5**).[17] The activity of each of these cannabinoids has been assayed in rhesus monkey behavioral tests. In these cannabinoids, a slight structural change (i.e., a change in the position of the double bond in ring A or loss of the double bond in ring A) produces a dramatic change in activity.

Several other investigators have alluded to the importance of the carbocyclic ring in cannabinoid compounds. Using Dreiding stereo models and NMR data, Binder, Edery, and Porath[17] measured the perpendicular distance of the C11 carbon above the plane of the aromatic ring in $\Delta^9$-THC, $\Delta^8$-THC, and $\Delta^{9,11}$-THC. They suggested that distances of 200 and 220 pm seemed to be critical for psychoactivity. In discussions of stereospecificity and stereoselectivity of cannabinoid actions, Mechoulam et al.[18] have also implicated the position of the C11 atom (i.e., the methyl group at C9). They have hypothesized that, in the absence of other molecular changes, the planarity of the methyl group at the C9 position determines cannabimetic activity.

In our early studies of 1, we found that the carbocyclic ring (ring A) assumes a half-chair conformation.[11] This result was in agreement with the theoretical and NMR results of Archer et al.[19] This half-chair conformation causes the top portion of the carbocyclic ring to move out of the plane of the aromatic ring (moving into what we call the "top face" of the molecule). Looking at the drawing of 1, this conformation causes the top portion of ring A to lift up out of the plane of the paper and toward the viewer. In Figure 1 we accentuate this conformation by turning the molecule sideways so that the side chain points toward the viewer (i.e., viewing it in the direction parallel to the vector from C2 to C10b). When viewed from this perspective, the top of the carbocyclic ring (ring A) can be seen to move to the left of the phenyl group hydroxyl oxygen and into the top face of the molecule. *We have taken this orientation as characteristic of active cannabinoids*, with the idea that cannabinoids whose carbocyclic rings are not in this orientation may lose activity. In order to assess the extent to which the carbocyclic ring and its substituent protrudes into the top face of the molecule in $\Delta^9$-THC and other cannabinoids, we have measured several nonbonded torsion angles. These angles help to quantify the orientation of the carbocyclic ring with respect to the phenyl group hydroxyl oxygen (see Methods). We have called these angles "protrusion" torsion angles.

In this paper, we report on the optimized structures of six cannabinoids (including the reoptimized structure of 1), on other minimum-energy accessible conformations of the carbocyclic ring in these molecules, on the minimum-energy positions of the phenyl group hydroxyl in each molecule, on the "protrusion" torsion angles of each, and, finally, on the correlations we were able to make between these "protrusion" torsion angles and cannabinoid psychopharmacological activity.

## Methods

The crystal structure of $\Delta^9$-THC acid B[20] was used as the starting geometry for all cannabinoids. The Modify facility within the CHEM-X molecular modeling system[21] was used to delete unnecessary atoms and to add necessary ones at standard bond lengths and bond angles.[22] The side chain on each molecule was shortened from pentyl to propyl in order to keep the molecule small enough for input into other computer programs in a future stage of our studies. Such a modification is acceptable since the focus of our study is on the fused ring structure of these molecules and not on their side chains.

The structures of all molecules were optimized by using the method of molecular mechanics as encoded in the MMP2(85) program.[23] Lone pairs on the oxygen of the phenyl group hydroxyl and on the ether oxygen were included in each optimization. Since our original characterization of 1 was performed with the MM2 program,[11] we also reoptimized the structure of 1 by using MMP2(85). In order to ascertain if any other minimum-energy conformations of the carbocyclic ring (ring A) exist, we performed dihedral driver studies of this ring.[24] In each case, one torsion angle [C10a–C6a–C7–C8 in $\Delta^9$-THC (**1**), C10–C10a–C6a–C7 in $\Delta^8$-THC (**2**), C6a–C7–C8–C9 in 11$\beta$-HHC (**3a**) and 11$\alpha$-HHC (**3b**), C6a–C10a–C10–C9 in $\Delta^7$-THC (**4**), and C10a–C10–C9–C8 in $\Delta^{9,11}$-THC (**5**)] was driven in increments of 5–10°.

