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TERPENOIDS OF THE MICROMERIAS.-I. TWO NEW TRITERPENIC ACIDS ISOLATED FROM MICROMERIA BENTHAMI WEBB ET BERTH.

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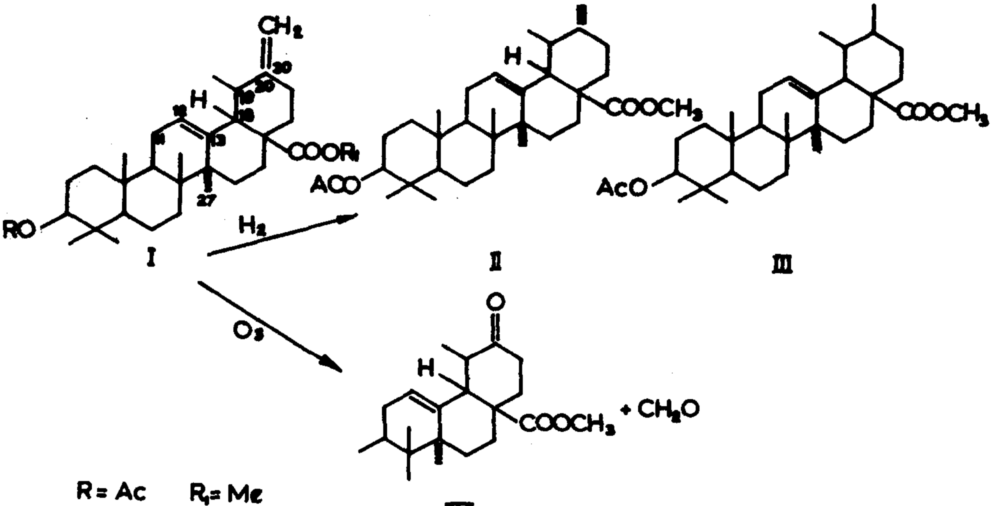
An acidic product isolated from the aerial parts of Micromeria Benthami WEBB et BERTH. (labiatae), a plant endemic to the Canary Isles, showed an intense violet colour in the Liebermann-Burchard reaction.

Chromatography of the methyl ester of this crude product on silica gel, yielded first with $C_6H_6/CHCl_3$ (3/1) the methyl ester of betulinic acid (m.p. 219-222º, $[\alpha]_D^{20}$ + 3.5º).

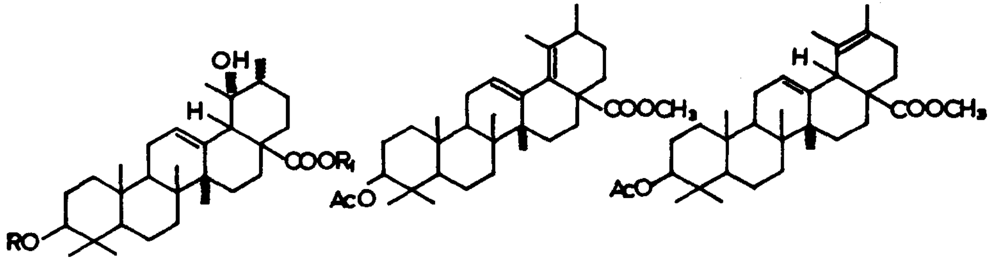
Secondly, a mixture of methyl esters (GLC) was obtained which acetylation after were rechromatographed on silica gel impregnated $AgNO_3$ (1) and gave, in the petroleum ether/C_6H_6 (4/1) fractions, a crystalline mass. This consisted of two substances which were separed by fractional crystallization and identified, by means of IR spectra, derivatives, and by comparison with original samples (mixed m.p.; IR identical), as the methyl ester acetates of ursolic and oleanolic acids, benzene eluted a new compoud I, the methyl ester acetate of micromeric acid,



