

93. *The Chemistry of the Triterpenes. Part XIV.\* Further Evidence concerning the Unsaturated Centres of Polyporenic Acid A.*

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Polyporenic acid A contains two double bonds one of which readily undergoes catalytic hydrogenation yielding two non-interconvertible series of dihydro-derivatives. The readily reducible double bond is present in a vinylidene group and the inert double bond is most probably of the tetra-substituted type. In contrast to the parent acid the stereoisomeric dihydro-acids do not decarboxylate, indicating that the reactive double bond is  $\beta\gamma$  to the carboxyl group in polyporenic acid A.

In the original investigation of polyporenic acid A (Cross, Eliot, Heilbron, and Jones, *J.*, 1940, 632), the presence of two double bonds was indicated by a microhydrogenation and by per-acid titrations, confirmed in the preceding paper. Although the acid reacted with 2 mols. of hydrogen under the vigorous conditions of a microhydrogenation at 95°, hydrogenation at 20° resulted in the uptake of only 1 mol. of hydrogen (Cross and Jones, *J.*, 1940, 1491). This procedure resulted in the isolation and characterisation of what was believed to be the dihydro-derivative of polyporenic acid A. Comparative ozonolysis experiments on a derivative of the latter and on the parent compound, from which yields of formaldehyde of 4% and 50% respectively were obtained, indicated that the readily reducible double bond was present as a vinylidene group.

The hydrogenation of acid A has now been studied in greater detail and chromatographic examination of the methyl esters and methyl ester acetates has revealed that the process is more complicated than was originally believed. The product of hydrogenation of the acid itself had the same physical constants as described by Cross and Jones (*loc. cit.*), although it proved to be dimorphous. Esterification of this acid or hydrogenation of methyl polyporenic acid A, gave, after repeated crystallisation, an ester with the properties described earlier. By careful chromatography this material yielded two isomeric compounds, m. p.s 159—160° and 132—133°, and more of the latter was isolated by chromatographic purification of the material contained in the initial mother liquors. Further experiments involving the catalytic hydrogenation of the methyl ester *a*-acetate and *a* : *b*-diacetate, and acetylation of the mixed methyl dihydro-esters also indicated the existence of two methyl dihydro-esters in both the monoacetate and the diacetate series. The results of these initial experiments were somewhat unpredictable and difficult to repeat. However, the possibility of polymorphism was ruled out in all cases by the separation of one and the same compound even when its solutions were seeded with the other isomer.

Further study of these reactions with careful chromatographic purification of the products conclusively established the existence of two non-interconvertible series of dihydro-derivatives. These have been arbitrarily designated (I) and (II), compounds in series

\* Part XIII, preceding paper.

(I) exhibiting higher melting points and lower rotations than their isomers in series (II). It has not been possible to separate the mixture of parent dihydro-acids, but the mixed isomers exist in two polymorphic forms.

The infra-red absorption spectra of methyl dihydro-(I)-polyporeneate A *a* : *b*-diacetate (m. p. 137.5—139°,  $[\alpha]_D + 80^\circ$ ) and methyl dihydro-(II)-polyporeneate A *a* : *b*-diacetate (m. p. 116.5—117.5°,  $[\alpha]_D + 87^\circ$ ) were compared. The two compounds displayed identical absorption curves in carbon disulphide solutions, but there were significant minor differences when the compounds were examined in the crystalline state (Nujol suspension). The identical solution spectra obviously preclude structural isomerism and any but minor stereochemical differences. The latter become apparent in the divergent spectra of the crystalline samples; the characteristic ester group absorptions at 1000—1050  $\text{cm}^{-1}$  and 1150—1200  $\text{cm}^{-1}$  are those revealing the greatest differences. This may be due to the close spacial proximity between the methyl group produced in the hydrogenation and the carboxylic ester group (see below).

The second double bond in polyporenic acid A is inert towards catalytic hydrogenation under normal conditions and is almost certainly cyclic. It does not migrate in the presence of hydrogen and a catalyst in an acidic medium (cf. 'α'-spinasterol; Fieser, Fieser, and Chakravarti, *J. Amer. Chem. Soc.*, 1949, **71**, 2226), since the same product was isolated from the hydrogenation of acid A, by employing a platinum catalyst, in both acidic and neutral media.

