60. *Triterpenoids. Part I. Morolic Acid, a New Triterpenoid Sapogenin.*

By D. H. R. Barton and C. J. W. Brooks.

The principal sapogenin from the heartwood of *Mora excelsa* Benth. has been shown to be a new triterpenoid hydroxy-acid, $C_{30}H_{48}O_3$, designated morolic acid. Replacement of the oxygen of this acid by hydrogen afforded the fundamental hydrocarbon identified as germanicene. Conversion of the carboxyl group into methyl gave germanicol. The action of acidic reagents on the oxide of methyl morolate acetate furnished, according to the proton-donating power of the medium, methyl olean-12 : 18-dienolate (methyl *iso*-dehydro-oleanolate) or methyl olean-11 : 13(18)-dienolate acetate (methyl dehydro-oleanolate acetate).

On melting, morolic acid decomposed to give oleanol in excellent yield, the same easy decarboxylation to the same product being observed with olean-13(18)-enolic acid.

Reduction of the above-mentioned oxide by lithium aluminium hydride followed by acetylation afforded a conjugated diene, shown to be norolean-16 : 18-dienyl acetate, the constitution of which was confirmed by a partial synthesis based on siaresinolic acid. This acetate was also obtained from the oxide of morolic acid acetate. It was smoothly isomerised by hydrogen chloride to norolean-12 : 18(17)-dienyl acetate, the constitution of which was proved by its formation from the facile decarboxylation of olean-12 : 18-dienolic acid.

These experiments provide evidence that morolic acid is olean-18-enolic acid.

The mechanism of the non-acid-catalysed decarboxylation of $\beta\gamma$-unsaturated acids is discussed. The theoretical correlation of the stabilisation of diolefinic systems with their degree of hyperconjugation is extended to include the conjugated dienes of the oleanolic acid series. Comment is made on the low intensity of absorption, in the ultra-violet region, of methyl olean-12 : 18-dienolate.

Naturally occurring saponins can be divided into two classes. The first of these gives steroidal sapogenins on hydrolysis, the second triterpenoid sapogenins. A consideration of molecular-rotation differences often helps in the assignment of a sapogenin to its correct class (Barton and Jones, *J.*, 1944, 659; Barton, *J.*, 1945, 813; 1946, 512, 1116; and later papers) and accordingly we welcomed the opportunity to investigate further a sapogenin obtained by

Farmer and Campbell (*Nature*, 1950, **165**, 237) at the Forest Products Research Laboratory, Princes Risborough, by acid hydrolysis of the saponin present in high concentration in the heartwood of *Mora excelsa* Benth. Our work on this problem was greatly facilitated by Mr. Campbell, who kindly provided us with an adequate supply of raw material, and by Sir John Simonsen, F.R.S., who gave us valuable advice in the initial stages of the investigation.

The first analyses of the crystalline sapogenin could be reconciled with the formula $C_{27}H_{42}O_3$ implying a steroid formulation. In apparent agreement acetylation afforded a monoacetate. However the specific rotation of the sapogenin ($[\alpha]_D$ +17° in acetone, +16° in dioxan, +13° in chloroform) rendered this somewhat questionable, since the steroid-sapogenin side chain normally confers pronounced lævorotation (Fieser and Fieser, " Natural Products Related to Phenanthrene," 3rd Edn., p. 591). Furthermore the change in molecular rotation on acetylation was +73°, which would correspond better with a triterpenoid than with a steroid formula (Barton, *loc. cit.*; the known steroidal sapogenins are all 3β-hydroxylated).

Further investigation of the sapogenin confirmed this view: it was shown to be a hydroxycarboxylic acid, $C_{30}H_{48}O_3$, by the preparation of a methyl ester, an acetate, an acetate methyl ester, and a benzoate methyl ester. The carboxyl group was sterically hindered and comparable in reactivity with that of oleanolic acid. The presence of 30 carbon atoms in the acid was shown by the equivalent of the acetate. The initial difficulty with regard to the analysis, referred to above, was due to solvation, the sapogenin crystallising well only from solvents containing methanol or another alcohol; analysis of a sample dried *in vacuo* at 160° was satisfactory for the unsolvated formula. The hydroxyl group was shown to be secondary by oxidation of the methyl ester by chromic acid to the corresponding ketone. Since this sapogenin is obtained from *Mora* wood and has one carboxyl and one hydroxyl group, we have named it morolic acid (Barton and Brooks, *J. Amer. Chem. Soc.*, 1950, **72**, 3314).

Although methyl morolate acetate resisted hydrogenation it gave a distinct colour with tetranitromethane indicative of unsaturation. Further, morolic acid, its acetate, methyl ester, methyl ester acetate, and methyl ester benzoate each absorbed one equivalent of oxygen from perbenzoic acid (see Table II, p. 269); the methyl ester acetate thus afforded the corresponding *oxide*, $C_{33}H_{52}O_4$, also obtained more expeditiously by the action of perhydrol–acetic acid at 100°. This oxide was saturated to tetranitromethane, thus indicating the presence of only one double bond in morolic acid. Triterpenoids of the β-amyrin series tend to furnish saturated ketones rather than epoxides under experimental conditions of this sort (*inter al.*, Ruzicka and Cohen, *Helv. Chim. Acta*, 1937, **20**, 804; Ruzicka, Müller, and Schellenberg, *ibid.*, 1939, **22**, 758; Picard, Sharples, and Spring, *J.*, 1939, 1045; Picard and Spring, *J.*, 1940, 1387). That the product from methyl morolate acetate was definitely an oxide was shown by its reaction with dry hydrogen chloride and with lithium aluminium hydride (see below).

Recently Ruzicka, Jeger, and their collaborators (*inter al.*, *Helv. Chim. Acta*, 1950, **33**, 672, 687, 711, 1050) have employed the presence or absence of an absorption maximum at about 11·8—12·4 μ. as indicative of a triply or fully substituted ethylenic linkage in triterpenoid compounds. Judged by this criterion the infra-red spectrum of methyl morolate acetate * (Fig. 1) indicated the presence of a triply substituted system $\rangle C{=}CH-$.

At this stage of the investigation it was clear that morolic acid was a pentacyclic hydroxy-acid very probably belonging to the triterpenoid series. The next step was to convert the hydroxy-acid into the fundamental hydrocarbon. This was carried out by reduction of morolic acid, or better of its methyl ester, by an excess of lithium aluminium hydride, to moradiol, $C_{30}H_{50}O_2$, a new triterpenoid diol. Chromic acid oxidation followed by careful chromatography furnished the corresponding dicarbonyl derivative, moronal, characterised as the bis-2 : 4-dinitrophenylhydrazone. Wolff–Kishner reduction of moronal gave morene, which corresponded in m. p. to the hydrocarbon germanicene prepared recently by David (*Bull. Soc. chim.*, 1949, [v], **16**, 427) from germanicol. Subsequent work (see below) confirmed that the two hydrocarbons were identical.

Moradiol readily afforded a diacetate which, by partial alkaline hydrolysis, gave the secondary monoacetate, a reaction sequence paralleled by the behaviour of betulin. Moradiol monoacetate contains a primary hydroxyl group and afforded a toluene-*p*-sulphonate, reduction of which by lithium aluminium hydride furnished only moradiol (cf. Schmid and Karrer, *Helv. Chim. Acta*, 1949, **32**, 1371). Chromic acid oxidation of moradiol monoacetate followed by careful chromatography afforded the corresponding aldehyde, acetoxymoral, which on Wolff–

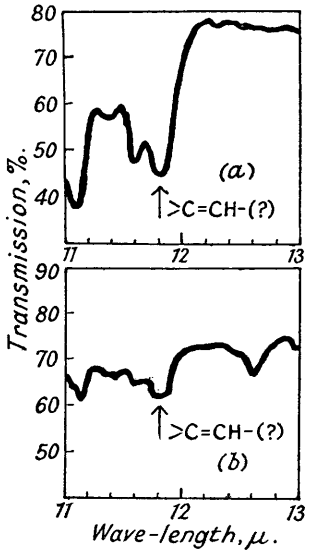
* We are indebted to Dr. R. Norman Jones of the N.R.C., Ottawa, for kindly making a preliminary determination of this spectrum. All the spectra reported here were taken at Harvard on the Baird Associates double-beam instrument (see Experimental).

Kishner reduction gave the alcohol, morol, $C_{30}H_{50}O$, together with some moradiol. Morol was shown to be identical with the known triterpenoid alcohol germanicol. This was established (see Experimental) by direct comparison with a specimen very kindly supplied by Dr. J. C. E. Simpson (*J.*, 1944, 283; cf. Dupont and Julia, *Bull. Soc. chim.*, 1947, [v], **14**, 1071) and confirmed by the identity of the infra-red spectra. A band near 12 μ. (see Fig. 2) confirms the trisubstituted nature of the ethylenic linkage.

The conversion of morolic acid into germanicol, which has been shown to belong to the β-amyrin–oleanolic acid group of triterpenoids (David, *ibid.*, 1949, [v], **16**, 155), establishes the triterpenoid nature of morolic acid and proves the position of the hydroxyl group, but it leaves undecided the precise location of the ethylenic linkage (see later) and the position of the carboxyl group. Evidence bearing on the latter problem was obtained by almost quantitative conversion of methyl morolate acetate oxide by hydrogen chloride into methyl dehydromorolate acetate, which was identical with methyl dehydro-oleanolate acetate obtained by selenium dioxide oxidation of methyl oleanolate acetate (Ruzicka, Grob, and van der Sluys-

FIG. 1.

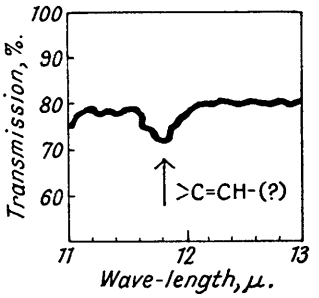
Methyl morolate acetate.