In our calculations, we have assumed that all minimum-energy conformations within 3 kcal/mol of the absolute minimum are accessible. This amount of energy is probably easily obtained by the interaction of the cannabinoid with bulk solvent or by interaction at its site of

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action.[25] In order to be certain that each accessible conformation identified above was capable of mimicking the low-energy phenyl group hydroxyl positions of 1, we performed an MMP2(85) dihedral driver study of molecular energy as a function of rotation about the C1–O axis for each conformer.[24] Rotations in 36 steps were made about this axis.

There are several ways in which the carbocyclic ring of some cannabinoids can protrude into the top face of the molecule. One or all of the following atoms can be involved in this protrusion: C8, C9, C10, or C11 (see the numbering system illustrated for 1). In order to describe the orientation of the top portion of the carbocyclic ring relative to the phenyl group hydroxyl oxygen, we measured four "protrusion" torsion angles by using the Calculate Geometry facility within the CHEM-X molecular modeling system.[21] These angles correspond to the following nonbonded torsion angles: C10–C10a–C1–O (angle 1), C8–C7–C1–O (angle 2), and C11–C9–C1–O (angle 3). In order to measure the inclination of C9 with respect to the phenyl group hydroxyl oxygen, it was necessary to define a point Q such that Q is the midpoint of the nonbonded distance between C8 and C10. We then measured the torsion angle, C9–Q–C1–O (angle 4). In each case, this "protrusion" torsion angle was the extent of clockwise rotation necessary to impose C1–O on C10a–C10 (or C7–C8, or C9–C11, or Q–C9) when viewed along the nonbonded distance C1→C10a (C1→C7, C1→C9), C1→Q).

## Results and Discussion

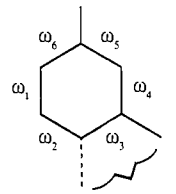
**Geometry Optimization.** The cyclohexene ring (ring A) in $\Delta^9$-THC (1), in $\Delta^8$-THC (2), and in $\Delta^7$-THC (4) was found to exist in a half-chair conformation as has been calculated for cyclohexene itself.[24] This half-chair conformation of the cyclohexene ring in 1, 2, and 4 is in keeping with our previous results[11] for 1 and those of others.[19] $\Delta^8$-THC (2) was found to be 3.36 kcal/mol lower in steric energy than $\Delta^9$-THC (1) and 4.36 kcal/mol lower in steric energy than $\Delta^7$-THC (4).

The cyclohexane ring (ring A) in 11$\beta$-HHC (**3a**) and in 11$\alpha$-HHC (**3b**) was found to exist in a chair conformation. The PMR results of Archer et al.[19] for one HHC isomer studied (**3b**) are in agreement with this result. 11$\beta$-HHC (**3a**), in which the C11 methyl group is equatorial, was found to be 1.68 kcal/mol lower in steric energy than 11$\alpha$-HHC (**3b**), in which the C11 methyl group is axial. This amount compares well with the 1.70 kcal/mol stabilization determined experimentally for equatorial versus axial methylcyclohexane in solution.[24]

The cyclohexane ring (ring A) in $\Delta^{9,11}$-THC (5) was also found to be in a chair conformation. This is the same conformation as predicted for methylenecyclohexane.[24] Binder, Edery, and Porath's NMR study of $\Delta^{9,11}$-THC (5)[17] also revealed that ring A exists in a chair conformation. Table I lists the torsion angles for ring A in each of these minimum-energy conformations.

