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CANNABINODIOL: CONCLUSIVE IDENTIFICATION AND SYNTHESIS OF A NEW CANNABINOID FROM *CANNABIS SATIVA*

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Key Word Index—*Cannabis sativa*; Cannabinaceae; cannabinoids; cannabinodiol.

Abstract—A constituent of Lebanese hashish was shown to be 2,6-dihydroxy-6′-isopropenyl-3′-methyl-4-*n*-pentyl-biphenyl, the aromatic analogue of cannabidiol. Synthesis and acid catalyzed conversion into cannabinol confirmed the assignment. This structure, cannabinodiol, was earlier erroneously assigned by others to a different compound; a suggestion for the correct structure of the latter product is given.

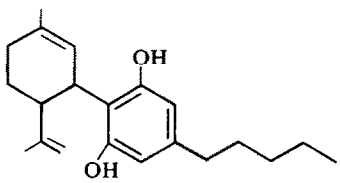
INTRODUCTION

In previous reports [1–4], we described the identification of several new cannabinoids. During GLC analysis of a large number of Cannabis preparations, it was found that accurate analysis of cannabidiol, a major cannabinoid, was frequently interfered with by the presence of an unknown compound sometimes present in substantial amounts.

Counter-current distribution was earlier applied as a convenient method for the preliminary separation of relatively large amounts of Cannabis preparations; subsequent GC–MS analysis showed that the unknown substance was present in the more polar fractions. The present paper describes the identification and synthesis of this unknown cannabinoid.

RESULTS AND DISCUSSION

The compound was purified by repeated column chromatography. The pure substance showed a somewhat shorter GC-retention time (R_x 0.91) than cannabidiol ((**1**), R_x 1.00). In its mass spectrum, the molecular ion was found at *m/e* 310 (11.5%) and the base peak at *m/e* 295, while most of the other fragment ions of the unknown compound occurred in nearly the same intensities as in the spectrum of cannabinol (**2**). The

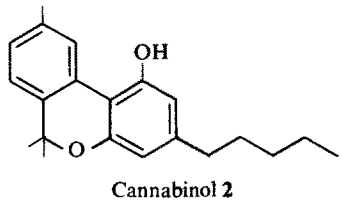


Cannabidiol **1**

molecular composition—as found by exact mass measurement—was found to be identical to **2**, $C_{21}H_{26}O_2$. However, trimethylsilylation of the unknown resulted in the formation of a *bis*-trimethylsilyl derivative, with a molecular ion at *m/e* 454 (310 + 2 × 72), thus suggesting the presence of two phenolic groups instead of one as in **2**.

A single coupling between the terpenoid and olivetol moiety was supported by the ^{1}H–NMR spectrum. A sharp singlet at δ 6.2 ppm was ascribed to two identical aromatic protons as occurring in e.g. **1**. The presence of the isopropenyl group was shown by signals at δ 4.82 (1H), 4.94 (1H) and 1.73 (3H) (compare similar resonances of **1** [5]. Resonances for the *n*-pentyl side chain were found in the appropriate regions. An aromatized terpene moiety, similar to **2**, was suggested by a methyl singlet at δ 2.34 ppm and additional signals in the—for cannabinoids—rather downfield aromatic region (δ 7.0–7.3 ppm). A 1,2,4-arrangement of these three aromatic protons was evident from the observed coupling constants. The above spectroscopic findings suggested structure **3**, 2,6-dihydroxy-6′-isopropenyl-3′-methyl-4-pentyl-biphenyl, for the unknown. The IR spectrum further supported this structure.

However, structure **3** was earlier assigned by Van Ginneken *et al.* [6] to a compound which they called cannabinodiol. Their product was observed by GC–MS measurements and it possessed, unlike our product, a longer retention time than **2**. Van Ginneken *et al.* observed great similarities between fragment ion intensities of their product and **2**. Therefore, the authors



Cannabinol **2**

assigned structure **3** to their product, based on the assumption that under the conditions of MS it was to a large extent ring-closed to cannabinol thus giving very similar spectra.

