Phytochemistry, 1977, Vol. 16, pp. 790–791. Pergamon Press. Printed in England

TRITERPENOIDS OF *SALVIA HORMINUM*, CONSTITUTION OF A NEW DIOL

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(*Revised received 16 December 1976*)

Key Word Index—*Salvia horminum*; Labiatae; lupeol; lup-(20)29-ene-2α,3β-diol; olean-(13)18-ene-2β,3β-diol.

Abstract—Lupeol, lup-(20)29-ene-2α,3β-diol and a new triterpenic alcohol olean-(13)18-ene-2β,3β-diol were isolated from the petrol extract of air dried *Salvia horminum* and their structures were determined.

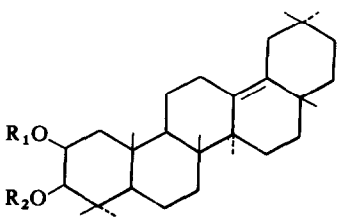
INTRODUCTION

In a previous study the presence of micromeric, ursolic and oleanolic acids in the chloroform extract of *Salvia horminum* was described [1]. A further investigation of the plant has led to the isolation of three triterpenic alcohols, one of them being new.

RESULTS AND DISCUSSION

Salvia horminum was collected from the Mediterranian coast of Turkey and identified by Prof. Dr. A. Baytop (Istanbul). A voucher sample, ISTE 8032, is deposited in the herbarium of the Faculty of Pharmacy, University of Istanbul.

Silica gel column chromatography of the petrol extract of the plant yielded lupeol, lup-(20)29-ene-2α,3β-diol and a new oleanene diol. The new diol (**1**) was obtained from the polar fractions eluted from the column, mp 228°, $C_{30}H_{50}O_2$, ν_{max}^{KBr} showed hydroxyl (3450 cm^{-1}) and a gem dimethyl (1370 cm^{-1}). NMR (CD_3OD, TMS) gave methyl singlets at δ0.79, 0.82, 0.94, 1.00, 1.06, 1.12 and 1.20 corresponding to eight methyls, two hydroxyl protons (δ2.25, 2H, *br.s*), two carbinol methine protons (δ3.5, 1H, *d*, J = 6.5 Hz, 3αH and δ4.5, 1H, *d*, J = 3 Hz, 2αH), no vinylic proton was present. Acetylation of the diol yielded a diacetate (**2**) mp 114–115°, ν_{max}^{KBr} ester (1725 cm^{-1}). NMR ($CDCl_3$, TMS) showed acetyl bands



1—$R_{1,2}$ = H
2—$R_{1,2}$ = Ac
3—$R_{1,2}$ = —O—O—$(Me)_2$

(δ1.93 and 2.02, 3H each, *s*). MS gave the characteristic peaks of olean-(13)18-ene fragmentation at *m/e* 442 (M^+), 424 (M–18), 218 and 205 [2].

The fragmentation pattern of the new diol indicated that two OH groups were located at rings *A/B*. The diol reacted with periodate and formed an acetonide (**3**) showing the presence of two hydroxyls on vicinal carbon atoms. Jones oxidation formed a diosphenol, the UV spectrum showed two maxima, one at 275 nm and the other at 256 nm. The former correlates with the 2, 3 positions of the hydroxyl groups, whereas the latter might come from the formation of keto group(s) conjugated with the 13(18) double bond. In order to confirm this point a sample of δ-amyrin was subjected to Jones oxidation and its UV spectrum was taken under the same conditions. A maximum at 257 nm clearly showed the same type of oxidation for the (13)18 double bond.

MS fragmentation of the sample of δ-amyrin gave peaks at *m/e* 426 (M^+), 408 (M-18), 218 and 205, which are in agreement with the MS of the new diol.

A study of the NMR spectrum of olean-(13)18-ene diol reveals the β, β positions of the OH groups. A doublet at δ3.5 J_{ae} = 6.5 Hz showed an equatorial OH group at C-3, on the other hand the doublet at δ4.15 J_{ea} = 3 Hz showed that the OH group at C-2 should be axial. As reported by Williams and Bhacca [3] the J_{ae} values denoting the coupling of an axial proton on the atom bearing the electro-negative substituent to an equatorial proton were observed to be of the order 6 cps, whereas J_{ea} values denoting the analogous coupling in which the proton on the electronegatively substituted carbon atom is equatorial, were of the order of 2–3 cps. The structure of the new oleanene diol was therefore concluded to be olean-(13)18-ene-2β,3β-diol (**1**).

