## 858. *Triterpenoids from New Zealand Plants. Isolation of Ursolic Acid from* Gaultheria Subcorymbosa *Col.*

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The recent extensive studies of Djerassi and his co-workers [1,2] have clearly demonstrated the great potentialities of mass spectrometry as a method for the rapid characterisation of triterpenoids. We now wish to report its application to the identification of a triterpene acid obtained by ethanolic extraction of the twigs and leaves of the New Zealand native shrub *Gaultheria subcorymbosa* (family Ericaceae). This acid, like its derived methyl ester, showed widely differing melting points on crystallisation from different solvents, but application of mass spectrometry to the different methyl ester fractions not only showed that all the spectra were essentially the same (within the normal variations to be expected from factors such as temperature differences), thus indicating that different solvations and/or different crystalline forms were responsible for the observed melting point differences, but identified the ester as methyl ursolate through comparison with the published spectrum.[1] The identity of the acid was then further confirmed by direct comparisons between its methyl ester and authentic methyl ursolate, between the acetyl methyl ester and authentic methyl *O*-acetylursolate and between the diol produced on lithium aluminium hydride reduction of the methyl ester and authentic uvaol.

*Experimental.*—M.p.s were taken on a Kofler block; $[\alpha]_D$'s were taken in $CHCl_3$. Infrared spectra were measured in $CCl_4$ solution unless otherwise stated. Light petroleum refers to the fraction of b. p. 60—80°. The mass spectra were determined with an A.E.I. M.S.9 double-focusing mass spectrometer using a direct inlet system. The energy of ionising electrons was 70 v, the ionising current was 100 μA and the source temperature was 90—110°.

*Isolation of ursolic acid.* Dried finely-ground twigs and leaves of *Gaultheria subcorymbosa* (800 g.) were exhaustively extracted with boiling ethanol (750 ml.) in a Soxhlet apparatus (24 hr.). Removal of solvent afforded 28 g. of total extractives which on extraction with cold light petroleum left a solid residue of ursolic acid (3·1 g.) showing m. p. 224—244° from ether, m. p. 260—280° from ethanol, and m. p. 280—281° from ether–light petroleum (lit.,[3] for ursolic acid, m. p. 279°). The analysis of the alkane fraction isolated from the petroleum ether-soluble material has been reported elsewhere.[4]

*Methyl ursolate.* Treatment of the acid isolated as described above with an excess of ethereal diazomethane afforded the methyl ester showing m. p. 112—114° from ether, m. p. 162—164° from ethyl acetate, m. p. 169—171° from ethanol, $[\alpha]_D = +58$ (*c* 2·0); lit.[5] for methyl ursolate, m. p. 169—170°, $[\alpha]_D = +62$. The m. p. of the specimen crystallised from ethanol was undepressed on admixture with authentic material. The infrared spectra were identical. Mass spectrum: parent molecular peak 470, corresponding to $C_{31}H_{50}O_3$; m/e 411, 410, 262 (intense), 249, 207, 203 (intense), 189, 133 with metastable peaks at 157·3, 87·2, and 172·8 corresponding to the transitions $262^+ \longrightarrow 203^+$, $203^+ \longrightarrow 133^+$ and $207^+ \longrightarrow 189^+$, respectively—comparable with that of authentic methyl ursolate.[1]

*Methyl O-acetylursolate.* Methyl ester (1 g.) prepared from the acid isolated from *Gaultheria subcorymbosa* was acetylated by adaptation of the method of Sengupta and Khastgir [6] by heating on the steam bath with acetic anhydride (10 ml.) and pyridine (10 ml.) for 4 hr. The crystalline product (1·05 g.) obtained after cooling the reaction mixture and pouring into water was separated and crystallised from ethanol; it then had m. p. 229—232°, $[\alpha]_D = +52$ (*c* 2·0); lit.,[7] m. p. 244—247°, $[\alpha]_D = +58$. There was no m. p. depression with authentic material and the infrared spectra were correct.

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*Uvaol.* Methyl ester (200 mg.) prepared from the acid isolated from *Gaultheria subcorymbosa* was treated in refluxing dry ether with an excess of lithium aluminium hydride (1·0 g.) for 8 hr. Excess of reagent was destroyed by careful addition of water, and the mixture was treated with 6N-HCl. Extraction with ether afforded the diol, m. p. 223—225° (from ether), $[\alpha]_D = +76$ (*c* 2·0); lit.,[8] for uvaol, m. p. 222—224°, $[\alpha]_D = +72$. There was no mixed m. p. depression and the infrared spectra in chloroform were correct. Mass spectrum: parent molecular peak 442, corresponding to $C_{30}H_{50}O_2$; m/e 411, 234 (intense), 221, 207, 203 (intense), 189, 133 with metastable peaks at 176·2, 87·0, and 172·6 corresponding to the transitions $234^+ \longrightarrow 203^+$, $203^+ \longrightarrow 133^+$, and $207^+ \longrightarrow 189^+$, respectively.

We thank Dr. R. I. Reed for his kind interest. We also thank Dr. A. T. Johns, Plant Chemistry Division, Grasslands Research, D.S.I.R., Palmerston North, New Zealand, for the use of drying facilities and Dr. W. Lawrie for kindly supplying authentic specimens of methyl ursolate, methyl *O*-acetylursolate and uvaol. M. M-S. gratefully acknowledges generous financial support from the Royal Society and the Pharmaceutical Society of Great Britain, T. A. B. a D.S.I.R. research studentship, and E.C. an Institute of Petroleum grant.

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