In order to clear up this confusion, it was decided to synthesize **3**. The issue seemed to require further clarification because of the interesting observation by Bowd

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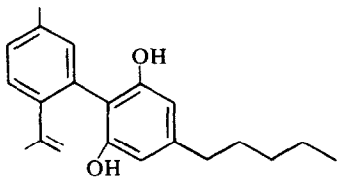
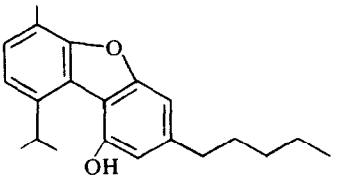


Cannabinodiol 3



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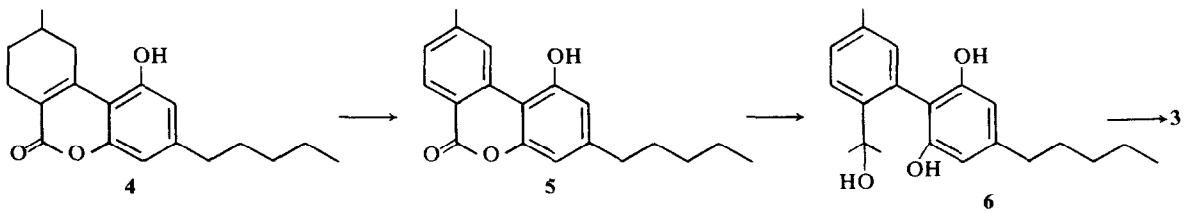
et al. [7] on the photochemical transformation of **2**. Irradiation of **2** yielded a product which Bowd *et al.* identified as the compound described by Van Ginneken. If our structure for the unknown were correct, Bowd *et al.* should have isolated a different compound from this. In fact, irradiation of cannabinol yielded about 50% of a product which, after purification, was in all aspects identical to our unknown.

The total synthesis of **3** was carried out as outlined in the Scheme. Product **4** was prepared according to Adams and Baker [8]; aromatization to **5** was achieved both by treatment with sulphur and 1,2-dichloro-4,5-dicyano-quinone (DDQ) [9] as well. **5** was treated with an excess of methyl-Grignard and **6** was obtained in an almost quantitative yield. Dehydration of **6** by flash-distillation yielded mainly two products, cannabinol and **3**, a compound which was identical in all respects with our isolated product.

Finally, acid catalyzed cyclisation under comparable conditions to those earlier applied to the conversion of **1** into its isomer $\Delta 1^{(2)}$-tetrahydrocannabinol [10, 11] yielded, both in the case of the isolated and the synthetic product, a compound which was identical with an authentic sample of cannabinol.

It can thus be concluded that the product both from our hashish sample and from the photochemical conversion of cannabinol is **3**. Consequently it must be concluded that the product reported by Van Ginneken *et al.* [6] as **3** must have a different structure. During other experiments [12], a product was isolated possessing MS and chromatographic properties closely resembling those described by Van Ginneken *et al.* Measurement of the ^{1}H–NMR spectrum of this compound suggested structure **7** for the product. Quite independently Friedrich–Fiechtl and Spiteller [13, 14] have suggested that the product described by Van Ginneken *et al.* is **7**. It can now be concluded that cannabinodiol **3** does in fact occur naturally.