Dihedral driver studies of ring A revealed no other minimum-energy conformations (other than the half-chair) for $\Delta^9$-THC (1), $\Delta^8$-THC (2), or $\Delta^7$-THC (4). This result is in keeping with the calculated minimum-energy conformations of cyclohexene.[24] Dihedral driver studies of ring A in the HHC's (**3a** and **3b**) revealed that a second minimum-energy conformation of ring A exists. This minimum corresponded to a twist conformation. For 11$\beta$-HHC (**3a**) this twist conformation was 5.55 kcal/mol above the chair conformation. This energy difference is quite similar to the 5.36 kcal/mol difference calculated for the chair versus twist forms of cyclohexane.[26] On the other hand, the second minimum for 11$\alpha$-HHC (**3b**), which also corresponded to a twist conformation, was only 1.72 kcal/mol above that of the chair conformation. The stabilization of the twist form of 11$\alpha$-HHC (**3b**) relative to the twist form of 11$\beta$-HHC (**3a**) is due primarily to lower torsional strain and to lower van der Waals 1,4 interactions. Thus, according to our 3 kcal/mol criteria, the twist conformation of 11$\alpha$-HHC (**3b**) is an accessible conformation. The ring torsional values of the twist form of **3b** are listed in Table I. Dihedral driver studies of ring A in $\Delta^{9,11}$-THC revealed a second minimum-energy conformation, a twist conformation 3.02 kcal/mol above the chair conformation. Since accessible conformations were taken to be those within 3 kcal/mol of the absolute minimum, we concluded that the only accessible conformation of ring A in $\Delta^{9,11}$-THC is the chair. Solution NMR studies performed by Binder et al.[17] are consistent with this conclusion.

**Table I.** Carbocyclic Ring Torsion Angles




| molecule | $\omega_1$, deg | $\omega_2$, deg | $\omega_3$, deg | $\omega_4$, deg | $\omega_5$, deg | $\omega_6$, deg |
|---|---|---|---|---|---|---|
| $\Delta^9$-THC | 37 | −66 | 62 | −32 | 4 | −6 |
| $\Delta^8$-THC | 17 | −51 | 67 | −48 | 15 | 2 |
| 11$\beta$-HHC | 55 | −61 | 63 | −60 | 56 | −53 |
| 11$\alpha$-HHC, chair | 54 | −61 | 62 | −59 | 54 | −50 |
| 11$\alpha$-HHC, twist | −29 | −34 | 66 | −31 | −31 | 64 |
| $\Delta^7$-THC | 2 | −30 | 59 | −66 | 39 | −7 |
| $\Delta^{9,11}$-THC | 54 | −61 | 62 | −59 | 57 | −53 |

In all molecules, ring B assumes a conformation such that the axial C6 methyl group is on the same side of the molecule as $H_{10a}$ and is much closer to $H_{10a}$ than is the other methyl group. The substituents on C6 and C6a are staggered with respect to one another. The optimized C10a–C6a–C6–O values were 62–63° for all of the molecules studied. Here the numbering system employed is the same as that for 1.

The phenyl group hydroxyl in all of the molecules is subject to steric interaction with the C10 proton(s). This interaction causes the phenyl group hydroxyl to bend slightly out of the plane of the benzene ring. The phenyl group hydroxyl optimized at a position pointing away from the carbocyclic ring. The optimized C2–C1–O–H values were 5–8° for all of the molecules studied.

Average optimized bond lengths for all molecules were within 0.01 Å of the following values: $C_{sp^2}$–$C_{sp^2}$ 1.34 Å, $C_{sp^2}$–$C_{sp^2}$ (aromatic) 1.40 Å, $C_{sp^2}$–$C_{sp^3}$ 1.51 Å, $C_{sp^3}$–$C_{sp^3}$ 1.54 Å, $C_{sp^2}$–O 1.37 Å, $C_{sp^3}$–O 1.39 Å, $C_{sp^2}$–H 1.10 Å, $C_{sp^3}$–H 1.12 Å, O–H 0.97 Å. Figures 2 and 3 illustrate a front view and a side view, respectively, of the optimized structure of $\Delta^7$-THC (4). This structure is included here as a representative example of the cannabinoid structures optimized in this study.