(*a*) *In* $CHCl_3$ (c, *approx.* 5·0).
(*b*) *In* CS_2 (c, *approx.* 5·0).

FIG. 2.

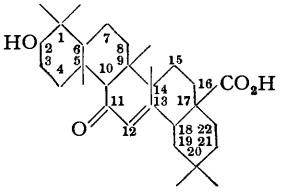
Morol, in $CHCl_3$ (c, *approx.* 5·0).

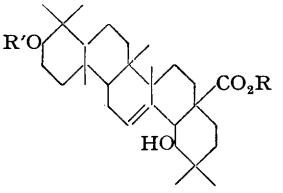


Veer, *Helv. Chim. Acta*, 1939, **22**, 788). This demonstrates that the carboxyl group of morolic acid is in the same position as in oleanolic acid.

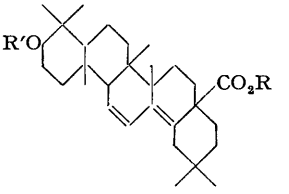
It is necessary at the present juncture to discuss the evidence bearing on the constitution of dehydro-oleanolic acid. This acid, or its simple functional derivatives, have been obtained in three ways. First, as the acetate methyl ester, by the route indicated above, secondly by the reduction of keto-oleanolic acid (I) by sodium and alcohol (Bilham, Kon, and Ross, *J.*, 1942, 532), and thirdly, as the acetate methyl ester, by the dehydration of methyl siaresinolate acetate (II; R = Me; R′ = Ac) by phosphoric oxide (Bilham, Kon, and Ross, *ibid.*, p. 540). Dehydro-oleanolic acid has two double bonds in conjugation in different rings as shown by the ultra-violet absorption maximum at 250 mμ. (Ruzicka, Grob, and van der Sluys-Veer, *loc. cit.*; cf. Ruzicka, Müller, and Schellenberg, *Helv. Chim. Acta*, 1939, **22**, 767) and, in view of the modes of formation mentioned above, two formulæ (III and IV; R = R′ = H) have been generally considered (compare the discussion by Green, Mower, Picard, and Spring, *J.*, 1944, 527). Ruzicka, Jeger, and Norymberski (*Helv. Chim. Acta*, 1942, **25**, 457) studied the oxidation of the analogous dehydro-β-amyrin acetate (V or VI) by lead tetra-acetate to give β-amyradienonyl acetate (VII) and considered that the formation of the latter provided firm evidence for the correctness of formula (VI). Actually this conclusion would be vitiated if, as is quite possible, the oxidation proceeded through the resonance-established radical (VIII)

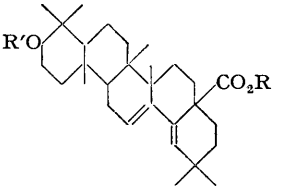
and too much significance should not be attached to it. However, there are two other recorded facts with regard to the formation and reactions of dehydro-oleanolic acid which we believe to be of considerable diagnostic value. The first of these is the method of formation from (I). Double bonds in conjugation do *not* move under the influence of alkali to thermodynamically more stable positions unless they move into conjugation with other *unsaturated* substituents. The rearrangement of (III; R = R′ = H), the first product of the reduction of (I), to (IV;

(I.) 



(II.)

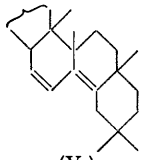
(III.) 

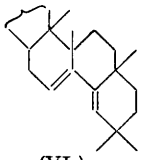


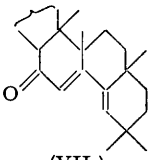
(IV.)

R = R′ = H) under the influence of alkali would, therefore, be very unlikely. This is strong evidence for formula (III). Secondly, the pyrolysis of dehydro-oleanolic acid affords " oleadienol I " (IX) (Bilham, Kon, and Ross, *J.*, 1942, 532), in which the two double bonds have moved *out* of conjugation. As is discussed in greater detail below this can best be explained if dehydro-oleanolic acid is formulated as (III; R = R′ = H).

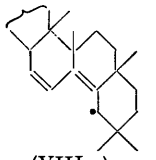
If (III; R = R′ = H) be the formula for dehydro-oleanolic acid then it should be possible to prepare an *iso*dehydro-oleanolic acid (IV; R = R′ = H). Furthermore the latter acid should be

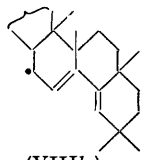

(V.)


(VI.)



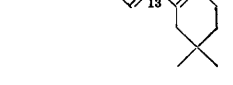
(VII.)


(VIII*a*.) ⟷


(VIII*b*.)

thermodynamically unstable with respect to (III; R = R′ = H), and should be rearranged thereto by acid in order to explain the observation by Bilham, Kon, and Ross (*loc. cit.*) on the dehydration of methyl siaresinolate acetate by phosphoric oxide. Both these predictions have been fulfilled. Oxidation of methyl morolate acetate by selenium dioxide afforded a methyl *iso*dehydro-oleanolate acetate which, unlike methyl dehydro-oleanate acetate ($[\alpha]_D$ −127°), exhibited pronounced dextro-rotation ($[\alpha]_D$ +209°). The corresponding methyl hydroxy-ester, obtained by fission of the oxide ring of methyl morolate acetate oxide, by heating with

(IX.) 

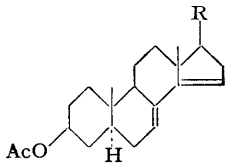


(X.)

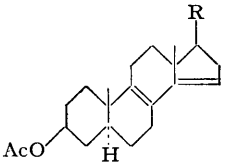
dilute methanolic sulphuric acid, also showed a pronounced dextrorotation ($[\alpha]_D$ +214°) and exhibited a maximum at 237 mμ. indicative of the presence of two double bonds in different rings. Isomerisation of methyl *iso*dehydro-oleanolate acetate by hydrogen chloride gave methyl dehydro-oleanolate acetate (III; R = Me, R′ = Ac) in almost quantitative yield. That *iso*dehydro-oleanolic acid, prepared by vigorous alkaline hydrolysis of the acetate methyl ester, possesses the formula (IV; R = R′ = H) was confirmed by pyrolysis, which furnished in good yield a nor-dienol, $C_{29}H_{46}O$, m. p. 189—191°, $[\alpha]_D$ +79°, in which the two double

bonds had *remained* in conjugation as shown by the ultra-violet absorption spectrum of the derived acetate ($\lambda_{max.}$ 238 and 244 mμ.; ε = 16,700, and 18,800 respectively). We formulate this dienol as (X) [norolean-12 : 18(17)-dienol],* the mechanism of its genesis being discussed in greater detail below.

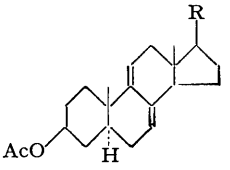
The correctness of the formulæ (III; R = R′ = H) and (IV; R = R′ = H) for dehydro- and *iso*dehydro-oleanolic acids is further confirmed by their absorption spectra. It has been enunciated as a general principle that cisoid chromophores absorb with lower intensity than the corresponding transoid chromophores (*inter al.*, Koch, *Chem. and Ind.*, 1942, **61**, 273; Braude *et al.*, *J.*, 1949, 1890, and references there cited). In agreement with this we have found that ergosterol B_3 acetate (XI; R = C_9H_{17}) (cf. Barton, *J.*, 1946, 512; for an improved method of preparation see Experimental) has a low-intensity absorption band ($\lambda_{max.}$ 242 mμ.; ε = 9900) relative to dehydro-α-ergostenyl acetate (XII; R = C_9H_{17}) ($\lambda_{max.}$ 248 mμ.; ε = 19,800) and ergosteryl D acetate (XIII; R = C_9H_{17}) ($\lambda_{max.}$ 242 mμ.; ε = 13,200) (see Barton and Cox, *J.*, 1949, 219). In the first-named of these acetates the chromophore is cisoid, in the other two it is transoid. Compounds of the β-amyrin series with two double bonds as in dehydro-



(XI.)



(XII.)



(XIII.)

oleanolic acid (III; R = R′ = H) absorb at 250 mμ. with ε about 25,000 (*inter al.*, Ruzicka, Grob, and van der Sluys-Veer, *Helv. Chim. Acta*, 1939, **22**, 788; Ruzicka, Müller, and Schellenberg, *ibid.*, p. 767), the high intensity of absorption corresponding to a transoid chromophore. In *iso*dehydro-oleanolic acid the value of ε is only about 10,000 (see Experimental) as expected for a cisoid chromophore.

It was of interest that the physical constants found by us for norolean-12 : 18(17)-dienol were quite close to those recorded by Noller and Carson (*J. Amer. Chem. Soc.*, 1941, **63**, 2238) and by Bilham, Kon, and Ross (*J.*, 1942, 535) for norechinocystadienol. Through the courtesy of Professor C. R. Noller (Stanford University, California) who kindly supplied us with an authentic specimen of norechinocystadienol, we have been able to compare the two alcohols directly. As shown in Table I and discussed at greater length in the Experimental section there can be little doubt that norechinocystadienol is substantially norolean-12 : 18(17)-dienol. This is in agreement with Noller and Carson's proposed formula (*loc. cit.*). The infra-red absorption spectra (Figs. 3 and 4) of the two alcohols were identical except for a small difference in relative intensity in two bands in the 9·6—10·2-μ. region.