Scheme

EXPERIMENTAL

Isolation procedure. 'Red' Lebanese hashish was extracted by hexane–Et_2O (1:1). The extract was concentrated to a dark syrup. 74 g of this residue was partionated by counter current distribution (175 steps) as earlier described [1]. The fractions 1–28 were combined and concentrated. The residue (24.5 g) was chromatographed over several columns (SiO_2 'Merck' for TLC) using *n*-hexane–Et_2O (9:1). The pure unknown was obtained as a colourless oil. GLC: R_x 0.91 (R_x cannabidiol 1.00) on a 1.80 m glass column of 3% OV-17 on Chromosorb G AW-DMCS 100–120 mesh. TLC: R_f 0.37 on precoated thin-layer plates 'Merck', eluent *n*-hexane–Et_2O (4:1), R_f cannabidiol 0.58. Spraying with an alkaline solution of Fast Blue salt B gives a typical orange colour. 100 MHz PMR-spectrum (in CCl_4): 0.90 (*t*, J = 6.0 Hz) (ω-Me), 1.73 (*d*, J = 1.0 Hz) (olefinic Me), 2.34 (*s*) (aromatic Me), 2.48 (*t*, J = 7.5 Hz) (benzylic CH_2), 4.83 and 4.94 ($C{=}CH_2$), 6.20 (*s*) (2 aromatic H's), 7.04 (1H) and 7.14–7.29 (2H's) ppm. MS (70 eV): 310 (11.5%), 295 (100%), 296 (22%), 238 (15%), 223 (15%), 165 (13.5%), 128 (10.5%), 115 (10%), and a very characteristic fragment at *m/e* 293 (9%).

Conversion to **2**. 0.5 mg of the above compound in 2 ml EtOH–0.01 N HCl was refluxed at 100° for 2 hr.

1-Hydroxy-9-methyl-6-oxo-3-pentyl-6H-dibenzo[b,d]pyran (**5**). Compound **4** was aromatized by treatment with S [8] and dichloro-dicyanoquinone (DDQ) [9], as earlier described. Using 50% excess of DDQ in C_6H_6 for 3 hr, gave the title product in a 80–84% yield (GC determined). Treatment of 4 with S gave a reaction product which was heavily contaminated with S. Mp 162°; IR (CCl_4) 3300, 2960, 1690, 1670, 1620, 1610, 1490 cm^{-1}; δ (60 MHz, CCl_4) 10.98 (1H), 8.9 ((1H), 8.1 (1H, J 8 Hz), 7.38 (1H, J 8 Hz), 6.70 (2H), 2.42 (5H), 1.30 (6H), 0.85 (3H); ms *m/e* 296, 253, 244, 241, 239 (100%).

2,6-Dihydroxy-6'-(2-hydroxy-isopropyl)-3'-methyl-4-n-pentyl-biphenyl (**6**). A methyl Grignard was prepared from 400 mg Mg in 10 ml Et_2O and 1 ml MeI in 25 ml Et_2O. A solution of 200 mg 5 in 10 ml Et_2O was added and the mixture was refluxed for 1 hr and left for 18 hr at room temp. The reaction mixture was then poured into a 10% NH_4Cl and the organic layer was extracted with hexane and evaporated, yield 193 mg. Purification was on pre-coated SiO_2 plates with hexane–Et_2O 4:1 (R_f 0.06). IR (CCl_4) 3600, 3555, 3400, 2960, 1740, 1640, 1565, 1490 cm^{-1}; δ (60 MHz, CCl_4) 7.45 (1H, J 8 Hz), 7.05 (1H, J 8 Hz), 6.9 (1H), 6.25 (2H), 6.1 (1H), 5.7 (2H), 2.3 (5H), 1.5–1.1 (12H), 0.9 (3H) ppm; MS identical to cannabinodiol, probably due to spontaneous dehydration under MS conditions.

Cannabinodiol, 2,6-dihydroxy-6'-isopropenyl-3'-methyl-4-n-pentyl-biphenyl (**3**). **6** (15 mg) was deposited as a thin film at the lower part of the inner-wall of a glass tube by evaporation of its ethereal solution. Air was replaced by N_2 and finally the tube was evacuated (10 mm Hg). The lower part of the reaction tube was then placed for 20 sec. in an oil bath at 200°. The reaction products condensed on the upper part of the glass wall. Thus TLC gave **3** in a 20% yield. Preparative TLC was carried out on pre-coated SiO_2 plates by three consecutive runs, using hexane–Et_2O (4:1).

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