15. *Triterpene Constituents of the Fruits of the Osage Orange* (Maclura pomifera).

By K. G. Lewis.

The fruits of the osage orange have yielded butyrospermol and a new triterpene diol (probably lupane-3 : 20-diol) in addition to the lupeol reported previously.

Lupeol was reported by Swift and Walter [1] and Beal and Wenzel [2] to be the only triterpene in the fruits of the osage orange (*Maclura pomifera*), but Wagner and Harris [3] later isolated a new substance which they called lurenol. The acetate of this substance was isolated from the crude material by acetylation and fractional crystallisation whereby lupenyl and lurenyl acetate were separated. On the basis of the properties of lurenol and its derivatives Wagner and Harris claimed that it was a new triterpene alcohol. They found [4] however, that when lurenol, obtained by hydrolysis of what they regarded as pure lurenyl acetate, was treated with benzoyl chloride and pyridine it yielded a product which was separable by fractional crystallisation into lupenyl benzoate and lurenyl benzoate. Further, when lurenol was re-acetylated by refluxing acetic anhydride and sodium acetate, lupenyl acetate and lurenyl acetate could be separated. Wagner and Harris claimed that lurenyl acetate was unaffected by the acetylating conditions used and they suggested that lurenol was related in structure to lupeol and that it could be converted into lupeol under acidic or basic conditions.

The crude material isolated by Wagner and Harris's procedure [3] was re-investigated in these laboratories, osage orange fruits * collected in the University grounds being used. The crude non-saponifiable material constituted about 5% of the dried fruit. Partial purification was effected by chromatography over alumina. The crystalline fractions eluted by light petroleum–benzene (fraction A) gave with the Liebermann–Burchard reagent a yellow colour, becoming brown with a strong green fluorescence, behaviour described by Wagner and Harris [5] as characteristic of lurenol. Further crystalline material (fraction B) was eluted by benzene–ether and this on purification afforded a product which gave a pale pink colour in the Liebermann–Burchard reaction and a negative test with tetranitromethane.

Fraction A was acetylated, affording lupenyl acetate and a more-soluble fraction, m. p. 144·5—147°. Hydrolysis of the latter acetate yielded the alcohol which was converted into the benzoate. The properties of these derivatives together with those recorded by

* Identified by the Botany Department of this University.

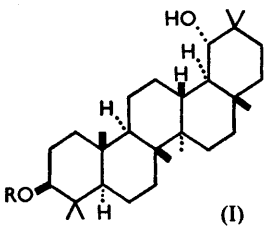
[1] Swift and Walter, *J. Amer. Chem. Soc.*, 1942, **64**, 2539.
[2] Beal and Wenzel, *Trans. Kansas Acad. Sci.*, 1951, **54**, 94 (*Chem. Abs.*, 1951, **45**, 5950).
[3] Wagner and Harris, *J. Amer. Pharm. Assoc.* (*Sci. edn.*), 1952, **41**, 494.
[4] *Idem, ibid.*, p. 497.
[5] *Idem, ibid.*, p. 500.

Wagner and Harris [3] for lurenol and by Heilbron, Jones, and Robins [6] for butyrospermol are compared in the Table.

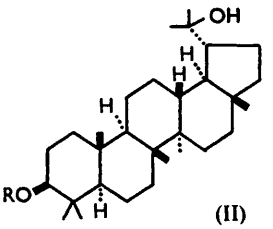
	Wagner & Harris		This work		Heilbron *et al.*	
	M. p.	$[\alpha]_D$	M. p.	$[\alpha]_D$	M. p.	$[\alpha]_D$
Alcohol	166—168°	+15·8°	111—112°	−13°	111—113°	−12°
Acetate	132—133·5	+19·9	144·5—147	+13	146·5—147·5	+11
Benzoate	125·5—126·5	+43·7	129—130	+33	130—133	+33·5

A mixed melting point of the alcohol obtained in this work with authentic butyrospermol showed no depression. It is possible that Wagner and Harris isolated a difficulty separable mixture of lupenyl and butyrospermyl acetate such as was encountered in working up the final mother-liquors from our acetate.

Fraction B melted over a rather wide range and was fractionally crystallised, to yield a product, m. p. 233—238°, unchanged on chromatography over alumina. This material gave an acetate which after chromatography had m. p. 248—250°. Analysis agreed with its formulation as a monoacetate of a diol $C_{30}H_{52}O_2$. It gave a negative tetranitromethane test and was transparent in the ultraviolet down to 212 mμ. It showed infrared bands at 3635 (non-bonded OH) and at 1738 cm.$^{-1}$ (CO of acetate) (in CCl_4). An unsuccessful attempt was made to form a diacetate and attempted oxidation with chromium trioxide–pyridine [7] yielded unchanged starting material. Dehydration by phosphorus oxychloride in pyridine gave in good yield lupenyl acetate.



(I)



(II)

This dehydration limits the possible structures for the parent diol (in the absence of other rearrangements) to 18α-oleanane-3β : 19α-diol (I; R = H) and lupane-3β : 20-diol (II; R = H). Ames, Davey, Halsall, and Jones [8] showed that the 19α-hydroxy-group of the monoacetate (I; R = Ac) was not acetylated under ordinary conditions and that this substance was dehydrated to form the lupeol-type of skeleton. It was however oxidizable to the 19-ketone. The evidence points to the structure of the diol monoacetate as (II; R = Ac) and experiments are in hand to confirm this.*

Experimental

M. p.s are corrected. Specific rotations were determined for chloroform solutions, ultraviolet absorption spectra for ethanol solutions with a Hilger Uvispek. Light petroleum refers to the fraction of b. p. 40—60°.