* The nomenclature used in this paper is as follows. The syllable " olean " is considered a trivial designation for the C_{30} carbon skeleton [as in (I)] having the steric orientation present in β-amyrin and oleanolic acid. The fully saturated C_{30} hydrocarbon derived from oleanolic acid formally by reduction and the changes $CO_2H \longrightarrow CH_3$ and $OH \longrightarrow H$ is termed oleanane, and the derived unsaturated hydrocarbons are termed oleanene, oleandiene, etc. The derived C_{30} alcohols and ketones are named by the usual variations of the termination. Acids derived from the hydrocarbons by oxidation of a methyl group to carboxyl are termed oleananoic, oleandienoic acid, etc. (names of the type oleanane-carboxylic acid, which have been used, are incorrect as the addition of the carboxylic acid suffix indicates a C_{31} structure). When a hydroxyl group is present in such acids, the name ends in " olic acid," as in oleanenolic acid, oleandienolic acid, thus preserving the termination current for triterpenoid sapogenin acids. When a methyl group is replaced by hydrogen, giving a C_{29} compound, the prefix " nor " is used; this gives, *e.g.*, the name noroleandienol for (X); it will be noted that this replacement of methyl by hydrogen in the hydrocarbon is, in some cases, equivalent to decarboxylation of the corresponding acid. Numerals are used, as usual, for designation of the position of unsaturation or groups; such numerals need not normally be used for the hydroxyl or the carboxyl group of hydroxy-acids whose names end in " olic acid," the syllables " olean " being held to denote specifically that the hydroxyl and the carboxyl group are present at position 2 and 17 respectively. The term " oleanolic acid " is regarded as wholly trivial (as are its dehydro- and other derivatives), being equivalent to olean-12-enolic acid. It is hoped that this system may prove of general applicability in the triterpene field in cases when a " parent " name has been agreed. It is in agreement with the system of nomenclature adopted by the Swiss School, except for minor points. It may be noted that the name " oleanane " does not itself define the stereochemical orientation at $C_{(13)}$ or, yet, at $C_{(18)}$; the " olean " prefix, which defines the stereochemistry, is derived from oleanolic acid which is unsaturated at positions 12 : 13; and the orientation of the hydrogen atom at position 18 is uncertain (cf. XIV and XV); these problems will be considered in a forthcoming paper. [D. H. R. B. and the Editor.]

As would be expected, the reactions described above for morolic acid and its derivatives are paralleled in a number of cases by those recorded recently by David (*Bull. Soc. chim.*, 1949 [v], **16**, 155, 427; 1950, [v], **17**, 169) for germanicol and its derivatives. Although David tentatively accepted the incorrect formula for dienes of the dehydro-oleanolic acid type, his

TABLE I.

	Alcohol,		Acetate,		Benzoate,	
Substance.	m. p.	$[\alpha]_D$.	m. p.	$[\alpha]_D$.	m. p.	$[\alpha]_D$.
Norolean-12 : 18(17)-dienol	189—191°	+79°	187—188°	+66°	227—229°	+80°
Norechinocystadienol	189—191	+82 *	186—188	+46 †	231—233 *	—

* In dioxan; Noller and Carson, *J. Amer. Chem. Soc.*, 1941, **63**, 2238.
† In dioxan; Bilham, Kon, and Ross, *J.*, 1942, 535.

FIG. 3.

Olean-12 : 18(17)-*dienol in* $CHCl_3$ (c, *approx.* 2·0).

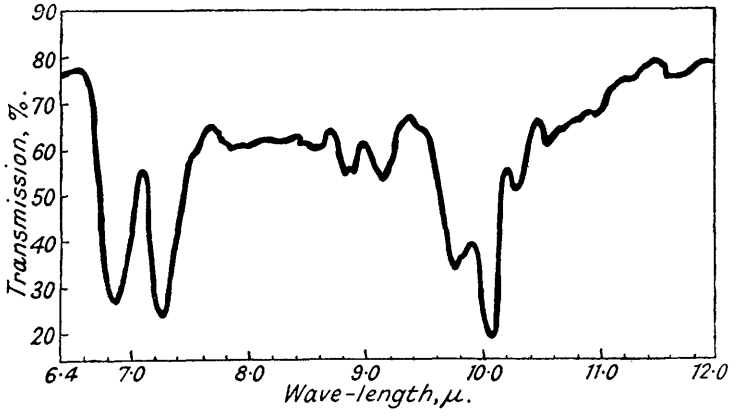
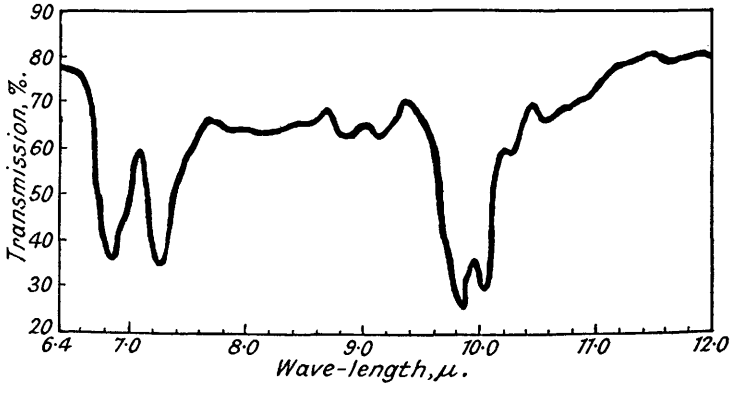


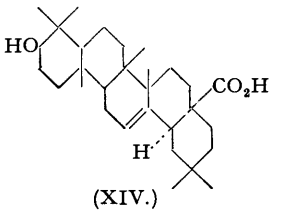
FIG. 4.

Norechinocystadienol in $CHCl_3$ (c, *approx.* 2·0).

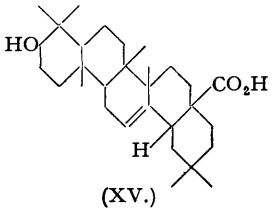


conclusion that the double bond in germanicol must be at one of the positions 12(13), 13(18), or 18(19) seems to us reasonable. There are then five possible formulæ (XIV—XVIII) for morolic acid. One of these (XIV or XV) must be the formula of oleanolic acid. The formula (XVI) has been proposed by Jeger, Norymberski, and Ruzicka (*Helv. Chim. Acta*, 1944, **27**, 1532; compare the corresponding β-amyrin analogue described by Ruzicka and Jeger, *ibid.*, 1941, **24**, 1236) for the acid from the hydrogenation product (methyl δ-oleanolate acetate) of methyl dehydro-oleanolate acetate (III; R = Me; R′ = Ac), on the grounds that it affords an oxide stable in hot acetic acid, conditions under which 12(13)-epoxides of the β-amyrin series are rearranged to the corresponding saturated ketones (cf. Ruzicka and Cohen, *ibid.*, 1937, **20**, 804; Ruzicka, Müller, and Schellenberg, *ibid.*, 1939, **22**, 758; Picard, Sharples, and

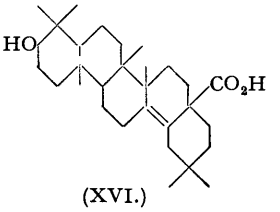
Spring, *J.*, 1939, 1045; Picard and Spring, *J.*, 1940, 1387, and later papers). Although we do not regard this evidence as particularly relevant a study of the reactions of the corresponding acid (δ-oleanolic acid) has confirmed this formula (see below). Methyl olean-13(18)-enolate acetate was also obtained (see Experimental) by the similar hydrogenation of methyl *iso*-dehydro-oleanolate acetate (IV; R = Me, R′ = Ac). The latter method of formation is a good example of 1 : 4-addition in catalytic hydrogenations (cf. Barton and Cox, *J.*, 1949, 214).



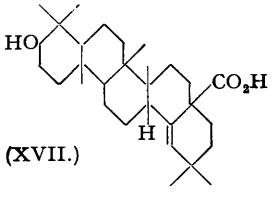
(XIV.)



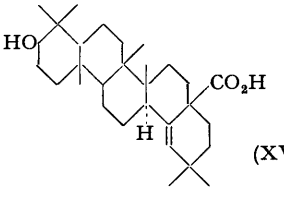
(XV.)



(XVI.)



(XVII.)



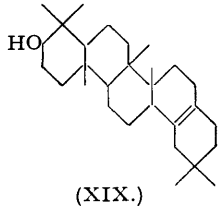
(XVIII.)

Attempts to characterise the double bond in morolic acid by the standard methods of oxidative attack proved fruitless. Methyl morolate acetate was completely resistant to osmium tetroxide. Ozonolysis afforded either unchanged starting material or, in quite good yield, the oxide. Chromic acid oxidation followed by careful chromatography furnished, besides unchanged starting material, the O_7 compound, $C_{33}H_{46}O_7$, previously obtained by Mower, Green, and Spring (*J.*, 1944, 256) by chromic acid oxidation of methyl dehydro-oleanolate acetate. Since the constitution of this compound is still unknown (see Jeger, Norymberski, and Ruzicka, *Helv. Chim. Acta*, 1944, **27**, 1532) its formation provides no evidence as to the position of the double bond in morolic acid.

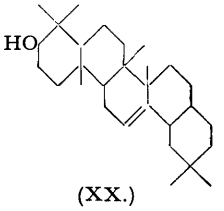
The first evidence bearing on the position of this double bond comes from the failure of morolic acid to lactonise under conditions adequate for the lactonisation of oleanolic acid. The carboxyl group in oleanolic acid must be polar in the stereochemical sense (for a discussion see Barton, *Experientia*, 1950, **6**, 316) in order to explain lactonisation. Inversion at $C_{(18)}$ should not alter this relationship and therefore 18-*iso*oleanolic acid should lactonise as readily as oleanolic acid.