The infra-red spectra of a number of derivatives of polyporenic acid A have been examined. In solution (chloroform) and in the solid state (Nujol suspension), methyl polyporeneate A and *a* : *b*-diketopolyporenic acid A display prominent bands at 902 and 1648  $\text{cm}^{-1}$  which are absent in the epimeric methyl dihydro-ester diacetates as well as in the decarboxy-*a* : *b*-diketo-compound (see preceding paper), and are attributed to the vinylidene group,  $\text{CH}_2=\text{C}<$ . This group clearly disappears when *a* : *b*-diketopolyporenic acid A is decarboxylated to the decarboxy-*a* : *b*-diketo-compound which shows no bands at 902 and 1648  $\text{cm}^{-1}$ . Methyl polyporeneate A and its derivatives display no marked absorption in the 650—850  $\text{cm}^{-1}$  range, and the presence of cyclic  $-\text{CH}=\text{CH}-$  or  $-\text{CH}=\text{C}<$  groups is therefore highly improbable. The second double bond would appear to be of the tetrasubstituted type.

Oxidation of the *b*-hydroxyl group in methyl polyporeneate A *a*-acetate gives rise to a carbonyl group which does not react with the usual carbonyl group reagents (previous paper), but for which evidence is obtained by infra-red examination. Thus, besides the carboxyl frequency at 1736  $\text{cm}^{-1}$ , crystalline *a* : *b*-diketopolyporenic acid A gives rise to two strong ketonic carbonyl bands at 1710 and 1692  $\text{cm}^{-1}$  which, in solution, appear as a single strong band at 1713  $\text{cm}^{-1}$  in both the *a* : *b*-diketo-acid and its decarboxy-derivative. Further, the ester groups in methyl polyporeneate A *a*-acetate display a single strong band at 1729  $\text{cm}^{-1}$  which is also present in methyl *b*-ketopolyporeneate A *a*-acetate. However, instead of the hydroxylic absorption of the former at 3500  $\text{cm}^{-1}$  the latter displays a second strong band at 1708  $\text{cm}^{-1}$  which must be attributed to the newly introduced carbonyl group. It may be noted that the values recorded above for the two ketonic carbonyl frequencies exclude the possibility of either group being present in a five-membered ring (cf., Jones, Williams, Whalen, and Dobriner, *J. Amer. Chem. Soc.*, 1948, **70**, 2024).

Further to the observation that polyporenic acid A readily decarboxylates when melted (preceding paper) it has been found that the dihydro-acid is unaffected after being kept in a molten state for some considerable time. The infra-red absorption-spectra bands displayed by polyporenic acid A at 902 and 1648  $\text{cm}^{-1}$ , attributable to the vinylidene group, disappear on both hydrogenation and decarboxylation of the acid. It is clear, therefore, that the close spatial proximity between the unsaturated methylene group and the carboxyl group must be responsible for the decarboxylation, and moreover in the absence of selective absorption characteristic of an  $\alpha\beta$ -unsaturated acid it must be concluded that polyporenic acid A contains the double bond  $\beta\gamma$  to the carboxyl group.

It is well known that  $\beta\gamma$ -unsaturated acids readily decarboxylate when heated and that the elimination of carbon dioxide is accompanied by a shift of the double bond to the former  $\alpha\beta$ -position (cf. Arnold, Elmer, and Dodson, *J. Amer. Chem. Soc.*, 1950, **72**, 4359).

Barton and Brooks (*J.*, 1951, 257) have shown that the triterpenoid morolic acid loses carbon dioxide when melted, producing oleanol in excellent yield. The decarboxylation of polyporenic acid A is analogous to that of morolic acid except that in the former the reacting centres are almost certainly located in a side-chain. The infra-red spectrum of the decarboxy-compound displays a band of medium strength at  $815\text{ cm.}^{-1}$ , not shown by the parent acid, which is indicative of the presence of a trisubstituted double bond (cf. Bladon, Fabian, Henbest, Koch, and Wood, *J.*, 1951, 2402), formed as the result of migration during decarboxylation.