Extraction of the Fruit of Maclura pomifera.—Ripe fruit were sliced and oven-dried and then ground. The powdered material (250 g.) was extracted (Soxhlet) with light petroleum. Evaporation of the extract gave a brown oil (36 g.) which was refluxed in alcohol (1 l.) containing potassium hydroxide (40 g.) for 10 hr. Evaporation of most of the alcohol, followed by dilution with water and extraction of the solution with ether, yielded the non-saponifiable material

* At this stage of the work the Organic Chemistry laboratories of the University were destroyed by fire, so that proof of the structure has had to be deferred for some time.

[6] Heilbron, Jones, and Robins, *J.*, 1949, 444.
[7] Poos, Arth, Beyler, and Sarett, *J. Amer. Chem. Soc.*, 1953, **75**, 422.
[8] Ames, Davey, Halsall, and Jones, *J.*, 1952, 286.

(12·5 g., 5%) as a yellow oil which slowly solidified. This material (19 g.) was chromatographed in light petroleum–benzene (9 : 1) on alumina (B.D.H.). The material (11·5 g.) eluted by light petroleum–benzene was treated overnight with pyridine (25 ml.) and acetic anhydride (25 ml.). The mixture was filtered and the crystals (5·2 g.) were crystallised several times from methanol–chloroform, to yield lupenyl acetate, m. p. 218—219° (needles), $[\alpha]_D$ +42° (*c* 1·61) (Found: C, 82·1; H, 11·2. Calc. for $C_{32}H_{52}O_2$: C, 82·0; H, 11·2%). There was no m. p. depression on admixture with an authentic sample. Hydrolysis of this acetate yielded lupeol, m. p. 213—215° (mixed m. p. 213—215°) (Found: C, 84·6; H, 11·9. Calc. for $C_{30}H_{50}O$: C, 84·4; H, 11·8%), which yielded lupenyl benzoate, m. p. and mixed m. p. 268—270° (from methanol–chloroform), $[\alpha]_D$ +64° (*c* 1·35) (Found: C, 83·6; H, 10·4. Calc. for $C_{37}H_{54}O_2$: C, 83·7; H, 10·25%)

Isolation of Butyrospermyl Acetate.—The filtrate from the acetylation above was evaporated under reduced pressure and the residue was boiled with methanol and again evaporated. The solid residue, recrystallised from methanol–chloroform, had m. p. 129—135° (3·4 g.). After several recrystallisations this yielded butyrospermyl acetate, m. p. 144·5—147°, $[\alpha]_D$ +13° (*c* 0·88) (Found: C, 81·9; H, 11·0. Calc. for $C_{32}H_{22}O_2$: C, 82·0; H, 11·2%). The mother-liquors from the crystallisation of lupenyl acetate yielded an additional 460 mg. (total 2·06 g., *ca.* 0·5%). Hydrolysis of this acetate with potassium hydroxide in dioxan–methanol yielded butyrospermol, m. p. and mixed m. p. 111—112° (from aqueous methanol), $[\alpha]_D$ −13° (*c* 1·22) (Found: C, 84·6; H, 11·8. Calc. for $C_{30}H_{50}O$: C, 84·4; H, 11·8%), which with pyridine–benzoyl chloride gave the benzoate, m. p. 129—130° (from methanol–chloroform), $[\alpha]_D$ +33° (*c* 1·18) (Found: C, 83·7; H, 10·1. Calc. for $C_{37}H_{54}O_2$: C, 83·7; H, 10·25%).

Isolation of a New Diol.—The fraction of the non-saponifiable material eluted from alumina by benzene–ether (4 : 1) (fraction B) was rechromatographed. The material (3 g.), m. p. 220—230°, eluted by benzene–ether (9 : 1) was fractionally crystallised from methanol–acetone, to yield a product, m. p. 233—238°. The m. p. was unchanged on chromatography over neutral alumina (Woelm) and this material was treated with pyridine–acetic anhydride overnight. Working up of the reaction mixture yielded an acetate, m. p. 247—250° (from methanol–chloroform), which was chromatographed over neutral alumina (Woelm-Act I). Elution with benzene–ether (9 : 1) yielded the *diol monoacetate*, m. p. 248—250° (from methanol–chloroform), $[\alpha]_D$ +18° (*c* 1·22) (Found: C, 79·1; H, 11·4. $C_{32}H_{54}O_3$ requires C, 79·0; H, 11·2%).

Attempted formation of a diacetate. Heating the monoacetate (170 mg.) in pyridine (1 ml.) and acetic anhydride (3 ml.) on a water-bath for 2½ hr. and recrystallisation of the product from methanol–chloroform yielded unchanged monoacetate, m. p. and mixed m. p. 247—250°.

Dehydration of the Diol Monoacetate.—The monoacetate (100 mg.) was refluxed in pyridine (15 ml.) and phosphorus oxychloride (3 ml.) for 2 hr. The cooled solution was poured on crushed ice, then extracted with benzene. The extracts were washed successively with dilute hydrochloric acid, dilute aqueous sodium hydroxide, and water. Evaporation yielded crystals which were chromatographed over neutral alumina (eluted by light petroleum), to give lupenyl acetate (85 mg.), m. p. and mixed m. p. 219—220° (from methanol–chloroform).

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THE UNIVERSITY OF NEW ENGLAND,
ARMIDALE, N.S.W., AUSTRALIA.

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