By elimination then, formula (XVII) or (XVIII) is indicated for morolic acid, the double bond being in the βγ-position to the carboxyl group. It is a general rule that βγ-unsaturated acids are decarboxylated with ease on pyrolysis (for example, Arnold, Elmer, and Dodson, *J. Amer. Chem. Soc.*, 1950, **72**, 4359) and, as originally shown by Wallach (*Annalen*, 1906, **347**, 316; 1907, **353**, 287; 1908, **359**, 291; 1908, **360**, 26), the elimination of carbon dioxide is always accompanied by a shift of the double bond to the αβ-position. It is also true that many αβ-unsaturated acids are readily decarboxylated but, at least in the case of tertiary βγ-unsaturated acids, a shift of the double bond to the αβ-position *cannot* precede decarboxylation. It was therefore an important observation in the chemistry of morolic acid when it was discovered that the acid was very easily decarboxylated, merely on melting, to give an almost quantitative yield of oleanol. Oleanol had previously been prepared by Winterstein and Stein (*Z. physiol. Chem.*, 1931, **202**, 222) by pyrolysis of oleanolic acid. It is important that this latter preparation proceeds slowly, at nearly 100° above the decomposition (melting) temperature of morolic acid, in half an hour as opposed to the time taken in melting the morolic acid (a few seconds) and gives a poor yield of an impure product. In our opinion this remarkable contrast in reaction rate is best explained if morolic acid is βγ-unsaturated and oleanolic acid γδ-unsaturated. Oleanol then must have the formula (XIX) and not that (XX) previously accepted for this alcohol (see Elsevier's " Encyclopædia of Organic Chemistry," Vol. XIV, p. 544). In agreement, the infra-red spectrum of oleanol (Fig. 5) showed no significant absorption maximum in the region 11·8—12·4 μ., thus indicating a fully substituted and not a trisubstituted ethylenic linkage. As to the mechanism whereby oleanol is obtained from oleanolic acid, it is possible

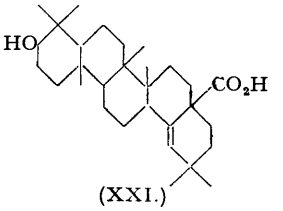
that the reaction proceeds by lactonisation, followed by isomerisation to olean-13(18)-enolic acid which, being βγ-unsaturated, would be decarboxylated easily. In agreement with this view pyrolysis of oleanolic lactone gave, in poor yield, oleanol.



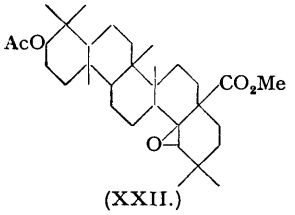
(XIX.)



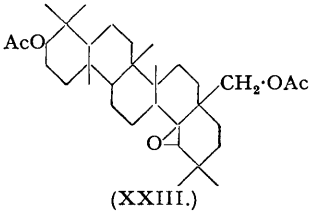
(XX.)



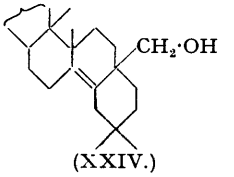
(XXI.)



(XXII.)



(XXIII.)

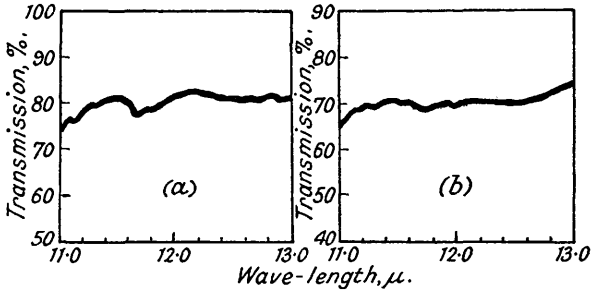


(XXIV.)

The above pyrolysis evidence is indicative of βγ-unsaturation in morolic acid. In order to exclude further consideration of (XVI) a brief study of δ-oleanolic acid was undertaken. Since this acid (as the methyl ester acetate) is obtained by the hydrogenation of (III; R = Me,

FIG. 5.

Oleanol, (a) in $CHCl_3$, *(b) in* CS_2 (c, *approx.* 5·0).

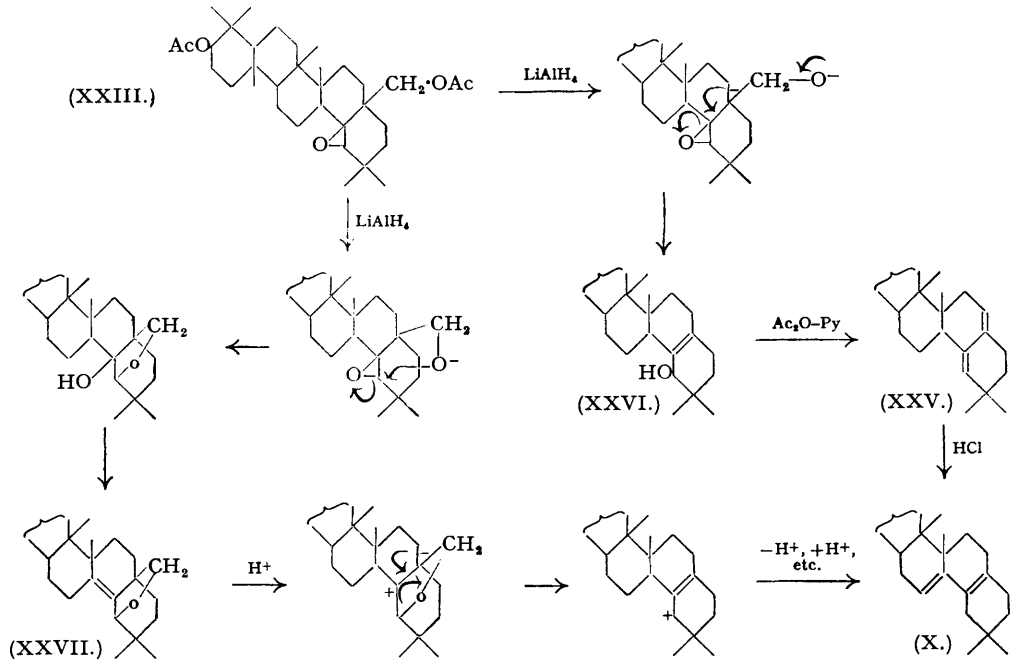


R′ = Ac) the ethylenic linkage must be at position 12(13) or 13(18) and not at 18(19).* δ-Oleanolic acid, prepared by vigorous alkaline hydrolysis of the methyl ester acetate, showed no tendency to give a bromo-lactone under conditions adequate for the formation of this derivative from oleanolic acid. It melted with vigorous decomposition (252—254°) and thereby afforded oleanol with the same ease as did morolic acid. Its formulation as the βγ-unsaturated olean-13(18)-enolic acid is thus confirmed. By exclusion then, morolic acid must be olean-18-enolic acid (XXI), and the methyl ester acetate oxide must be (XXII).

Further evidence as to the βγ-unsaturation of morolic acid was obtained in the following way. It was expected that lithium aluminium hydride reduction of moradiol diacetate oxide (XXIII) might open the oxide ring and give a triol or its dehydration product, for example (XXIV). In fact the reduction product, after acetylation and careful chromatography, furnished two compounds. The major product was an unsaturated acetate, $C_{31}H_{48}O_2$, m. p. 220—222°, $[\alpha]_D$ −19°. The preparation of (XXIV) by lithium aluminium hydride reduction of methyl olean-13(18)-enolate acetate proceeded without difficulty and the derived diacetate was quite different from the oxide reduction product. Hydrolysis of the acetate of m. p. 220—222° gave the corresponding alcohol, $C_{29}H_{46}O$, m. p. 181—183°, $[\alpha]_D$ −45°. A rational interpretation of these compounds became possible when it was discovered that they were

* Double bonds at positions 12(13), 13(18), and 18(19) in the triterpenoid skeleton, unlike those at 7(8) and 8(9) in the steroid series, show no indication of rearrangement under standard hydrogenating conditions (Pt–H_2 and acetic acid). Therefore this evidence is probably of diagnostic significance.

unsaturated conjugated dienes ($\lambda_{max.}$ 241 mμ.; ε = 17,200) with the double bonds in different rings. We formulate the dienol acetate as norolean-16 : 18-dienyl acetate (XXV) and interpret its formation by the mechanism outlined in the scheme. This mechanism implies that an intermediate allylic alcohol (XXVI) is formed, and this was proved to be the case although the intermediate was unstable and was not characterised. The second product of



the reduction was obtained as the acetate, $C_{32}H_{50}O_3$, m.p. 248—250°, $[\alpha]_D$ −2°, which was unsaturated to tetranitromethane, gave no hydroxyl band in the infra-red spectrum (confirmed by the ready elution from alumina), and showed the ordinary acetate carbonyl band at 5·75 μ. In view of the mode of preparation a ketonic carbonyl group can hardly be present. The infra-red spectrum (Fig. 6) showed a small absorption band near 9 μ. Such bands as this have been interpreted (Ruzicka, Baumgartner, and Prelog, *Helv. Chim. Acta*, 1949, **32**, 2069; Meyer, Jeger, and Ruzicka, *ibid.*, 1950, **33**, 687) as indicative of ethereal oxygen.* If this assignment is correct then a possible formula for the by-product would be (XXVII), its formation proceeding as indicated.