Analysis of all the available analytical data shows unequivocally that polyporenic acid A is tetracyclic. Of 34 analyses of the acid and its derivatives, 22 agree within the usual limits with a  $\text{C}_{29}$ , 30 with a  $\text{C}_{30}$ , and 19 with a  $\text{C}_{31}$  formula for acid A. The evidence is thus somewhat in favour of  $\text{C}_{30}\text{H}_{48}\text{O}_4$ . As indicated in the preceding paper, however, equivalent-weight determinations gave results in good agreement with a  $\text{C}_{31}$  formula (Cross, Eliot, Heilbron, and Jones, *loc. cit.*). Furthermore, an X-ray crystallographic examination of the methyl ester failed to distinguish between the  $\text{C}_{30}$  and  $\text{C}_{31}$  formulæ for the acid. The molecular weight observed ( $496 \pm 8$ ) slightly favours  $\text{C}_{32}\text{H}_{52}\text{O}_4$  ( $M=501$ ) for methyl polyporenic acid A. Attempts to prepare a pure derivative containing sufficient halogen to be of value in establishing the empirical formula of the acid by microanalysis have not yet been successful.

The molecular rotation differences consequent upon hydrogenation of the reactive double bond in polyporenic acid A are shown in the Table. In neither series is there any great change in  $[M]$ . Barton and Jones (*J.*, 1944, 659) have pointed out that the tetracyclic triterpenes containing reactive double bonds form a single group, the members of which show little change in  $[M]$  on hydrogenation. In this respect polyporenic acid A fits into the group.

Compound	[M]			$\Delta_{\text{I}}$	$\Delta_{\text{II}}$
	Parent	Dihydro- (I)-	Dihydro- (II)-		
Polyporenic acid A .....	+303° (pyr.)	+313° (pyr.) *		-12°	+10°
Methyl ester .....	+386	+374°	+379°	-5	+32
Methyl ester <i>a</i> -acetate .....	+175	+170	+207	-39	+1
Methyl ester <i>a</i> : <i>b</i> -diacetate ...	+497	+458	+498		

\* Mixed dihydro-acids.

$\Delta_{\text{I}} = [M]$ , dihydro-(I)- -  $[M]$ , parent.  $\Delta_{\text{II}} = [M]$ , dihydro-(II)- -  $[M]$ , parent.

## EXPERIMENTAL

Rotations were determined in chloroform. M. p.s were determined on a Kofler block and are corrected unless otherwise stated. Analytical specimens were dried at a suitable temperature for 5—10 hours in a high vacuum. Alumina of activity I—II was employed for all chromatograms and except where specified, light petroleum refers to the fraction, b. p. 60—80°.

*Hydrogenation of Polyporenic Acid A.*—Polyporenic acid A (10 g.) in glacial acetic acid (150 c.c.) was hydrogenated at atmospheric temperature and pressure in the presence of Adams's catalyst (50 mg.). The absorption of hydrogen was rapid and ceased when one mol. had been taken up. The product crystallised from aqueous acetic acid as needles (7.4 g.), m. p. 216—218° (Found: C, 75.8; H, 10.55. Calc. for  $\text{C}_{30}\text{H}_{50}\text{O}_4$ : C, 75.9; H, 10.6%). Recrystallisation from methanol-light petroleum (1 : 1) or methanol-chloroform gave fine needles, m. p. 222—224°. Recrystallisation of the latter from aqueous acetic acid again gave the low-melting form; the mixed m. p. of the two forms was 216—220°. Both forms had  $[\alpha]_{\text{D}}^{20} + 66^\circ$  (*c*, 0.4 in pyridine). [Cross and Jones, *loc. cit.*, gave m. p. 216° (capillary uncorr.),  $[\alpha]_{\text{D}}^{20} + 66^\circ$  (*c*, 1.1 in pyridine) for dihydropolyporenic acid A.] It is now considered that this material is a mixture of two stereoisomers and that the mixture is dimorphous, both crystalline forms having the same heterogeneous composition.

*Esterification of the Product from the Hydrogenation of Polyporenic Acid A.*—The above dihydro-product (600 mg.) was esterified with diazomethane. The ester crystallised from aqueous acetic acid as needles (470 mg.), m. p. 140—144°, raised by repeated crystallisation from aqueous methanol to 145—148°,  $[\alpha]_{\text{D}}^{20} + 77^\circ$  (*c*, 1.24) (cf. below).