I II III

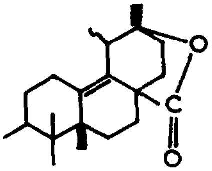
IV

R = Ac R1 = Me

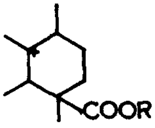


V VI VII

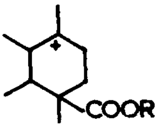
Va R = R1 = H
Vb R = H, R1 = Me
Vc R = Ac R1 = Me



VIII



IX



X

$C_{33}H_{50}O_4$, m.p. 181-183º, $[\alpha]_D^{20}$ + 127º. IR 3090, 1650 and 900 ($=CH_2$), 1730 (acetate and methyl ester), 835 and 827 cm^{-1} (trisubstituted double bond), NMR spectrum TABLE I.

TABLE I

(τ -values)

Compound	H_3	H_{12}	H_{18}	COOMe	MeCO	Me27	Me29	$C_{20}=CH_2$
I	5.46	4.70		6.40	7.94	8.85		5.32
II	5.48	4.73		6.38	7.94	8.91		
III	5.48	4.72		6.40	7.96	8.90		
IV	5.46	4.66		6.34	7.96	8.85		
V_c	5.45	4.65	7.37	6.38	7.95	8.78	8.75	
VI	5.50	4.64		6.40	7.95		8.30	
VII	5.55	4.55	7.13	6.40	7.95		8.37 and/or 8.53	
VIII	5.50				7.95	8.85		

Hydrogenation of I with $Pd/CaCO_3$ in ethyl acetate produced a mixture of two substances. One of them was identified (mixed m.p.; IR identical) as acetyl methyl ursolate II, and formed the basis for the general structure of the product.

The position of the methylene group was established by ozonolysis, which yielded formaldehyde (identified by means of its dimedone and 2,4-dinitrophenylhidrazine derivative) and a norketone, m.p. 236-247º, $[\alpha]_D^{20}$ + 71.5º. The deep violet colour of the latter with the Zimmermann reagent, as well as its condensation with ethyl formate, indicated the presence of a $-CO-CH_2-$ group, in accordance with structure IV proposed for the norketone. Consequently, the position of the methylene group must be at C_{20}.

This conclusion was supported by the NMR spectrum which does not show the diamagnetic shift to 0.14 ppm characteristic of the Me-27 in pentacyclic triterpenes with $C_{19}=O$ (2).

The second substance obtained by hydrogenation of I has m.p. 173-175ºº, $[\alpha]_D^{20}$ + 75º. Its NMR and IR spectra are very similar to those of II and it was assigned structure III, i.e. methyl ester acetate of 20β-Me ursolic acid, in accordance with the following: hydrogenation of I should give a mixture of C_{20}-epimers; the 20α-Me compound is the methyl ester acetate of ursolic acid with C_{19}-β Me, C_{20}-α Me; hence, III has the same configuration at C_{19} but the opposite one at C_{20}: C_{19}-β Me, C_{20}-β Me.

Consequently, ring E of substance I is like the one in taraxasterol, but with 18β-H configuration.

Further elution of the chromatograph with $C_6H_6/CHCl_3$ (65/35) yielded the methyl ester of a new diol V_b, benthamic acid methyl ester:[+] $C_{31}H_{50}O_4$, m.p. (from petroleum ether).

[+] When our wort was already finished, Brieskorn and Wunderer published a short note with their results on the triterpenic acids from the peel of apples (Z. Naturforschg. 21.b, 1005 (1966). They isolated the 19 hydroxiursolic acid and named it pomolic acid. Consequently our name of Benthamic acid must be changed to polomic acid.

105-115° (from MeOH) 127-129°, $[\alpha]_D^{20}$ + 39°, IR 3530 (OH) and 1728 cm^{-1} (methyl ester); acetylation V_b yielded a monoacetate $C_{33}H_{52}O_5$ V_c, m.p. 235-238°, $[\alpha]_D^{20}$ + 41°, IR 3550 (OH) and 1730 cm^{-1} (methyl ester and acetate), NMR spectrum TABLE 1. The tertiary character of the hydroxyl group, which could not be acetylated, was deduced from its resistance to oxidation. The similarity of the NMR and IR spectra of this compound and the ursane and oleane suggested a pentacyclic skeleton.

