Triterpenoids. Part XVI. The Constitution of Rehmannic Acid.*

By D. H. R. Barton and P. de Mayo.

[Reprint Order No. 4799.]

The triterpenoid, rehmannic acid, isolated from the roots of *Lippia rehmanni* Pears, has been shown by degradation experiments parallel to those applied to icterogenin to be 22β-angeloyloxyoleanonic acid (deoxyicterogenin).

In Part XV * the isolation of a new triterpenoid acid, rehmannic acid, $C_{35}H_{52}O_5$, was briefly reported. As already mentioned this compound, like icterogenin, possesses a functionally reactive ketone grouping. The ultra-violet spectrum of rehmannic acid was similar to that of icterogenin and thus suggested that the remaining two (uncharacterised) oxygen atoms were part of an αβ-unsaturated ester residue. An even closer relationship to icterogenin was indicated by identical Zimmermann and tetranitromethane colours. On the basis of these findings and of further evidence outlined in the sequel the constitution (I; R = H) was deduced for rehmannic acid.

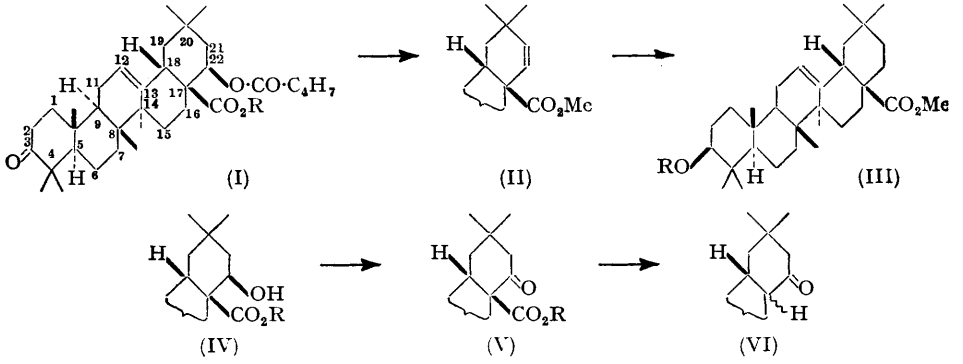
Pyrolysis of methyl rehmannate (I; R = Me) furnished angelic acid (thus confirming the presence of an αβ-unsaturated ester function as well as accounting for five of the carbon atoms of the parent compound) and an olefin (II). On hydrogenation, the latter afforded methyl oleanolate (III; R = H), further characterised as the acetate and the benzoate (III; R = Ac and Bz, respectively). All 35 carbon atoms of rehmannic acid are thus accounted for.

Vigorous alkaline hydrolysis of rehmannic acid furnished the corresponding hydroxy-acid (IV; R = H), chromic acid oxidation of which gave a β-keto-acid (V; R = H). The latter was not isolated but was characterised by its ready decarboxylation in warm benzene solution to a crystalline diketone, $C_{29}H_{44}O_2$ (VI), m. p. 237—240°, $[\alpha]_D$ +49°.

As with icterogenin, the placing of the ester side-chain at $C_{(22)}$ rather than at $C_{(16)}$ is based on exclusion evidence. Both the $C_{(17)}$ stereoisomeric nor-diketones, (VII) and

* Part XV, preceding paper.

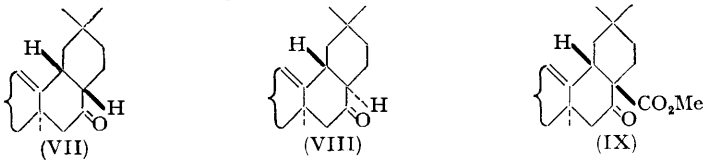
(VIII), have been obtained from echinocystic acid (White and Noller, *J. Amer. Chem. Soc.*, 1939, **61**, 983). These have m. p. 210—212°, $[\alpha]_D$ −93° (dioxan), and m. p. 230—233°, $[\alpha]_D$ +86°, and are clearly different from (VI). Indeed the diketone from rehmannic acid gave a m. p. depression on admixture with the latter compound. In agreement, conversion of (IV; R = H) into its methyl ester (IV; R = Me) and oxidation gave a diketo-methyl ester (V; R = Me), m. p. 197—199°, $[\alpha]_D$ +37°, different from the analogous echinocystic



(I) (II) (III) (IV) (V) (VI)

acid derivative (IX), of m. p. 166—168°, $[\alpha]_D$ +2° (dioxan) (White and Noller, *loc. cit.*) (see also Experimental).