*Hydrogenation of Methyl Polyporenic Acid A.*—The ester (2.5 g.) in acetic acid (40 c.c.) was hydrogenated at atmospheric temperature and pressure in the presence of Adams's catalyst

(30 mg.). One mol. of hydrogen was rapidly taken up. The product crystallised from aqueous ethanol as rosettes of needles (2 g.), m. p. 132—134°. Repeated crystallisation from aqueous ethanol and aqueous methanol gave needles (700 mg.) with constant m. p., 144—147°,  $[\alpha]_D^{20} + 78^\circ$  (*c*, 1.81). A mixed m. p. with a specimen prepared as above was 144—147°. [Cross and Jones, *loc. cit.*, gave m. p. 142° (capillary uncorr.),  $[\alpha]_D^{20} + 76^\circ$  (*c*, 1.0), for methyl dihydropolyporene A.]

*Hydrolysis of the Product from the Hydrogenation of Methyl Polyporene A.*—The dihydro-ester (100 mg.), m. p. 144—147°, was hydrolysed with methanolic potassium hydroxide (15%) at 20° for 72 hours. The acidic product (70 mg.) was isolated in the usual way, and two crystallisations from methanol gave fine needles of the mixed dihydro-acids, m. p. 222—224°,  $[\alpha]_D^{20} + 65.5^\circ$  (*c*, 1.06 in pyridine).

*Methyl Dihydro-I-polyporene A.*—The product (700 mg.), m. p. 144—147°, from the hydrogenation of acid A methyl ester, was dissolved in a minimum of light petroleum-benzene (1 : 1) and adsorbed on alumina (70 g.). The fraction (60 mg.) eluted with benzene-ether (2 : 1) (100 c.c.) crystallised from nitromethane as prismatic needles, m. p. 132—133°, which were identical with methyl dihydro-II-polyporene A (see below). The fraction (200 mg.) eluted with benzene-ether (1 : 1) (200 c.c.) crystallised from aqueous methanol as a mixture of prisms and needles, m. p. 136—151°. Finally, further elution with benzene-ether (1 : 1) (800 c.c.) gave a fraction (400 mg.) which crystallised from methanol giving *methyl dihydro-I-polyporene A* as stout needles, m. p. 159—160°,  $[\alpha]_D^{20} + 76.5^\circ$  (*c*, 1.19) (Found: C, 75.9; H, 10.7.  $C_{31}H_{52}O_4$  requires C, 76.15; H, 10.75%). A chloroform solution gave a faint yellow colour with tetranitromethane.

*Methyl Dihydro-II-polyporene A.*—Evaporation of the mother liquors from the first three crystallisations of the product from the hydrogenation of acid A methyl ester yielded a gummy solid, m. p. 125—130°, which was exceedingly soluble and crystallised with difficulty. The solid (2 g.) in benzene (30 c.c.) was adsorbed on alumina (160 g.). The fraction eluted with ether was crystallised from nitromethane-methanol giving *methyl dihydro-II-polyporene A* (1 g.) as prismatic needles, m. p. 132—133° undepressed on admixture with the specimen described above,  $[\alpha]_D^{20} + 77.5^\circ$  (*c*, 2.8) (Found: C, 76.15; H, 10.85.  $C_{31}H_{52}O_4$  requires C, 76.15; H, 10.75%). A chloroform solution gave a faint yellow colour with tetranitromethane. Further elution gave a number of gummy fractions (600 mg.) which could not be induced to crystallise. The remaining material (400 mg.) was not eluted with methanol, and may have been hydrolysed.

*Methyl Dihydro-I-polyporene A a-Acetate.*—(a) The product (1.5 g.), m. p. 145—147°, from the hydrogenation of acid A methyl ester, was acetylated in pyridine at 20° in the usual manner. The product crystallised from methanol as needles (760 mg.), m. p. 172—174°; chromatographic purification of these gave *methyl dihydro-I-polyporene A a-acetate* as blunt needles, m. p. 176—178°,  $[\alpha]_D^{20} + 32^\circ$  (*c*, 3.9) (Found: C, 74.6; H, 10.0.  $C_{33}H_{54}O_5$  requires C, 74.7; H, 10.25%).

(b) The dihydro-I-acid methyl ester (100 mg.), acetylated in the same way, gave methyl dihydro-I-polyporene A *a-acetate* (60 mg.) as needles, m. p. 176—178° undepressed on admixture with a specimen prepared as above. A chloroform solution gave a faint yellow colour with tetranitromethane.