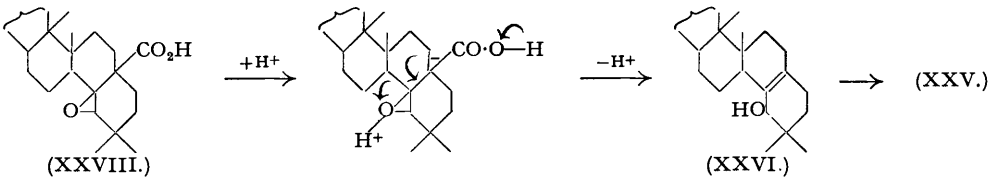
On treatment with dry hydrogen chloride norolean-16 : 18-dienol acetate furnished, in excellent yield, norolean-12 : 18(17)-dienol acetate (as X). The same acetate was obtained in the same way from the by-product acetate (? XXVII). Whilst the first-mentioned rearrangement is easily explained in terms of the addition and abstraction of protons, it is rather more difficult to rationalise the second. One possibility is indicated in the scheme above.

Reduction of methyl morolate acetate oxide by lithium aluminium hydride followed by acetylation also afforded norolean-16 : 18-dienol acetate, but there was no indication of the formation of the by-product acetate. It is very difficult to explain these remarkable elimination reactions other than on the basis of a βγ-oxido-structure for the starting material.

A further indication of the βγ-unsaturation of morolic acid was furnished by examination of the action of perbenzoic acid on morolic acid acetate. As mentioned above (p. 258) this compound consumes one equivalent of oxidant only. However, working up the product in the usual way afforded both an acid and a neutral fraction. We defer our report on the nature of the former fraction, but the neutral fraction was identified as pure norolean-16 : 18-dienol

* It is our experience that substances containing ethereal oxygen do exhibit a band at about 9 μ., but, on the other hand, so do many compounds in the steroid field lacking this function. It is, therefore, necessary to interpret evidence of this type with caution.

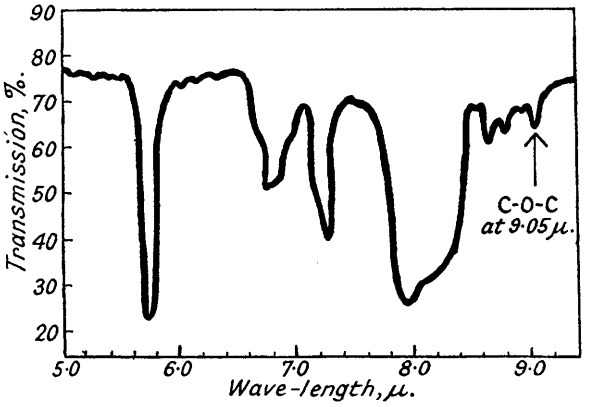
acetate. Presumably the βγ-oxido-acid acetate (XXVIII) is unstable under the mild acid conditions used in working up and decomposes as indicated below.



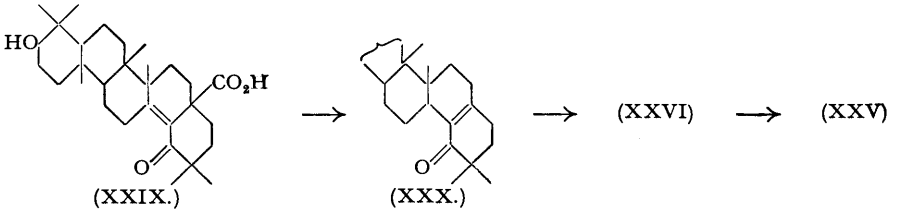
The evidence so far presented may be construed as a proof of the correctness of formula (XXI) for morolic acid. Confirmation of this has been obtained by what we regard as an unambiguous synthesis of norolean-16 : 18-dienol acetate starting from siaresinolic acid. Siaresinolic acid (II; R = R′ = H) was converted by known methods (Bilham, Kon, and Ross, *J.*, 1942, 540; Ruzicka, Grob, Egli, and Jeger, *Helv. Chim. Acta*, 1943, **26**, 1218) into 2-hydroxy-19-keto-olean-13(18)-en-17-oic acid (XXIX), pyrolysis of which furnished, after acetylation of the product and chromatography, 2-acetoxynorolean-17-en-19-one (XXX).

FIG. 6.

By-product of $LiAlH_4$ *reduction in* $CHCl_3$ (c, *approx.* 1·00).

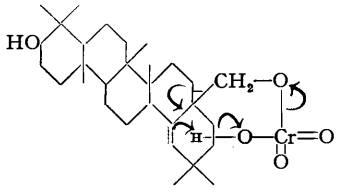
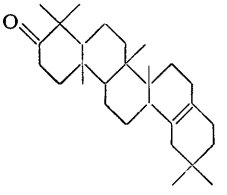


The position of the double bond in this compound is based on the general rule of βγ ⟶ αβ shift in the decarboxylation of βγ-unsaturated acids (see above) and is confirmed by the large change in rotation {the methyl ester of (XXIX) has $[\alpha]_D$ −209°; the acetate (XXX) shows $[\alpha]_D$ +138°}. The product was further characterised by hydrolysis to 2-hydroxynorolean-17-en-19-one. Reduction of 2-acetoxynorolean-17-en-19-one by lithium aluminium hydride gave an unstable compound of indefinite properties [presumably (XXVI)] which, on acetylation, furnished authentic norolean-16 : 18-dienol acetate, identical with the acetate obtained earlier from morolic acid.

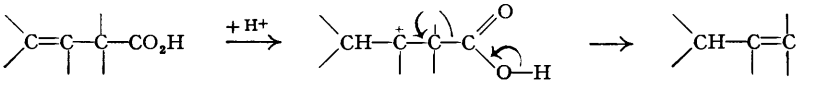


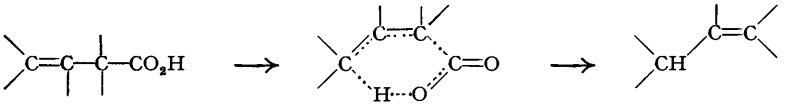
The behaviour of moradiol and of moradiol monoacetate on chromic acid oxidation, some aspects of which have already been discussed above, is deserving of further mention. The reaction products in both cases proved unexpectedly complex (for details see Experimental). Chromatography of the moradiol oxidation product afforded, in the most easily eluted fraction, oleanone (XXXI). The most easily eluted fraction of the monoacetate oxidation product

was likewise indicated to be the acetate of oleanol (XIX). Under the same conditions morolic acid acetate was recovered unchanged. These interesting by-products may be formed from the corresponding aldehydes, or possibly by an intramolecular process within a chromic acid ester as *illustrated* only in (XXXII). In any case we regard these reactions as additional evidence for $\beta\gamma$-unsaturation in morolic acid.

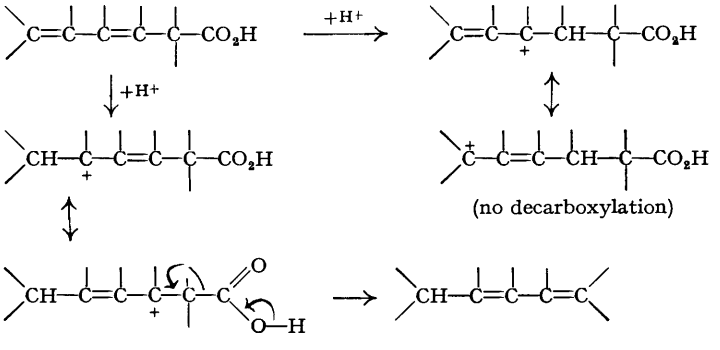
(XXXII.)  (XXXI.) 

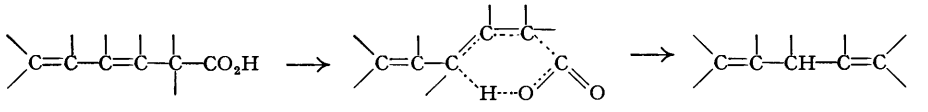
The evidence which has been accumulated on the constitution of morolic acid also enables one point of stereochemistry to be elucidated. There are two possible formulæ for morolic acid, (XVII) and (XVIII). In our opinion the evidence shows that the former of these is correct. There are two (more probable) general mechanisms which will explain the shift of the double bond from the $\beta\gamma$- to the $\alpha\beta$-position during decarboxylation of acids of the morolic acid type. In the first of these (*A*) a β-carbonium ion is formed, in the second (*B*) the transition state is of the intramolecular type without separation of charge (for example see Johnson and Hunt, *J. Amer. Chem. Soc.*, **1950**, **72**, **935**; we are indebted to Professor W. S. Johnson, University of Wisconsin, for an interesting discussion). Mechanism (*A*) should proceed best in the presence

(*A*) 

(*B*) 

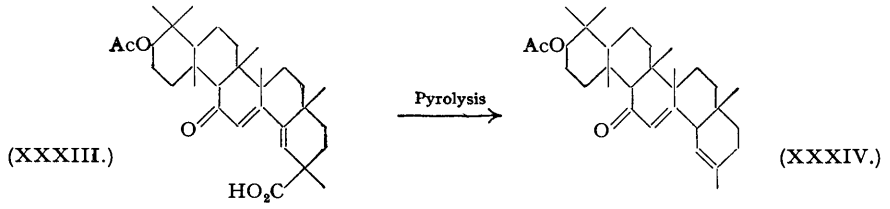
of acids, mechanism (*B*) should be the preferred one in the gaseous phase. It should be possible to distinguish between these two mechanisms by studying the decomposition of $\beta\gamma$: $\delta\varepsilon$-dienoic acids, for according to mechanism (*A*), as in (*C*), the two double bonds in the decarboxylated product should remain in conjugation, whereas according to mechanism (*B*), as in (*D*), they should shift out of conjugation.

(*C*) 

(*D*) 

Two examples are recorded in the triterpenoid field whereby to test this theory. The first has been mentioned in the discussion of the constitution of dehydro-oleanolic acid (see above).

