Phytochemistry, 1970, Vol. 9, pp. 1887 to 1889. Pergamon Press. Printed in England.

SHORT COMMUNICATION

GLAUCARUBOL AND GLAUCARUBOL-15-ISOVALERATE FROM *CASTELA NICHOLSONI**

W. STÖCKLIN and T. A. GEISSMAN

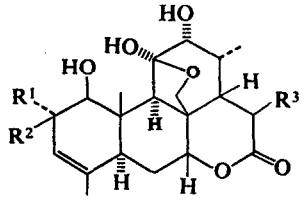
Department of Chemistry, University of California, Los Angeles 90024, U.S.A.

(*Received* 28 *October* 1969)

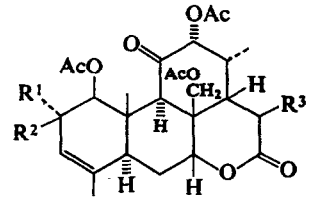
Abstract—Crude chaparrin from *Castela nicholsoni* Hook was acetylated and then chromatographed on silica gel. Two minor constituents, glaucarubol and glaucarubol-15-isovalerate, were isolated and identified as their per-*O*-acetyl derivatives.

INTRODUCTION

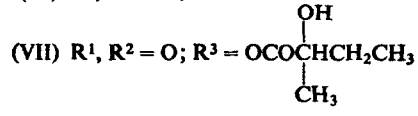
THE MAJOR component of *Castela nicholsoni* Hook (Simarubaceae) is chaparrin (I),[1] whose structure has been established.[2] The only minor constituent so far isolated and properly identified from this plant is glaucarubolone (II).[3] The presence of glaucarubol (III) had been noted but not established conclusively in earlier work.[1] While preparing pure acetates from crude chaparrin, we have now isolated glaucarubol pentaacetate (IV) and glaucarubol-15-isovalerate tetraacetate (V).



(I) $R^1 = OH$; $R^2 = R^3 = H$
(II) $R^1, R^2 = O$; $R^3 = OH$
(III) $R^1 = R^3 = OH$; $R^2 = H$



(IV) $R^1 = R^3 = OAc$; $R^2 = H$
(V) $R^1 = OAc$; $R^2 = H$; $R^3 = OCOCH_2CH(CH_3)_2$
(VI) $R^1, R^2 = O$; $R^3 = OAc$



(VII) $R^1, R^2 = O$; $R^3 = OCOCHCH_2CH_3$ (CH_3 branch)

* Contribution No. 2484 from the Department of Chemistry, U.C.L.A.

[1] T. A. GEISSMAN and K. R. CHANDORKAR, *J. Org. Chem.* **26**, 1217 (1961).
[2] T. A. GEISSMAN and G. A. ELLESTAD, *Tetrahedron Letters* 1083 (1962); T. A. DAVIDSON, T. R. HOLLANDS and P. DE MAYO, *Tetrahedron Letters* 1089 (1962); T. A. DAVIDSON, T. R. HOLLANDS, P. DE MAYO and M. NISBET, *Can. J. Chem.* **43**, 2996 (1965); T. R. HOLLANDS, P. DE MAYO, M. NISBET and P. CRABBÉ, *Can. J. Chem.* **43**, 3008 (1965).
[3] J. POLONSKY and N. BOURGUIGNON-ZYLBER, *Bull. Soc. Chim. France* 2793 (1965).

RESULTS AND DISCUSSION

Crude chaparrin from an earlier investigation[1] was acetylated with acetic anhydride and pyridine. Chromatography of this material on silica gel gave besides different acetates of chaparrin, two crystalline compounds, A and B, in yields of 2 and 0·1 %, respectively.

Compound A, m.p. 203–204° was identified as glaucarubol penta-acetate (IV) by TLC, mixed m.p. with an authentic sample,[4] and examination of the NMR spectra.