The assignment of the β-configuration for the side-chain at $C_{(22)}$ is based in the main part on molecular-rotation considerations. Thus, it can be shown that the 22β-angeloyloxy-grouping of methyl icterogenin (see Part XV, *loc. cit.*) contributes +50° in $[M]_D$. Addition of this to the $[M]_D$ of methyl oleanonate (+417°; see Barton and Jones, *J.*, 1944, 659) gives a calculated $[M]_D$ of +467°, or $[\alpha]_D$ +83°, for the structure (I; R = Me). This is in close agreement with the observed $[\alpha]_D$ of +86° for methyl rehmannate. Further the conversion of methyl rehmannate into methyl dihydro-rehmannate causes an $[M]_D$ change of −26°, clearly comparable with that, −37°, observed



(VII) (VIII) (IX)

in the reduction of methyl 22β-angeloyloxyhedragonate to the α-methyl-*n*-butyroyloxy-derivative. Other values which support this assignment are the increment for the hydrolysis of the angelate residue (−109° for the rehmannate series and −91° in the icterogenin series), and that for the oxidation of the 22β-hydroxyl to the ketone (−199° in the rehmannate series and −193° in the icterogenin series) (see Part XV, *loc. cit.*).

In the course of this work we prepared methyl deoxodihydrorehmannate and methyl deoxodihydroicterogenin benzoate. Details are given in the Experimental Section.

It is not without interest that the ultra-violet absorption spectrum of rehmannic acid ($\lambda_{max.}$ 212 mμ; ε, 12400), like that of icterogenin, corresponds to what would be expected from the superimposition of a β-amyrin-type of ethylenic linkage and of an angelate grouping rather than of a tiglate residue (see Adams and van Duuren, *J. Amer. Chem. Soc.*, 1953, **75**, 4631). Furthermore the absorption spectrum of icterogenin bromolactone (see Part XV, *loc. cit.*) is comparable with that of angelic acid rather than that of tiglic acid. Additional confirmatory evidence is thus forthcoming for the angelate side-chain in these compounds.

Experimental

For general experimental detail see Part VII (*J.*, 1952, 2339). Rotations were determined in chloroform solution. Ultra-violet absorption spectra were determined in ethanol solution, the Unicam S.P. 500 Spectrophotometer being used. M. p.s are uncorrected.

Methyl Dihydrorehmannate.—Rehmannic acid (110 mg.) in ethyl acetate (25 ml.) was hydrogenated for 1 hr., a palladised charcoal (50 mg.; 5%) catalyst being used. Crystallisation from methanol afforded *dihydrorehmannic acid*, m. p. *ca.* 160° (with solvent evolution), $[\alpha]_D$ +88° (*c*, 1·73), $\lambda_{max.}$ 282 mμ (ε, 30) (Found: C, 73·7; H, 9·75. $C_{35}H_{54}O_5,CH_3{\cdot}OH$ requires C, 73·7; H, 9·95%). Treatment with ethereal diazomethane gave *methyl dihydrorehmannate*, solvated needles (from methanol), m. p. *ca.* 125°, $[\alpha]_D$ +81° (*c*, 1·80) (Found: C, 76·15; H, 9·6. $C_{36}H_{56}O_5$ requires C, 76·0; H, 9·9%).

Degradation of Rehmannic Acid.—Rehmannic acid (100 mg.) was refluxed with ethanolic potassium hydroxide (4%; 10 ml.) for 6 hr. The product was chromatographed over silica gel (2 g.; 6 fractions). Elution with ether–benzene (1 : 19) (3 fractions) afforded 22β-*hydroxyoleanonic acid*. Recrystallised from aqueous methanol (fine needles) this had m. p. 233—236°, $[\alpha]_D$ +92° (*c*, 0·78) (Found: C, 74·1; H, 9·45. $C_{30}H_{46}O_4,CH_3{\cdot}OH$ requires C, 74·05; H, 10·05%).

The hydroxy-acid (101 mg.) in "AnalaR" acetic acid (3 ml.) was treated with chromium trioxide (15·5 mg.) in "AnalaR" acetic acid (1 ml.) at room temperature overnight. The product was separated into an acid and a neutral fraction. The latter (very small) fraction was discarded. The acidic fraction was decarboxylated by heating it in benzene solution (10 ml.) on the steam-bath for 15 min. Separation again into an acidic and a neutral fraction and filtration of the latter in benzene solution through alumina (1·0 g.) gave the *diketone*. Recrystallised from methanol as plates, this had m. p. 237—240°, $[\alpha]_D$ +49° (*c*, 1·06) (Found: C, 82·1; H, 10·55. $C_{29}H_{44}O_2$ requires C, 82·0; H, 10·45%). The m. p. was depressed to 210—217° on admixture of our sample with an authentic specimen of *iso*norechinocystenedione (White and Noller, *J. Amer. Chem. Soc.*, 1939, **61**, 983).

22β-Hydroxyoleanonic acid (see above) (35 mg.), with ethereal diazomethane, afforded the corresponding *methyl ester*. Recrystallised from methanol, this had m. p. 183—185°, $[\alpha]_D$ +78° (*c*, 1·02) (Found: C, 77·0; H, 10·3. $C_{31}H_{48}O_4$ requires C, 76·8; H, 10·0%).