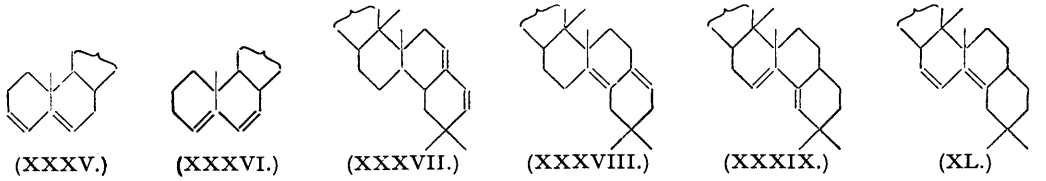
The second is the pyrolysis of dehydroglycyrrhetic acid acetate (XXXIII) at 205—210° to give nor-β-amyradienonyl acetate (XXXIV) (Ruzicka and Jeger, *Helv. Chim. Acta*, 1942, **25**, 775). In both cases the double bonds move out of conjugation; therefore the reactions must proceed by mechanism (*B*).



(XXXIII.) (XXXIV.)

Now norolean-16 : 18-dienol acetate has been obtained by two routes. That from morolic acid does not involve $C_{(13)}$; the other proceeds from siaresinolic acid and includes a decarboxylation. Siaresinolic acid has the $C_{(17)}$-carboxyl group in the same configuration as in oleanolic acid and it must be polar in the stereochemical sense in order to explain the ease of lactonisation on to $C_{(13)}$. In the decarboxylation of (XXIX) the C–H bond formed at $C_{(13)}$ must be on the *same* side of the molecule as the original C–carboxyl bond. The configuration at $C_{(13)}$ must be the same in morolic acid and therefore the formula (XVII) is correct.

A further point of theoretical interest in connection with these researches concerns the thermodynamic stability of olefinic or dienic systems. De La Mare, Hughes, and Ingold (*J.*, 1948, 17) have pointed out that in a number of aliphatic systems the stability of ethylenic linkages is governed by the number of C–H bonds that can hyperconjugate therewith. This is a useful generalisation which can be extended, albeit with caution for there are exceptions, to conjugated dienes in the steroid and di- and tri-terpenoid fields. Thus cholesta-3 : 5-diene



(XXXV.) (XXXVI.) (XXXVII.) (XXXVIII.) (XXXIX.) (XL.)

(XXXV) is more stable than cholesta-4 : 6-diene (XXXVI). The former has four hyperconjugating C–H bonds, the latter only three. Dehydro-oleanolic acid (III; R = R′ = H) (three hyperconjugating C–H bonds) is more stable than *iso*dehydro-oleanolic acid (IV; R = R′ = H) (two hyperconjugating C–H bonds). Similarly norolean-12 : 18(17)-dienol (X) with six hyperconjugating C–H bonds is more stable than norolean-16 : 18-dienol (XXV) with only two such. It must also be more stable than the alcohols (XXXVII), (XXXVIII), (XXXIX), and (XL) which, in agreement, have two, four, two, and three hyperconjugating C–H bonds respectively. The most serious exception to the rule that we have encountered so far is the instability of neoabietic acid relative to abietic acid (see Barton, *Quart. Reviews*, 1949, **3**, 36).

In connection with these aspects of thermodynamic stability it should be emphasised that, although norolean-12 : 18(17)-dienol is the most stable of the nor-dienes, its formation in the pyrolysis of *iso*dehydro-oleanolic acid is not due to the isomerisation of an initially formed and less stable nor-dienol. Under the same pyrolytic conditions and in the presence of oleanolic acid the thermodynamically unstable norolean-16 : 18-dienol acetate showed no indication of isomerisation.

Experimental.

M.p.s are uncorrected. Unless specified to the contrary rotations were determined for the sodium D line, in chloroform solution, at room temperature (15—25°). For polarimetry all specimens were dried *in vacuo* at 20° below their m. p.s or at 110°, whichever was the lower. Values of $[\alpha]_D$ have been approximated to the nearest degree. For the calculation of molecular rotations, $[\alpha]$ at *c*, 2·00, or at the nearest concentration to this at which measurements were made, have been used.

Unless specified to the contrary, infra-red spectra were determined in chloroform solution, with a Baird Associates (Cambridge, Mass., U.S.A.) self-recording double-beam instrument.

We are indebted to Dr. E. A. Braude (Imperial College) and Mr. Huang (Harvard) for determination of ultra-violet absorption spectra; solutions in absolute alcohol were used.

Merck's alumina for chromatography was used unless specified to the contrary.

In the text below the phrase " in the usual way " refers to dilution with water, extraction with ether, washing successively with aqueous potassium hydroxide, aqueous hydrochloric acid, and water, followed by evaporation of the ethereal solution *in vacuo*. Where necessary, water was removed from the residue by azeotropic distillation with benzene *in vacuo*.

Alkaline hydrolyses were effected by using several equivalents of potassium hydroxide and refluxing the reactants for 30—60 minutes in methanol or dioxan–methanol according to the solubility of the ester.

For methylations with diazomethane the acid was treated with an excess of ethereal diazomethane. After the mixture had been kept at room temperature until the evolution of nitrogen had ceased (about 30 minutes for 2-g. portions of morolic acid) the excess of diazomethane and the ether were removed *in vacuo* or on the steam-bath.

Morolic Acid from Mora *Saponin.*—The following procedure was found to be satisfactory. The saponin was heated under reflux (oil-bath) with alcohol (10 ml. per g. of saponin) containing concentrated hydrochloric acid (2 ml. per g. of saponin). The saponin first formed a deep-red solution which, after refluxing for a short time, began to deposit a crystalline precipitate of sapogenins. After 2 hours' refluxing, the mixture, now thick from the crystalline sapogenins, was allowed to cool and the precipitate was filtered off and washed with methanol. The crude sapogenin mixture was recrystallised several times from dioxan–methanol. 480 G. of saponin furnished 35 g. of pure *morolic acid*, m. p. 273° (decomp.), $[\alpha]_D$ +16° (*c*, 1·80 in dioxan), +33° (*c*, 0·59), +31° (*c*, 0·71) (Found: C, 76·5; H, 10·7. $C_{30}H_{48}O_3,CH_3{\cdot}OH$ requires C, 76·2; H, 10·7%). This represents a yield of 7·3%, as against 9·4% found by the Forest Products Research Laboratory (see Table III below) for material of somewhat lower m. p. The yield of acid of m. p. 263° was 42 g. (8·8%) and thus the results are in good agreement. Morolic acid was sparingly soluble in ethyl acetate or chloroform and tended to separate in an amorphous state unless methanol (or another alcohol) was present. Morolic acid showed no absorption in the ultra-violet region above 220 mμ. The acid was recovered unchanged after treatment adequate for the formation of a lactone (Winterstein and Stein, *Z. physiol. Chem.*, 1931, **199**, 64) or bromo-lactone (Winterstein and Stein, *ibid.*, pp. 50, 64) from oleanolic acid. On titration with perbenzoic acid in chloroform solution morolic acid consumed one equivalent of oxidant only (see Table II below).

Morolic Acid Acetate.—Morolic acid was refluxed with acetic anhydride for 1 hour. Working up in the usual way afforded the *acetate* which, recrystallised from chloroform–methanol or ethyl acetate–methanol, had m. p. 256—257° (decomp.), $[\alpha]_D$ +42° (*c*, 2·31), +44° (*c*, 1·77) (for analysis see Table III below). The equivalent weight of morolic acid acetate was determined by titration (phenolphthalein) with a solution of potassium hydroxide in dioxan–methanol, oleanolic acid having been used for standardisation (Found: equiv., 498. $C_{32}H_{50}O_4$ requires equiv., 499). The acetate gave a very pale yellow colour with tetranitromethane. For the titration with perbenzoic acid see Table II.

Methyl Morolate.—Morolic acid was esterified with diazomethane in the usual way. Recrystallised from ethyl acetate–methanol, the *methyl* ester had m. p. 228—229°, $[\alpha]_D$ +26° (*c*, 3·27), +26° (*c*, 2·21) +26° (*c*, 1·73) (Found: C, 78·7; H, 10·4. $C_{31}H_{50}O_3$ requires C, 79·1; H, 10·7%). 30 Mg. of the ester, refluxed with 200 mg. of potassium hydroxide in 10 ml. of methanol for half an hour and then left for 3 days, was almost quantitatively recovered unchanged. For the titration with perbenzoic acid see Table II.

TABLE II.

*Perbenzoic acid titrations.**

Substance.	Equivs. of per-acid consumed.	Substance.	Equivs. of per-acid consumed.
Morolic acid	0·91	Morolic acid acetate methyl ester	0·89, 1·10
Morolic acid acetate	0·97	Morolic acid benzoate methyl ester	1·00
Morolic acid methyl ester	0·89		

* In chloroform solution. The course of the reaction was followed until the uptake of per-acid was complete.

Methyl Morolate Acetate.—The methyl ester was refluxed with acetic anhydride for 30 minutes. Working up in the usual way and crystallisation of the product from chloroform–methanol afforded *methyl morolate acetate*, m. p. 263—264°, $[\alpha]_D$ +37° (*c*, 1·82), +38° (*c*, 1·76) (Found: C, 77·0; H, 10·05. $C_{33}H_{52}O_4$ requires C, 77·3; H, 10·2%). This gave a yellow colour with tetranitromethane, but resisted hydrogenation over a platinum catalyst in ethyl acetate at room temperature, in acetic acid at room temperature, or in acetic acid at 50°. In the Liebermann–Burchard reaction the initial colour was pink-blue, deepening to a permanent violet. This ester acetate was not isomerised when 100 mg. were heated in 10 ml. of acetic acid, to which two drops of concentrated sulphuric acid had been added, for 3 hours on the steam-bath. It was completely resistant to osmium tetroxide in ether (6 days at room temperature) and in dry pyridine (8 weeks at room temperature, then 8 hours at 100°); the starting material was recovered almost quantitatively in both cases. For the titration with perbenzoic acid see Table II.