Compound B, m.p. 238–239·5°, was found to be glaucarubol-15-isovalerate tetraacetate (V). Its close relationship to glaucarubol pentaacetate was indicated by the NMR spectrum, which differed essentially only at three points from that of glaucarubol pentaacetate. In particular, it showed the presence of only four acetoxy groups; the signal at 2·16 ppm in the NMR spectrum of glaucarubol pentaacetate was missing. Instead of a doublet at 1·03 ppm (3 H, $J = 6{\cdot}5$ c/s; 13–CH_3) in the NMR spectrum of glaucarubol pentaacetate, a complex nine-proton signal was observed between 0·90 and 1·10 ppm. Furthermore, the position of the signal of the proton at C-15 appeared at 6·08 ppm (d, $J = 10$ c/s) in compound B, whereas it appears at 6·20 ppm (d, $J = 10{\cdot}5$ c/s) in glaucarubol pentaacetate.[4]

These results indicated that the 15-hydroxy group is not esterified with acetic acid but with another acid which produces the signals around 1 ppm in the NMR spectrum. The mass spectrum, showing strong peaks at *m/e* 85 (C_5H_9O), established the presence of a saturated C_5 acid. By comparing the NMR spectra of all possible saturated C_5 acids with that of compound B the presence of an isovaleroyl residue could be established. Although the secondary methyl groups of the isovaleric acid residue of compound B do not appear as a clear doublet at 0·94 ppm ($J = 6$ c/s) as in isovaleric acid, they appear at the correct chemical shift. In glaucarubol-15-isovalerate tetraacetate the secondary methyl groups are in an asymmetric environment and therefore are not expected to be equivalent.

The upfield shift of the position of the signal of the C-15 proton corresponds to that observed in comparing glaucarubolone tetraacetate (VI) (H-15 at 6·15 ppm) and glaucarubinone triacetate (VII) (H-15 at 6·00 ppm).[5] It is to be noted, however, that in other examples (steroids) no upfield shift was observed by comparing C*H*OAc and C*H*OCOCH$_2$(CH$_3$)$_2$.[6] The isolation of glaucarubol pentaacetate and glaucarubol-15-isovalerate tetraacetate establishes the presence of glaucarubol and glaucarubol-15-isovalerate in *Castela nicholsoni*, although it cannot be completely excluded that these compounds may also have been present in the plant in an already partly acetylated form.

Castela nicholsoni Hook ("chaparro amargosa") has been used as antiamebic drug in Mexico.[7] Since neither chaparrin nor glaucarubol showed antiamebic activity in *in vitro* experiments,[8] it will be interesting to test glaucarubol-15-isovalerate which has—as has the amebicidal glaucarubin[9] (structure II but $R^3 = OCOCOHCH_3CH_2CH_3$)—an ester residue at C-15.

EXPERIMENTAL

Acetylation of Crude Chaparrin

40 g of crude chaparrin from an earlier investigation[1] was suspended in 250 ml pyridine and 200 ml Ac_2O and the mixture allowed to stand at 23° for 48 hr. After destroying the excess Ac_2O with methanol the solvents were evaporated. Initial chromatographies on alumina and on silica gel allowed the separation of some chaparrin tetra-acetate and chaparrin triacetate.[2]

[4] W. STOCKLIN, L. B. DE SILVA and T. A. GEISSMAN, *Phytochem.* **8**, 1565 (1969).
[5] A. GAUDEMER and J. POLONSKY, *Phytochem.* **4**, 149 (1964).
[6] H. H. SAUER, EK. WEISS and T. REICHSTEIN, *Helv. Chim. Acta* **49**, 1655 (1966).
[7] M. MARTINEZ, *Las Plantas Medicinales de Mexico*, Ediciones Botas Mexico (1944).
[8] N. ENTNER, personal communication.
[9] J. DRUEY, *Angew. Chem.* **72**, 677 (1960).

Isolation of Glaucarubol-15-isovalerate Tetraacetate and Glaucarubol Pentaacetate

The remaining acetate mixture (21·9 g) was adsorbed on 40 g of silica gel and chromatographed on 450 g of silica gel with fractions of 25 ml. Fractions 71–120 (eluted with EtOAc–petrol. ether, 1:2; 165 mg) yielded, after crystallization from EtOAc–petrol. ether and ether–petrol. ether, 43 mg of glaucarubol-15-isovalerate tetra-acetate. Fractions 121–170 (same eluant, 1070 mg) yielded, after 2 crystallizations from ether–petrol. ether, 914 mg of glaucarubol pentaacetate. The further fractions give different acetates of chaparrin.