**Phenyl Group Hydroxyl Conformation.** Since we were interested in studying only molecules whose phenyl group hydroxyl positions were similar to those in $\Delta^9$-THC (1), we next screened all of the molecules for phenyl group hydroxyl position. For each molecule, we performed mo-

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Table II. Protrusion Torsion Angles

| molecule | activity[a] | angle 1, C10–C10a–C1–O, deg | angle 2, C8–C7–C1–O, deg | angle 3, C11–C9–C1–O, deg | angle 4, C9–Q–C1–O, deg |
|---|---|---|---|---|---|
| $\Delta^9$-THC | active[b] | −47 | −49 | −49 | −45 |
| $\Delta^8$-THC | active[b] | −49 | −34 | −38 | −33 |
| 11$\beta$-HHC | active[c] | −48 | −46 | −54 | 5 |
| 11$\alpha$-HHC | minimally active[c] | −48 | −45 | 48 | 2 |
| $\Delta^7$-THC | inactive[d] | −46 | −15 | 36 | −9 |
| $\Delta^{9,11}$-THC | inactive[e] | −47 | −45 | 7 | 7 |

[a] Activity as assessed in rhesus monkey behavioral tests. [b] See ref 14. [c] See ref 15. [d] See ref 16. [e] See ref 17.

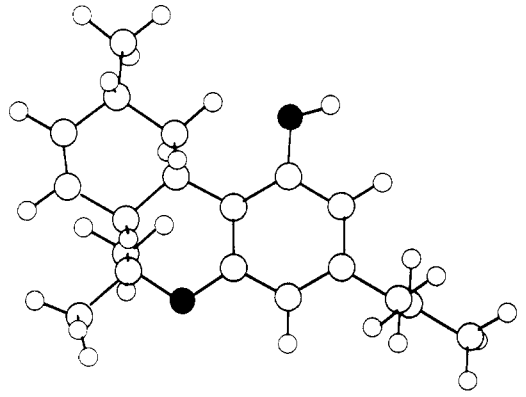

**Figure 2.** Front view of the conformation of $\Delta^7$-THC (**4**) (with propyl side chain) as determined by MMP2(85). The oxygens are shown as blackened circles.

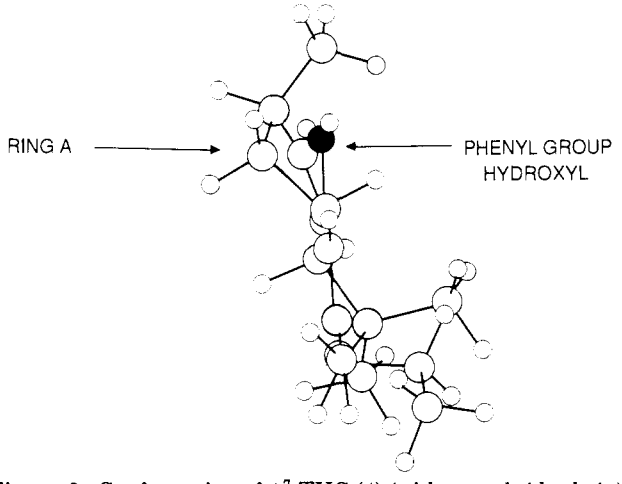


**Figure 3.** Conformation of $\Delta^7$-THC (**4**) (with propyl side chain) as determined by MMP2(85). See Figure 1 for details.

lecular mechanics dihedral driver studies using the MMP2(85) program.[23] We found that all of the molecules, including the twist conformation of **3b**, possessed two minima and that the location of these minima were very similar to those of **1**. In conformation I (C2–C1–O–H = 5–8°), the phenyl group hydroxyl is essentially in the plane of the aromatic ring. In this conformation the lone pairs of electrons of the oxygen point toward the carbocyclic ring. In conformation II (C2–C1–O–H = 160–170°), the lone pairs of the oxygen point away from the carbocyclic ring.