In comparative experiments, methyl oleanolate acetate and olean-13(18)-enolate acetate were each recovered (almost quantitatively) unchanged after similar treatment with osmium tetroxide in dry pyridine.

Methyl Morolate Benzoate.—The methyl ester was heated in pyridine solution for 1½ hours at 100° with an excess of benzoyl chloride. Working up in the usual way afforded the *benzoate*, m. p. 194—195° (from ethyl acetate–methanol), $[\alpha]_D$ +52° (*c*, 2·08) (Found: C, 78·7; H, 9·2. $C_{38}H_{54}O_4$ requires C, 79·4; H, 9·45%). For the titration with perbenzoic acid see Table II.

Methyl Moronate.—Methyl morolate (0·5 g.) in ether–acetic acid was oxidised by chromic acid (excess) overnight. Working up in the usual way gave *methyl moronate* (400 mg.) which, recrystallised from methanol, had m. p. 165°, $[\alpha]_D$ +59° (*c*, 4·71), +58° (*c*, 4·59) (Found: C, 79·65; H, 10·2. $C_{31}H_{48}O_3$ requires C, 79·45; H, 10·3%).

Examination of Total Sapogenin Fraction.—By hydrolysis of *Mora* saponin with alcoholic hydrochloric acid the Forest Products Research Laboratory (F.P.R.L.), had obtained the total sapogenin fraction and had divided this into four main sub-fractions, hereinafter referred to as sapogenins I, II, III, and IV. The data * on these sapogenins, collected by the F.P.R.L., are summarised in Table III.

TABLE III.

Sapogenin fractions from Mora excelsa *Benth.*

Sapogenin.*	I.	II.	III.	IV.
Yield (on wt. of saponin), %	9·4	3·9	3·5	1·0
M. p.	261—263°	267—274°	241—245°	283—286°
$[\alpha]_D$ (in $CHCl_3$)	+30°	+62°	+64°	+79°
	+17° (in $COMe_2$)			
Found (%)	C, 76·7; H, 10·6	C, 77·8; H, 10·6	C, 77·7; H, 10·35	C, 78·75; H, 10·6
$C_{30}H_{48}O_3,C_2H_5{\cdot}OH$ requires (%)	C, 76·45; H, 10·8	—	—	—
Calc. for $C_{30}H_{48}O_3$ (%)	—	—	—	C, 78·9; H, 10·6
Loss in wt. at 160° *in vac.* (2 hrs.) (%)	4·2	0·5	0·0	—
Found (after heating as above) (%)	C, 78·5; H, 10·5	C, 78·55; H, 10·5	C, 77·8; H, 10·3	—
Acetate, m. p.	245—247°	252—254·5°	—	256—258°
Found (%)	C, 76·9; H, 10·15	C, 77·10; H, 10·1	—	—
$C_{32}H_{50}O_4$ requires (%)	C, 77·05; H, 10·1	C, 77·05; H, 10·1	—	—

* Arranged in order of increasing solubility in alcohol. Sapogenin IV was only deposited on prolonged storage of the filtered hydrolysate.

The least soluble fraction, sapogenin I, consisted of morolic acid.

Sapogenin IV was methylated with diazomethane and then acetylated; one recrystallisation from chloroform–methanol furnished methyl oleanolate acetate, m. p. 218°, $[\alpha]_D$ +69° (*c*, 4·09), which gave no depression in m. p. on admixture with an authentic specimen of m. p. 218°, $[\alpha]_D$ +69° (*c*, 1·99), obtained from oleanolic acid extracted from spent cloves.† Alkaline hydrolysis of the acetate ester of sapogenin IV afforded the corresponding methyl ester, m. p. 196—198° (from ethyl acetate–methanol). Authentic methyl oleanolate, obtained by similar hydrolysis of its acetate, had m. p. 196—198° and there was no depression in m. p. on admixture.

Sapogenin II was converted into the acetate methyl ester, m. p. *ca.* 203°, in the usual way and the product subjected to prolonged fractional crystallisation (with triangulation) from ethyl acetate–methanol. After nine steps in the triangulation (32 fractions) methyl morolate acetate was isolated having m. p. 259—261°, $[\alpha]_D$ +39° (*c*, 4·04), mixed m. p. 261—263° with authentic material (see above), and also methyl oleanolate acetate, m. p. 218°, likewise undepressed in m. p. on admixture with an authentic specimen. No other substance could be isolated. A mixture of equal amounts of morolic and oleanolic acids, dissolved in a little dioxan and precipitated with water, had m. p. 262—267°, mixed m. p. with sapogenin II 268—273°. Recrystallisation of the mixture from ethanol afforded material of m. p. 268—270° closely resembling sapogenin II in appearance. A similarly prepared mixture of the corresponding acetate methyl esters had m. p. 202—203°.

Sapogenin III (2 g.) was converted into the (amorphous) acetate methyl ester in the usual way. Chromatography over alumina (Birlec; each fraction crystallised once from chloroform–methanol) gave the following fractions.

Fraction 1: Eluant 300 Ml., 1 : 1 light petroleum (b. p. 40—60°)–benzene; cryst., m. p. *ca.* 200°.
,, 2: ,, 50 Ml., benzene; traces only.
,, 3: ,, 30 Ml., benzene; 50 mg., cryst., m. p. 250—255°.
,, 4: ,, 100 Ml., 1 : 1 benzene–ether; 20 mg., amorphous yellow solid, m. p. 230—250°.

Recrystallisation of fraction 1 gave methyl oleanolate acetate. Fractions 3 and 4 were not examined further.

Methyl Morolate Acetate Oxide.—(a) *Perbenzoic acid procedure* (cf. Table II). Methyl morolate acetate (1·07 g.) in dry chloroform (20 ml.) was treated with 1·31N-perbenzoic acid (5 ml.), also in chloroform, and left at 0°. After 20 hours 95% of the amount of per-acid required for one double bond

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* We are indebted to Mr. Campbell and Dr. Farmer for permission to quote their findings.

† The spent cloves were kindly placed at our disposal by Professor G. A. R. Kon, F.R.S., to whom we are much indebted.

had been consumed; after 43 hours the titre remained unchanged at a value corresponding to 1·1 double bonds. The solution was worked up in the usual way to give *methyl morolate acetate oxide*, m. p. 288°, $[\alpha]_D$ +30° (*c*, 1·73), +30° (*c*, 2·59) (Found: C, 74·95; H, 9·85. $C_{33}H_{52}O_5$ requires C, 74·95; H, 9·9%). This substance crystallised from chloroform–methanol in two distinct crystalline forms. However, its homogeneity was established by triangulation, carried to seven stages, and by extensive chromatography.

(b) *Perhydrol–acetic acid procedure.* Methyl morolate acetate (0·75 g.) in acetic acid (100 ml.) was heated on the steam-bath and perhydrol (2 ml., Merck) added during half an hour. Heating was continued for a further 2 hours. Dilution with water, filtration, and crystallisation from chloroform–methanol gave the oxide (0·66 g.), m. p. 288°, identical with that obtained by procedure (a). This is by far the more convenient method of preparation.

Methyl Morolate Oxide.—Alkaline hydrolysis of the foregoing acetate afforded *methyl morolate oxide* which, recrystallised from ethyl acetate–methanol, had m. p. 216—218°, $[\alpha]_D$ +21° (*c*, 2·79), +21° (*c*, 2·09) (Found: C, 76·4; H, 10·0. $C_{31}H_{50}O_4$ requires C, 76·5; H, 10·35%).

Methyl Morolate Benzoate Oxide.—Methyl morolate benzoate in warm glacial acetic acid was treated with perhydrol as described in procedure (b) above. Recrystallisation from methanol afforded the corresponding *oxide*, m. p. 203—204°, $[\alpha]_D$ +43° (*c*, 4·58) (Found: C, 77·65; H, 9·6. $C_{38}H_{54}O_5$ requires C, 77·25; H, 9·2%).

Action of Ozone on Methyl Morolate Acetate.—The acetate methyl ester (700 mg.) in ethyl chloride (50 ml.) was treated with ozone at −60° for half an hour. No volatile products could be detected and 650 mg. of unchanged starting material was recovered. 650 Mg. treated in the same way in carbon tetrachloride at 0° for 1 hour gave 450 mg. of unchanged starting material and no volatile products. 450 Mg. ozonised in 20 ml. of carbon tetrachloride at room temperature for 6 hours gave no volatile products; the solid product, m. p. 270—275°, obtained by evaporation of the carbon tetrachloride was chromatographed as indicated below, each fraction being crystallised once from chloroform–methanol:

Fraction 1: Eluant 50 Ml., benzene; m. p. 273—276°.
,, 2: ,, 50 Ml., benzene; m. p. 286—288°.
,, 3: ,, 50 Ml., benzene; m. p. 285—287°.

Further elution gave only traces of material. Fractions 1, 2, and 3 weighed 290 mg. Fraction 2 gave no depression in m. p. on admixture with pure methyl morolate acetate oxide, m. p. 288°.