Characterization of the Isolated Compounds

Glaucarubol-15-isovalerate tetraacetate. M.p. 238–239·5°, $\alpha_D^{24} + 27{\cdot}9°$ (*c*, 0·47 in pyridine). (Calc. for $C_{33}H_{44}O_{13}$: C, 61·10; H, 6·84. Found: C, 61·35; H, 6·90%.) The m.s. showed the following principal peaks: 588 (M–AcOH); 546 (588–CH_2CO); 531 (546–CH_3); 503 (588–85); 443 (503–AcOH); 425 (485–AcOH); 401 (443–CH_2CO); 383 (425–CH_2CO); 359 (401–CH_2CO); 341 (401–AcOH); 85 (C_5H_9O); 60 (AcOH); 57 (C_4H_9); 43 (CH_3CO). The NMR spectrum (100 Mc, in $CDCl_3$ with TMS as internal standard) showed the following signals: *ca.* 1·04 ppm (3 H, $J = 6{\cdot}5$ c/s; 13-CH_3); *ca.* 1·00 ppm and 1·04 ppm (each 3 H, d, $J = 6{\cdot}5$ c/s; $CH(CH_3)_2$); 1·43 ppm (3 H, s; 10-CH_3); 1·69 ppm (3 H, s, br; 4-CH_3); 1·81 ppm; 1·97 ppm; 2·08 ppm; 2·24 ppm (each 3 H, s; 4 acetoxy groups); 5·04 ppm (1 H, d, $J = 8$ c/s; H-1); *ca.* 5·37 ppm (1 H, m; H-2); 5·41 ppm (1 H, s, br; H-3); 4·72 ppm (1 H, t-like; H-7); 3·86 ppm and 4·67 ppm (each 1 H, d, J = 12·2 c/s. 8-CH_2O); 3·33 ppm (1 H, s; H-9); 5·07 ppm (1 H, d, $J = 2{\cdot}5$ c/s; H-12); 6·08 ppm (1 H, d, $J = 10{\cdot}5$ c/s; H-15). Chemical shifts are given in ppm on the δ-scale, coupling constants in c/s; s = singlet, d = doublet; t = triplet; m = multiplet; br = broad signal. Insufficient material was available for further characterisation of the isovaleric acid residue in V.

Glaucarubol pentaacetate. M.p. 203–204° (not depressed on admixture with authentic material); $[\alpha]_D^{23} + 28{\cdot}7°$ (*c*, 0·48 in pyridine) (reported, +23° in pyridine;[10] +25·4° in pyridine.[11] (Calc. for $C_{30}H_{30}O_{13}$: C, 59·40; H, 6·31. Found: C, 59·27; H, 6·41%.) The NMR spectrum was identical with the reported values.[4] The circular dichroism curve (*c*, 0·057 in methanol) showed maxima at 309 nm ($[\theta]_{309} = -3210$) and 237 nm ($[\theta]_{237} = -2580$).

Acknowledgement—The 100 Mc NMR spectrum was recorded by Mrs. Dr. G. Chmurny, Department of Chemistry, UCLA, to whom we are very grateful. These studies were supported by a research grant AI-07435 from the U.S. Public Health Service and part of the costs of the mass spectrometer and NMR spectrometer was provided by a grant to the Department of Chemistry from E. I. du Pont de Nemours and Compagny and by the National Science Foundation. Analyses are by Miss Heather King, Department of Chemistry, UCLA.

[10] A. Ham, H. M. Schafer, R. G. Denkewalter and N. G. Brink, *J. Am. Chem. Soc.* **76**, 6066 (1954).
[11] J. Polonsky, C. Fouquey and A. Gaudemer, *Bull. Soc. Chim. France* 1818 (1964).