*Methyl Dihydro-II-polyporene A a-Acetate.*—The dihydro-II-acid methyl ester (500 mg.) in pyridine was acetylated as above. Crystallisation of the product from aqueous methanol yielded needles of *methyl dihydro-II-polyporene A a-acetate* (400 mg.), m. p. 155—157°, raised by repeated crystallisation from methanol and ethanol to 158.5—159.5°,  $[\alpha]_D^{20} + 39^\circ$  (*c*, 1.15) (Found: C, 75.0; H, 10.2.  $C_{33}H_{54}O_5$  requires C, 74.7; H, 10.25%). A chloroform solution gave a faint yellow colour with tetranitromethane.

*Methyl Dihydro-I-polyporene A a : b-Diacetate.*—(a) The dihydro-(I)-acid methyl ester *a-acetate* (600 mg.) was further acetylated by Whitman and Schwenks's method (*J. Amer. Chem. Soc.*, 1946, 68, 1865). Crystallisation of the product from aqueous methanol gave *methyl dihydro-I-polyporene A a : b-diacetate* (410 mg.) as blunt needles, m. p. 137—139°,  $[\alpha]_D^{20} + 80.5^\circ$  (*c*, 1.80) (Found: C, 73.4; H, 9.65.  $C_{35}H_{56}O_6$  requires C, 73.4; H, 9.85%). A chloroform solution gave a faint yellow colour with tetranitromethane.

(b) The dihydro-I-acid methyl ester (100 mg.) was acetylated as in (a) above, giving methyl dihydro-I-polyporene A *a : b-diacetate* as blunt needles (from aqueous methanol), m. p. 137.5—139° undepressed on admixture with a specimen prepared as above,  $[\alpha]_D^{20} + 80^\circ$  (*c*, 3.3). On titration with perbenzoic acid in chloroform solution methyl dihydro-I-polyporene A *a : b-diacetate* had consumed 1.16 equiv. when the uptake of per-acid was complete.

*Methyl Dihydro-II-polyporene A a : b-Diacetate.*—(a) Methyl polyporene A *a : b-diacetate*

(2 g.) in acetic acid (60 c.c.) was hydrogenated at atmospheric temperature and pressure in the presence of Adams's catalyst (30 mg.) until sufficient hydrogen to saturate one double bond had been taken up. After filtration of the catalyst and removal of the solvent, crystallisation of the residue from aqueous ethanol gave needles (1.6 g.), m. p. 112—114°. This latter material (1 g.) was adsorbed from light petroleum-benzene (1 : 1; 30 c.c.) on alumina (100 g.). Elution with benzene and crystallisation of the eluate from aqueous ethanol gave *methyl dihydro-II-polyporene A a : b-diacetate* (460 mg.) as blunt needles, m. p. 116.5—117.5°,  $[\alpha]_D^{20} + 85^\circ$  (*c*, 1.86) (Found : C, 73.2; H, 9.65.  $C_{35}H_{56}O_6$  requires C, 73.4; H, 9.85%). A chloroform solution gave a faint yellow colour with tetranitromethane. The remaining material (450 mg.) eluted from the column melted over a considerable range.

(*b*) Methyl dihydro-II-polyporene A *a*-acetate (175 mg.), m. p. 158—159°, was further acetylated in the presence of perchloric acid. Crystallisation of the product from aqueous methanol gave methyl dihydro-II-polyporene A *a : b-diacetate* (120 mg.) as elongated prisms, m. p. 114—116°, raised by several crystallisations from aqueous methanol and methanol to 116.5—117.5°, which was undepressed on admixture with a specimen as prepared in (*a*);  $[\alpha]_D^{20} + 89^\circ$  (*c*, 4.24).

(*c*) Methyl dihydro-II-polyporene A (300 mg.), was acetylated as in (*b*). The product isolated in the usual way and crystallised several times from aqueous methanol gave blunt needles of methyl dihydro-II-polyporene A *a : b-diacetate* (200 mg.), m. p. 116—117.5° undepressed on admixture with specimens prepared as in (*a*) and (*b*),  $[\alpha]_D^{20} + 87^\circ$  (*c*, 3.26).

*Attempted Decarboxylation of the Product from the Hydrogenation of Polyporenic Acid A.*—Mixed dihydro-acids A (256 mg.), m. p. 216—218°, dried for 6 hours at 130°/0.05 mm., were heated for 45 minutes at 225—230° in an inert atmosphere. No carbon dioxide or water was eliminated. The residue crystallised from aqueous acetic acid as fine needles (200 mg.; 78% recovery), m. p. 214—216°, undepressed on admixture with the starting material.

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