

277. *Constituents of the Higher Fungi. Part II. The Unsaturated System of Polyporenic Acid A.*

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Dihydropolyporenic acid A has been prepared by partial catalytic hydrogenation and its *methyl ester* and *methyl ester-monoacetate* have been obtained. Ozonolysis of the methyl ester-acetate of polyporenic acid A gives a 50% yield of formaldehyde, and, since none is obtained on ozonolysis of the corresponding dihydro-compound, the reactive double bond of acid A must be present in an exocyclic methylene group.

In Part I (this vol., p. 632) it was demonstrated that polyporenic acid A, a constituent of the fungus *Polyporus betulinus* Fr., is a diethenoid dihydroxy-acid of probable formula $C_{30}H_{48}O_4$, its diethenoid nature being established by quantitative microhydrogenation and by titration with perbenzoic acid. We have now examined in greater detail the hydrogenation of acid A and find that in acetic acid solution in the presence of platinum-black at atmospheric pressure and temperature one mole of hydrogen is rapidly absorbed, after which the rate of hydrogenation diminishes abruptly. The product of this partial reduction, *dihydropolyporenic acid A*, $C_{30}H_{50}O_4$, has been characterised by the preparation of the *methyl ester* and the *methyl ester-monoacetate*.

The marked difference in reactivity between the two double bonds of polyporenic acid A suggested that the readily reducible bond might be located in a side chain and consequently the methyl ester-acetate was ozonised, a 50% yield of formaldehyde being obtained. In order to confirm that it is the readily reducible double bond of polyporenic acid A which is present in an exocyclic methylene group it has been established that no more than a 4% yield of formaldehyde is obtained on ozonolysis of the methyl ester-acetate of dihydro-acid A under similar conditions. It is now evident that polyporenic acid A may be closely related to those triterpenes known to possess an exocyclic ethenoid linkage, viz., *basseol* (Beynon, Heilbron, and Spring, J., 1937, 989), *lupeol* and *betulin* (Jones and Meakins, this vol., p. 456). The facility with which the one ethenoid linkage in acid A can be hydrogenated, however, is more reminiscent of the behaviour of *lupeol* than of *basseol*, the reduction of the latter being difficult to accomplish. Further, polyporenic acid A is rather more stable to cyclising agents than was at first thought and treatment of the methyl ester with formic acid in the cold, effective in cyclising *basseol acetate* into β -*amyrin acetate*, merely results in the formation of the *methyl ester-formate* of polyporenic acid A, which regenerates the acid A on hydrolysis with alcoholic alkali.

The neutral portion of the non-volatile residue from the ozonolysis of the methyl ester-acetate of polyporenic acid A yielded a small quantity of a crystalline *methyl ester keto-*

acetate (?), $C_{32}H_{50}O_6$. Quantitative microhydrogenation indicates the presence of an ethenoid linkage or carbonyl group in the compound and the absorption spectrum, although somewhat ambiguous, also suggests the presence of a carbonyl group. The ozonolysis of the dihydromethyl ester-acetate also yielded a neutral substance, m. p. 189° , probably formed by the action of ozone on the second ethenoid linkage of polyporenic acid.

EXPERIMENTAL.

All m. p.'s are uncorrected. Analytical specimens were dried in a high vacuum (10^{-3} mm.) for several hours at temperatures about 30° below the m. p.

Dihydropolyporenic Acid A.—When polyporenic acid A (7 g.) in acetic acid (200 c.c.) was hydrogenated at 20° under atmospheric pressure in the presence of platinum oxide (1.1 g.), a rapid absorption of hydrogen ensued until one mole had been taken up. The catalyst was removed, and careful dilution of the hot filtrate with water gave the *dihydro-acid A*, which crystallised from aqueous acetic acid in fine needles (6 g.), m. p. 216° . The acid is readily soluble in alcohol, acetone, acetic acid and pyridine but sparingly soluble in benzene. With chloroform and sulphuric acid, it slowly gives a bright red coloration, with the Liebermann-Burchard reagent it gives a red colour without fluorescence, and with tetranitromethane in chloroform solution it gives a pale yellow coloration. $[\alpha]_D^{20} + 66^\circ$ ($l = 1, c = 1.1$ in pyridine) (Found: C, 76.15; H, 10.9. $C_{30}H_{50}O_4$ requires C, 75.9; H, 10.6%). The *methyl ester* was prepared in the usual manner with diazomethane and after several crystallisations from aqueous methyl alcohol had the constant m. p. 142° , mixed with the methyl ester of acid A, m. p. 132° . $[\alpha]_D^{20} + 76^\circ$ ($l = 1, c = 1.0$ in chloroform) (Found: C, 76.5; H, 11.0. $C_{31}H_{52}O_4$ requires C, 76.15; H, 10.7%). The *methyl ester-acetate* was obtained in only poor yield by treatment of the ester with pyridine and acetic anhydride at 100° . With the same reagents at 20° for 24 hours, however, good yields of the acetate, which separated from aqueous methyl alcohol in fine needles, m. p. 142° (softening at 138°), were obtained; $[\alpha]_D^{20} + 36^\circ$ ($l = 1, c = 1.1$ in chloroform) (Found: C, 74.65; H, 10.0. $C_{33}H_{54}O_5$ requires C, 74.7; H, 10.3%).