Methyl 22β-hydroxyoleanonate (45 mg.) in "AnalaR" acetic acid (2 ml.) was treated with chromium trioxide (7 mg.) in "AnalaR" acetic acid (1 ml.) at room temperature overnight, giving *methyl 22-oxo-oleanonate*. Recrystallised from aqueous methanol, this had m. p. 197—199°, $[\alpha]_D$ +37° (*c*, 1·03); the m. p. was depressed to 145—153° on admixture of our sample with an authentic specimen of methyl 16-oxo-oleanonate (m. p. 169°) from echinocystic acid (White and Noller, *loc. cit.*) (Found: C, 76·9; H, 9·2. $C_{31}H_{46}O_4$ requires C, 77·15; H, 9·6%).

Pyrolysis of Methyl Rehmannate.—The methyl ester (280 mg.) was pyrolysed at 530°/1 mm. in three batches under the general conditions specified in Part XV (*loc. cit.*). The pyrolysate was chromatographed over alumina (11 g.); (8 fractions). Elution with benzene–light petroleum (b. p. 40—60°) (3 : 7) (2 fractions) furnished *methyl olean-21-enonate*. Recrystallised (with difficulty) from a small volume of methanol, this had m. p. 117—119°, $[\alpha]_D$ +19° (*c*, 1·28) (Found: C, 79·2; H, 9·6. $C_{31}H_{46}O_3,\frac{1}{2}CH_3{\cdot}OH$ requires C, 78·6; H, 9·85%). Elution with benzene–light petroleum (b. p. 40—60°) (3 : 2) (3 fractions) gave unchanged methyl rehmannate (m. p. and mixed m. p.).

Methyl olean-21-enonate (12 mg.) in "AnalaR" acetic acid (5 ml.) was hydrogenated at room temperature overnight, a platinum catalyst (50 mg.) being used. The catalyst was removed by filtration and the acetic acid by evaporation *in vacuo*. The residue was treated with methanolic potassium hydroxide (5%; 5 ml.) under reflux for 20 min. and the neutral product was crystallised from aqueous methanol, giving methyl oleanolate, m. p. 198—200°, $[\alpha]_D$ +70° (*c*, 0·7), undepressed in m. p. on admixture with an authentic specimen. Acetylation with pyridine–acetic anhydride at room temperature overnight gave methyl oleanolate acetate (m. p. and mixed m. p.).

In a second experiment the identity of the oleanolate was further confirmed by conversion into the benzoate (pyridine–benzoyl chloride at room temperature overnight). This had m. p. 260—261° (Kofler block), $[\alpha]_D$ +82° (*c*, 0·74), m. p. undepressed on admixture with an authentic specimen of the same m. p. and rotation. The acid resulting from the pyrolysis which collected in the cooled receiver was identified by m. p. (42°), mixed m. p., and crystal form as angelic acid (cf. Part XV, *loc. cit.*).

Methyl Deoxodihydrorehmannate.—Methyl dihydrorehmannate (see above) (67 mg.) in benzene (1 ml.) and toluene-ω-thiol (1 ml.) was shaken with perchloric acid (5 drops; 70%) (see Barton and Rosenfelder, *J.*, 1951, 1048) for 15 min. Crystallisation of the product from chloroform–methanol afforded the *dithioketal*, m. p. 183—185° (Found: C, 75·5; H, 9·0. $C_{50}H_{70}O_4S_2$ requires C, 75·15; H, 8·85%). The dithioketal (60 mg.) in purified dioxan (10 ml.) was refluxed with Raney nickel (5 g.) for 2 hr., giving *methyl deoxodihydrorehmannate*, m. p. 166—

168° (from methanol), $[\alpha]_D$ +100° (Found : C, 78·05; H, 10·2. $C_{36}H_{58}O_4$ requires C, 77·95; H, 10·55%).

Methyl Deoxodihydroicterogenin Benzoate.—Methyl dihydroicterogenin benzoate (270 mg.) in benzene (10 ml.) and toluene-ω-thiol (2 ml.) was shaken with perchloric acid (1 ml.; 70%) for 15 min. Crystallisation from chloroform–methanol gave the dithioketal (150 mg.), m. p. 168—170°. This was dissolved in dioxan (10 ml.) and refluxed with Raney nickel (5 g.) for 2 hr., giving *methyl deoxodihydroicterogenin benzoate*, fine needles (from chloroform–methanol), m. p. 192—193·5°, $[\alpha]_D$ +113° (*c*, 1·96), $\lambda_{max.}$ 229 mμ (ε, 15,500) (Found : C, 76·55; H, 9·0. $C_{43}H_{62}O_6$ requires C, 76·5; H, 9·25%).

We thank the Government Grants Committee of the Royal Society, the Central Research Fund of London University, and Imperial Chemical Industries Limited for financial assistance. One of us (P. de M.) is grateful to the Colonial Products Research Council for a Research Studentship.

BIRKBECK COLLEGE, LONDON, W.C.1. [*Received, November 14th,* 1953.]