Figure 4 illustrates the rotational energy profile for $\Delta^7$-THC (**4**). This profile is included here as a representative example of the energy profiles obtained for all of the molecules studied. In the rotational energy profiles for the accessible conformations of all six cannabinoids, the first minimum was separated from the second by a 4.40–4.70 kcal/mol barrier. The energy profile was not symmetrical; thus the barrier separating the second minimum from the first was 3.10–3.90 kcal/mol (see Figure

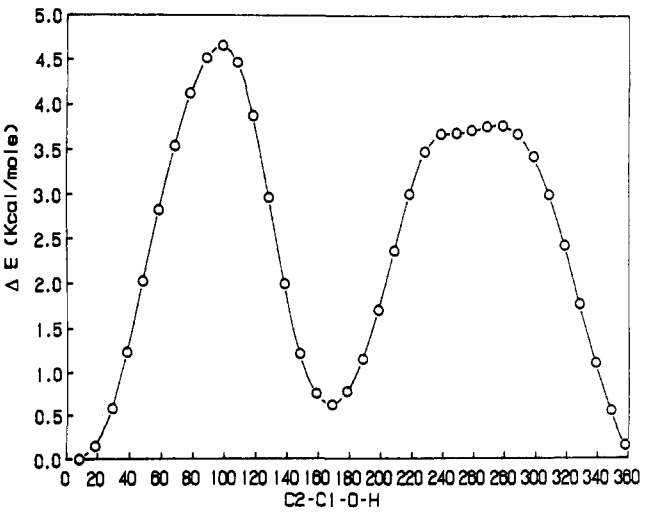


**Figure 4.** Rotational energy behavior of the phenyl group hydroxyl of $\Delta^7$-THC (**4**) as determined by an MMP2(85) dihedral driver calculation.

4, for an example). Conformation II was found to be 0.30–0.80 kcal/mol higher in energy than conformation I for the molecules studied.

The results of these rotational energy studies demonstrate that the position of the phenyl group hydroxyl in each of the molecules is similar to that of the phenyl group hydroxyl in **1**. Thus the variation of the activities of these compounds *cannot be attributed to phenyl group hydroxyl orientation.*

**Protrusion Torsion Angles.** Table II contains a summary of the results of "protrusion" torsion angle measurements for each of the absolute minimum-energy conformations of the investigated molecules. The molecules are arranged here in order of decreasing activity. We found a correlation between activity and the value of angle 3 (C11–C9–C1–O). All active cannabinoids possess a large negative value for angle 3. Since angle 3 measures the inclination of the C9 substituent (i.e., the C11 methyl group) with respect to the oxygen position, a negative value means that the methyl group protudes into the top face of the molecule (moves to the left of the phenyl group hydroxyl oxygen when viewed in sideways perspective).

As the value of angle 3 approaches zero, the molecules lose activity. A positive value of angle 3 results when the C9 substituent (i.e., the C11 methyl group) protrudes into the bottom face of the molecule (moving to the right of the phenyl group hydroxyl oxygen when viewed sideways). Figure 5 illustrates side views of the chair conformations of the hexahydrocannabinols **3a** and **3b**. As can be seen here, the C11 methyl group of the active isomer (**3a**, Figure 5A) moves to the left of the phenyl group hydroxyl oxygen (negative "protrusion" torsion angle 3), and the C11 methyl group of the isomer that is reduced in activity (3b, Figure 5B) moves to the right (positive "protrusion" torsion angle 3). The protrusion torsion angles were also measured in all molecules when the phenyl group hydroxyl was in its

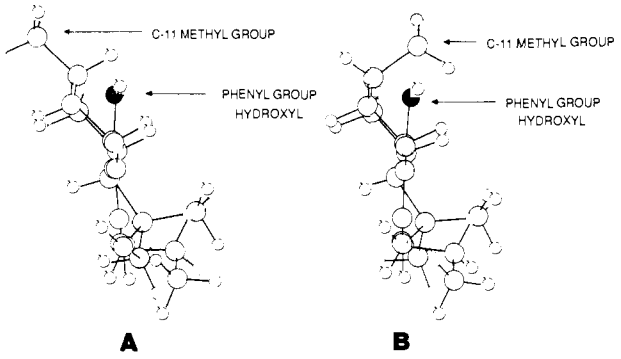


**Figure 5.** Side views of the MMP2(85)-optimized chair conformations (with propyl side chains) of (A) 11$\beta$-HHC (C11 methyl equatorial; **3a**) and (B) 11$\alpha$-HHC (C11 methyl axial; **3b**). See Figure 1 for details.

second minimum-energy position (conformation II). In each case the angles were found to vary less than 4° from those listed in Table II.