Dehydration of V_c with $SOCl_2$ in pyridine gave a mixture of conjugated and nonconjugated dienes from which were isolated by fractionated crystallization, vanguerolic and tomentosolic acid acetate methyl ester VI and VII (3), both identified by physical constants and a comparison with original samples (mixed m.p. IR identical).

Treatment of V_a (m.p. 272-275°, $[\alpha]_D^{20}$ 0°) with a mixture of HCl and acetic acid hat yielded the acetate of a lactone, VIII, which proved to be identical with the lactone of Merz and Tschubel (4) (mixed m.p.; IR superimposable). The formation of these compounds shows that V_a is a hydroxyl derivative of ursolic acid with the OH group at C_{19} or C_{20}. The preparation, in good yield, of VI and the fact that V_b is not identical with the methyl ester of 20β-OH ursolic acid (5) suggests position 19 for the tertiary alcoholic group.

The NMR spectrum of V_c shows a signal at τ 8.75 due to the methyl on the C-atom which bears the OH. The signal at τ 8.78

attributed to the Me-27, is paramagnetically displaced by 0.15 ppm relative to the Me-27 of the methyl acetyl ursolate (8.93). This shift is explained by the close proximity of the Me-27 and the OH on C_{19} in position α (6).

A relatively sharp signal in the region of the allylic protons, at τ 7.37, is assigned to the H_{18}. Its shape indicates that there are no hydrogen atoms in the inmediate neighbourhood and that C_{13} and C_{19} are therefore completily substituted, which is in accordance with the structure of 19α-OH ursolic acid proposed for V_a.

The C_{20}-Me in V probably is α orientated for the following reasons: The formation from V_c of VII, with the normal 18-βH orientation, suggests that benthamic acid has the same stereochemistry at C_{18}. If the dehydration of V_c with $SOCl_2$ proceeds via <u>trans</u> diaxial elimination, the C_{19}-OH will have the α and the C_{20}-H β orientation as in ursolic acid.

Vanguerolic acid VI, derived from V_c should have the same stereochemistry as benthamic and ursolic acids at C_{20} which is opposite to the that reported by Barton and seems to be well documented (3). This question should therefore be revised.

V_a as well as 3β-OH ursa$\Delta^{12,20(30)}$-dien-28-oic acid I ($R=R_1=H$) have the normal configuration 18β-H. Their biogenesis probably is achieved in a normal way. The acid V_a may be at common precursor to VI and VII. By OH elimination, the cation

IX may be formed instead of X proposed by Barton and co-workers (7). On the other hand, X on losing an H_{30} would form the acid I ($R=R_1=H$).

The analyses of all the compounds mentioned were satisfactory. If not otherwise stated, the IR spectra as well as the optical activities were determined in $CHCl_3$, the NMR spectra in $CDCl_3$ with TMS as internal standard, on a PE R-10 model (60 Mc).

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REFERENCES

1. A.S.Gupta & S.Deo, J. Chromatog. **12**, 139 (1963).
2. B.Tursch, R.Savoir, R.Ottinger & G.Chiurdoglu, Tetrahedron Letters, **1**, 205 (1966).
3. D.R.H.Barton, H.T.Cheung, P.J.L. Daniels, K.G.Lewis & J.F. McGhie, J. Chem. Soc. **1962**, 5163.
4. K.W.Merz & H.Tschubel, Ber. **72**, 1017 (1939).
5. W.Lawrie, J.McLean & M.E.Garby Younes, Chem. & Ind. **1966** 1720.
6. P.Potier, B.C.Das, A.M.Bui, M.M.Janot, A.Pourrat & H.Pourrat Bull. Soc. Chim. (France), **1966**, 3458.
7. D.R.H.Barton, loc. cit.