Ozonolysis of the Methyl Ester-Acetate of Polyporenic Acid A.—A slow stream of ozonised oxygen was passed into a solution of the methyl ester-acetate (830 mg.) in purified acetic acid (20 c.c.) at 20° for 2 hours, the issuing gases being led into water. The acid and water solutions were combined and distilled in steam until the distillate no longer gave a turbidity with 2:4-dinitrophenylhydrazine hydrochloride. The whole distillate was then treated with this reagent; the dried precipitate (162 mg., corresponding to a 50% yield of formaldehyde) crystallised from alcohol in fine needles, m. p. 159° , undepressed by an authentic specimen of the formaldehyde derivative. In a second experiment, with 1 g. of the methyl ester-acetate, the formaldehyde was estimated as the dimedon derivative, 265 mg. being obtained, equivalent to a 45% yield; after crystallisation from alcohol, it had m. p. 187° , undepressed by an authentic specimen.

The non-volatile portions of both experiments were combined and extracted with ether, and the ethereal solution washed with 2*N*-sodium carbonate, dried, and evaporated. The residual gum crystallised on contact with methyl alcohol, giving the *methyl ester keto-acetate* (?) (250 mg.) in needles which after several recrystallisations had m. p. 194° , $[\alpha]_D^{20} + 121^\circ$ ($l = 1, c = 0.75$ in chloroform) (Found: C, 72.4, 72.4; H, 9.6, 9.6. $C_{32}H_{50}O_6$ requires C, 72.45; H, 9.5%). *Quantitative microhydrogenation*: The compound, m. p. 194° (11.74 mg.), absorbed 0.540 c.c. of hydrogen at 18° and 752 mm., corresponding to one double bond. The ozonolysis product reacted with both hydroxylamine and 2:4-dinitrophenylhydrazine, but in neither case could a homogeneous product be isolated. *Light absorption in chloroform-alcohol*: Maximum 2500 μ , inflexions 2720 and 3160 μ ; $E_{1\%}^{1\text{cm}}$ = 71, 15, and 1.4 respectively.

Ozonolysis of the Methyl Ester-Acetate of Dihydro-acid A.—The methyl ester-acetate (950 mg.) was ozonised exactly as described above and a yield of formaldehyde (estimated as the dimedon derivative) of less than 4% was obtained. The neutral gum isolated from the non-volatile portion by means of ether yielded crystalline material (150 mg.) which after three crystallisations from aqueous methyl alcohol had the constant m. p. 189° (Found: C, 74.6; H, 9.8%). *Light absorption in alcohol*: Maxima, 2500 and 3200 μ , $E_{1\%}^{1\text{cm}}$ = 83 and 70 respectively.

Methyl Ester-Formate of Acid A.—A solution of the methyl ester (500 mg.) in formic acid (50 c.c.; 90%) was set aside at 20° for 40 hours and then diluted with water. The neutral product, isolated with ether, crystallised in contact with methyl alcohol. Several crystallisations from this solvent gave the *methyl ester-formate* (180 mg.) in plates, m. p. 148° , $[\alpha]_D^{20} + 84^\circ$ ($l = 1, c = 1.0$ in chloroform) (Found: C, 73.5, 73.6; H, 9.6, 9.3. $C_{32}H_{50}O_5 \cdot \frac{1}{2}CH_3 \cdot OH$ requires C, 73.5; H, 9.8%). The formate (100 mg.) was refluxed for 5 hours with methyl-alcoholic

potassium hydroxide (20 c.c., 10%); the acid portion yielded polyporenic acid A, m. p. 192°, undepressed by an authentic specimen.

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