Protrusion torsion angle measurements were also performed for the twist conformation of 11$\alpha$-HHC (**3b**). In this case angles 1, 2, 3, and 4 were found to be −48°, −20°, −24°, and −82°, respectively. Clearly, angle 3 for the twist form of 11$\alpha$-HHC (**3b**) does not have a positive value as does its chair form. In this sense, the higher energy conformation of **3b** mimics the active cannabinoids $\Delta^9$-THC (**1**), $\Delta^8$-THC (**2**), and 11$\beta$-HHC (**3a**). Taking into account that ring A in **3b** can exist as a chair or a twist and that the phenyl group hydroxyl in each can exist in conformation I or II, there are four forms of **3b** that can be considered to exist. In form 1, ring A is in a chair conformation and the phenyl group hydroxyl is in conformation I. In Form 2, ring A is also in a chair conformation, but the phenyl group hydroxyl is in conformation II. In form 3, ring A exists in a twist conformation and the phenyl group hydroxyl is in conformation I. In form 4, ring A exists in a twist conformation also, but the phenyl group hydroxyl is in conformation II. Our studies of **3b** have revealed that form 2 is 0.54 kcal/mol higher in steric energy than form 1. Form 3 was found to be 1.72 kcal/mol above form 1, while form 4 was found to be 2.46 kcal/mol above form 1. By use of the Boltzmann relationship at 298 K and assuming no significant entropic or solvation differences, the relative amounts of forms 1–4 of 11$\alpha$-HHC (**3b**) would be 68%, 27%, 4%, and 1%, respectively. The fact that the twist conformation of **3b** (which exists in much smaller amounts) possesses a negative value of angle 3 may account for the minimal activity of this molecule. Thus each of the predominant forms of 11$\alpha$-HHC (**3b**) (forms 1 and 2) possesses a C11 methyl group that protrudes into the bottom face of the molecule (positive "protrusion" torsion angle) as does the C11 methyl group in $\Delta^7$-THC (**4**) and the C11 methylene group in $\Delta^{9,11}$-THC (**5**), both inactive cannabinoids. But each of the forms of 11$\alpha$-HHC present in much lower amounts (forms 3 and 4) has a C11 methyl group that protrudes into the top face (negative "protrusion" torsion angle) as does the C11 methyl group in $\Delta^9$-THC (**1**), $\Delta^8$-THC (**2**), and 11$\beta$-HHC (**3a**), all active cannabinoids.

## Conclusion

Our results indicate that, for cannabinoids which differ from $\Delta^9$-THC (**1**) only in the position or absence of a double bond in the carbocyclic ring (ring A), the orientation of the C9 substituent relative to the oxygen of the phenyl group hydroxyl is a determinant of activity. As measured by the "protrusion" torsion angle 3, those molecules whose C11 methyl groups (C9 substituents) point into the top face of the molecule are active (negative "protrusion" torsion angles). Those whose C11 methyl groups protrude into the bottom face (positive "protrusion" torsion angles) are inactive. This result is in keeping with Mechoulam's emphasis on the position of the C11 methyl group in cannabinoid compounds.[18]

It is important to emphasize that the C11 methyl group itself is not necessary for activity. The activity of desmethyl compounds such as 9-nor-$\Delta^9$-THC[27] clearly argues against such a posture. Here there is no substituent at C9, and the molecule is active. Instead, it seems that there may be a critical area near the top of the carbocyclic ring that must *not be blocked*. In this study, blocking was produced by the C11 methyl group (C9 substituent) in inactive or low-activity molecules. In principle, blocking could equally well be provided by another functional group. Our results, then, argue strongly for a steric requirement at the site of action. Protrusion into the bottom face may hamper or prevent molecules from binding at their site of action.

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