Although lup-(20)29-ene-2α,3β-diol and its 2β, 3β isomer were isolated from *Pterocarpus santalinus* [4, 5] for the first time, this is the first time it was found in a *Salvia* species, olean-(13)18-ene-2β,3β-diol was also a new diol for salvia species.

EXPERIMENTAL

Mps were uncorrected, NMR spectra were taken at 60 MHz.

Extraction and isolation. The dried and powdered (2 kg) herbage of *S. horminum* was first macerated then percolated with petrol (40–60°), $CHCl_3$ and EtOH successively. The petrol extract was chromatographed on a Si gel column and eluted with various solvents. C_6H_6–$CHCl_3$ (1:1) fractions yielded lupeol. $CHCl_3$ elution gave lupenediol and $CHCl_3$–EtOH (9:1) fractions gave oleanenediol.

Lupeol. mp 215°; IR ν_{max}^{KBr} cm^{-1}: 3340(OH), 1640, 875 (=CH_2); NMR ($CDCl_3$, TMS); δ0.76, 0.79, 0.84, 0.96 and 1.02 (six methyl singlets), 1.66 (vinylic methyl, 3H, *s*), 3.16 (carbinol hydrogen, 1H, *d*, J = 6 Hz), 4.54 and 4.66 (=CH_2, 1H each, *d*, J = 1.5 Hz). Acetylation formed a monoacetate, mp 205°; IR ν_{max}^{KBr} cm^{-1}: 1735, 1250 (ester). NMR ($CDCl_3$, TMS): δ 2.00 (acetyl, 3H, *s*), C-3 proton shifted to δ4.4 ppm (1H, *d*, J = 5.5 Hz). MS *m/e*: 426 (M^+), base peak at 218, other peaks at 220, 189 and 207 [6]. Jones oxidation yielded lupen-3-one, mp 169–170°. Reduction of lupen-3-one with $NaBH_4$ gave mainly lupeol and a small amount of epilupeol. Comparison with an authentic sample confirmed the product as lupeol.

Lup-(20)29-ene-2α,3β-diol. mp 234°; $C_{30}H_{50}O_2$ M^+ 442; IR ν_{max}^{KBr} cm^{-1}: 3340(OH), 1380(gem dimethyl), 880, 1640(=CH_2); NMR ($CDCl_3$, TMS): δ0.79, 0.88, 0.93, 0.98, 1.02 (tertiary methyl singlets), 1.68 (3H, *s*, vinylic methyl), 3.2 (1H, *dd*, J = 10 Hz, 11 Hz) and 3.55 (1H, *m*, $W_{\frac{1}{2}}$ 20 Hz) (3αH and 2βH respectively), 4.55 and 4.70 (1H each, *d*, J = 1.5 Hz, exo = CH_2). MS showed diagnostically important peaks for lupeol derivatives at *m/e* 442 (M^+), 427 (M–Me), 424 (M-18), 236, 223, 218, 189, 203, 218, 205 [2]. Acetylation yielded a diacetate, mp 131°; $C_{34}H_{54}O_4$, M^+ 526; IR ν_{max}^{KBr} cm^{-1}: 1740 (ester), 885, 1640 (=CH_2); NMR ($CDCl_3$, TMS): δ 1.92 and 2.01 (3H each, *s*, acetyl). Jones oxidation yielded a diosphenol, UV λ_{max} 273 nm (ε 7800); IR ν_{max}^{KBr} cm^{-1}: 885, 1650 (=CH_2), 1385 (gem dimethyl), 3450 (OH); NMR ($CDCl_3$, TMS) δ0.78, 0.82, 0.95, 1.01, 1.20 (six tertiary Me, *s*), 1.68 (3H, *s*, vinylic Me), 4.50 and 4.65 (=CH_2), 6.29 (1H, *s*, vinylic proton, C-1).

Olean-(13)18-ene-2β,3β-diol. The new oleanenediol (160 mg), crystallized from EtOH, mp 228° (Found C, 80.95; H, 11.05. $C_{30}H_{50}O_2$ requires: C, 81.40; H, 11.40%). IR ν_{max}^{KBr} cm^{-1}: 1385 (gem dimethyl), 3450 (OH). NMR and MS described in the results. A diacetate was formed upon acetylation, mp 114° (Found: C, 77.95; H, 10.27. $C_{34}H_{54}O_4$ requires: C, 77.50; H, 10.30%) IR ν_{max}^{KBr} cm^{-1} 1385 (gem dimethyl), 1725, 1250 (ester); NMR ($CDCl_3$, TMS); δ0.8, 0.86, 0.98, 1.10 and 1.20 (8 tertiary Me, *s*), 1.94 and 2.02 (3H, each, *s*, 2 × OCOMe), 4.50 (1H, *d*, J = 6.5 Hz, 3αH), 5.20 (1H, *d*, J = 2.5 Hz, 2αH).

Olean-(13)18-ene-2β,3β-acetonide (3). 30 mg of the diol dissolved in 1 ml dry Me_2CO was added to 200 mg *p*-toluensulfonic acid in Me_2CO and kept at room temp. for 1 hr. Purification on a small column of Si gel gave the amorphous acetonide (Found: C, 81.95; H, 11.35. $C_{33}H_{54}O_2$ requires: C, 82.10; H, 11.20%). IR ν_{max}^{KBr} cm^{-1} 858, 1050, 110, 1158 (acetonide); NMR ($CDCl_3$, TMS): δ0.78, 0.82, 0.94, 1.00, 1.11, 1.20 (eight tertiary Me, *s*), 1.38 and 1.42 (3H each, *s*, methyls of acetonide), 3.52 (1H, *d*, J = 6.5 Hz, 3αH) and 4.2 (1H, *d*, J = 2.5 Hz, 2αH).

Diosphenol. 50 mg of the diol dissolved in 5 ml Me_2CO was treated with CrO_3–AcOH (25 mg of CrO_3 in 1 ml AcOH) at room temp. for 5 min, then diluted with H_2O, extracted with Et_2O and the residue purified by PLC (Si gel G, $CHCl_3$–C_6H_6), mp 263°, +$FeCl_3$ reaction, UV λ_{max}^{EtOH} nm: 275 (ε 7600) and 256 (ε 12000); IR ν_{max}^{KBr} cm^{-1}: 3430 (OH), 1380 (gem dimethyl), 1725, 1630 (carbonyl bands); NMR ($CDCl_3$, TMS): δ0.79, 0.84, 0.96, 1.05, 1.12, 1.24 (eight tertiary Me, *s*), 6.33 (1H, *s*, C-1).

Acknowledgement—One of the authors (AU) thanks DAAD for a three months grant for the work at the Faculty of Pharmacy (Würzburg). The authors would like to thank Dr. B. S. Joshi, Ciba Research Center (Bombay) for samples of lupeol and lupen-3-one.

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Phytochemistry, 1977, Vol. 16, pp. 791–793. Pergamon Press. Printed in England.

STRUCTURE OF BAROGENIN FROM *SOLANUM TUBEROSUM*

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(*Revised received* 14 *December* 1976)

Key Word Index—*Solanum tuberosum*; Solanaceae; steroidal sapogenin; kryptogenin; barogenin; spirostan biosynthesis.

Abstract—The budding tuber of *Solanum tuberosum* accumulated barogenin. Its structure was determined by chemical and spectroscopic studies as (25*S*)-3β,26-dihydroxy-cholest-5-ene-16,22-dione, the (25*S*)-epimer of kryptogenin. The biogenetic relationship between barogenin and spirostanols is discussed.

INTRODUCTION

The biogenesis of spirostanols, for example, diosgenin (**1a**) and yamogenin (**1b**), is presently under investigation in several laboratories [1–7]. Most of the hypothesis, based on the results of feeding experiments, suggest that cholesterol (**2a**) is oxygenated at C-26 (**2b**), C-16, and C-22, in that order, and then enzymatically converted to a spirostan. However, there is no report on the presence of these compounds, and the presence of 26-hydroxycholesterol (**2b**) or its derivatives which carry an oxygen function at C-16 or C-22 has not been recorded in plants which contain spirostanol. Therefore it seemed significant to