Chromic Acid Oxidation of Methyl Morolate Acetate.—The acetate methyl ester (1·75 g.) was dissolved in acetic acid (150 ml.) at 100°. Chromic anhydride (0·9 g.) in 90% (vol.) aqueous acetic acid (50 ml.) was added during half an hour and the heating continued for a further 1·5 hours. After being worked up in the usual way the oxidation product was separated into neutral and acid fractions. The latter was very small and amorphous and was not examined further. The neutral fraction was chromatographed over alumina with the results indicated below, each fraction being crystallised once from chloroform–methanol before the m. p. determination:

Fraction	Eluant.	Yield (mg.).	M. p.
1	50 Ml., benzene	500	255—257°
2	50 Ml., ,,	50	253—255
3	150 Ml., ,,	Traces	Amorphous
4	70 Ml., 5 : 95 ether–benzene	Small	*ca.* 200
5	70 Ml., ,, ,,	50	245—247
6	140 Ml., 10 : 90 ,,	Small	278—280
7	70 Ml., 50 : 50 ,,	Traces	—
8	140 Ml., ether	—	—

Recrystallisation of fractions 1 and 2 afforded unchanged starting material. Fraction 6 depressed the m. p. of methyl morolate acetate oxide to 240—250° and was not investigated further. Recrystallisation of fraction 5 from methanol afforded the O_7 acetate, m. p. 247—248°, $[\alpha]_D$ +31° (*c*, 0·89) (Found: C, 71·0; H, 8·45. Calc. for $C_{33}H_{46}O_7$: C, 71·45; H, 8·35%). An authentic specimen of the O_7 acetate, prepared from methyl dehydro-oleanolate acetate by the method of Mower, Green, and Spring (*J.*, 1944, 256), had m. p. 248—249° and gave no depression in m. p. on admixture with the product described above.

Moradiol.—(a) *From methyl morolate.* Methyl morolate (1·0 g.) in dry ether (100 ml.) was mixed with lithium aluminium hydride (0·5 g.) in dry ether. After being left at room temperature for 10 minutes with occasional shaking, the mixture was worked up by acidification with aqueous acetic acid, etc., in the usual way. The reaction product, recrystallised from methanol, furnished beautiful long flat needles (615 mg.), m. p. 220°, $[\alpha]_D$ −11° (*c*, 1·61), of pure *moradiol* (Found: C, 81·25; H, 11·2. $C_{30}H_{50}O_2$ requires C, 81·4; H, 11·4%). Further batches were prepared with equal facility from the acetate methyl ester. In each case the yield of pure product was 60—70%.

(b) *From morolic acid.* Morolic acid (4·2 g.) in dry ether (1 l.) was treated with 0·31M-ethereal lithium aluminium hydride (350 ml.). The mixture was left overnight and then refluxed gently for 2 hours. Working up in the usual way gave pure moradiol (2 g.), m. p. 220°, and *ca.* 0·5 g. of recovered morolic acid. This is the less satisfactory method of preparation.

Moradiol (200 mg.) in dry ether was treated with excess of lithium aluminium hydride in ether and the suspension refluxed for 48 hours. Working up in the usual way gave 160 mg. of unchanged moradiol.

Moradiol Diacetate.—Moradiol, refluxed in benzene solution with excess of acetic anhydride for 1 hour and worked up in the usual way, afforded *moradiol diacetate* which, recrystallised from chloroform–methanol, had m. p. 273°, $[\alpha]_D$ +23° (*c*, 2·46) (Found: C, 76·2; H, 10·3. $C_{34}H_{54}O_4,0{\cdot}5CH_3{\cdot}OH$ requires

C, 76·3; H, 10·4%). It gave a pink colour, deepening to a permanent red, in the Liebermann–Burchard reaction.

Moradiol 2-Monoacetate.—Moradiol diacetate (1·59 g.) was refluxed with ethanol (50 ml.) and 0·023N-potassium hydroxide (in dioxan–ethanol) (150 ml.) for 2 hours, 2 ml. of water being added during this period. After working up of the mixture by dilution with water and extraction with chloroform 795 mg. of pure *moradiol 2-monoacetate* were obtained, which, recrystallised from chloroform–methanol, had m. p. 282—283°, $[\alpha]_D$ +5° (*c*, 1·88) (Found: C, 78·8; H, 10·5. $C_{32}H_{52}O_3$ requires C, 79·3; H, 10·8%). In the Liebermann–Burchard reaction the monoacetate gave a pink colour, deepening to a permanent red colour.

Moradiol Diacetate Oxide.—Moradiol diacetate (500 mg.), dissolved in warm glacial acetic acid (75 ml.), was heated on the steam-bath for 30 minutes whilst perhydrol (Merck; 2 ml.) in acetic acid (10 ml.) was added. A further hour's heating, dilution with water, filtration, and recrystallisation of the product from chloroform–methanol gave *moradiol diacetate oxide* (460 mg.), m. p. 253—255°. Repeated recrystallisation raised the m. p. to 255—256°, $[\alpha]_D$ +17° (*c*, 10·0) (Found: C, 75·4; H, 9·9. $C_{34}H_{54}O_5$ requires C, 75·25; H, 10·0%). A further experiment with 1·8 g. of moradiol diacetate furnished 1·66 g. of the oxide.

Moradiol 2-Monoacetate Toluene-p-sulphonate.—To the 2-monoacetate (220 mg.) in dry pyridine (2 ml.) and dry benzene (3 ml.) toluene-*p*-sulphonyl chloride (200 mg.) was added and the solution refluxed for 5 hours. Working up in the usual way furnished the required *ester* which, recrystallised from chloroform–methanol, had m. p. 184° (decomp.) (Found: C, 74·1; H, 9·45. $C_{39}H_{58}O_5S$ requires C, 73·3; H, 9·15%). This ester (100 mg.) was reduced by lithium aluminium hydride according to the directions of Schmid and Karrer (*Helv. Chim. Acta*, 1949, **32**, 1371) for cholesteryl toluene-*p*-sulphonate. The only product was moradiol, m. p. 218°, mixed m. p. with pure diol (m. p. 220°) 220°.

Chromic Acid Oxidation of Moradiol.—Several experiments were carried out. In each the reaction product was heterogeneous and could only be separated into its components by careful chromatography. The most easily eluted substance [from alumina by 50 : 50 light petroleum (b. p. 40—60°)–benzene], recrystallised from chloroform–methanol, had m. p. 174—175°, $[\alpha]_D$ +95° (*c*, 1·66) (Found: C, 83·85; H, 11·15. Calc. for $C_{29}H_{46}O$: C, 84·8; H, 11·3%), not depressed in m. p. on admixture with authentic oleanone, m. p. 174—176°, $[\alpha]_D$ +96° (*c*, 3·08), prepared by chromic acid oxidation of oleanol according to the directions of Winterstein and Stein (*Annalen*, 1933, **502**, 223), followed by chromatography. The identity was confirmed by the preparation of the corresponding 2 : 4-dinitrophenylhydrazones. In both cases purification was effected by chromatography. Authentic *oleanone 2 : 4-dinitrophenylhydrazone*, recrystallised from chloroform–methanol, had m. p. 245—246° (Found: N, 9·2, 9·8. $C_{35}H_{50}O_4N_4$ requires N, 9·5%), and mixed m. p. 245—246°, with the 2 : 4-dinitrophenylhydrazone, m. p. 245—246°, prepared from the moradiol oxidation product.

A typical oxidation experiment was as follows: Moradiol (1·93 g.) in 1 : 4 ether–acetic acid (500 ml.) was treated dropwise with chromic anhydride (880 mg.) in 95% aqueous acetic acid (125 ml.) during 2 hours. After being left overnight the solution was worked up by dilution with water, ether extraction, etc., in the usual way. The partly crystalline reaction product was chromatographed over 60 g. of alumina with the following results, each fraction being crystallised once from chloroform–methanol:

Fraction.	Eluant.	Product.
1	1000 Ml., 50 : 50 light petroleum (b. p. 40—60°)–benzene	Contained the oleanone (see above)
2	50 Ml., 70 : 30 light petroleum (b. p. 40—60°)–benzene	Cryst., m. p. 175—192°
3	50 Ml., ,, ,,	Cryst. (main fraction), m. p. 180—194°, $[\alpha]_D$ +85° (*c*, 5·14)
4	50 Ml., ,, ,,	Cryst., m. p. 175—194°
5	50 Ml., ,, ,,	Cryst., m. p. 175—190°, $[\alpha]_D$ +88° (*c*, 2·06)
6	50 Ml., ,, ,,	Cryst., m. p. 167—182°, $[\alpha]_D$ +87° (*c*, 2·04)
7	100 Ml., ,, ,,	Cryst., m. p. 182—195°
8	50 Ml., ,, ,,	Trace, m. p. 160—180°
9	200 Ml., benzene	Nothing
10	200 Ml., 5 : 95 ether–benzene	Trace, oil
11	150 Ml., 25 : 75 ,,	,,
12	50 Ml., ,, ,,	Cryst., m. p. 176—195°, $[\alpha]_D$ +41° (*c*, 2·42)
13	50 Ml., ,, ,,	Cryst., m. p. 160—162°, $[\alpha]_D$ +41° (*c*, 3·10)
14	50 Ml., ,, ,,	Cryst., m. p. 175—185°, $[\alpha]_D$ +33° (*c*, 4·17)
15	50 Ml., ,, ,,	Cryst., m. p. 185—192°
16	50 Ml., ,, ,,	Trace, cryst., m. p. 182—192°
17	100 Ml., ether	Cryst., m. p. 196—197°, $[\alpha]_D$ +26° (*c*, 2·30)
18	150 Ml., 1 : 1-chloroform—methanol	Cryst., m. p. 215—218°

Fractions 5—7, combined and recrystallised from chloroform–methanol, gave *moronal*, m. p. 185—200° (indefinite), $[\alpha]_D$ +88° (*c*, 3·48) (Found: C, 81·5; H, 10·6. $C_{30}H_{46}O_2$ requires C, 82·1; H, 10·6%). The orange *bis*-2 : 4-*dinitrophenylhydrazone*, prepared in the usual way and purified by chromatography followed by recrystallisation from chloroform–methanol, melted sharply at 240—241° (Found: N, 14·2. $C_{42}H_{54}O_8N_8$ requires N, 14·05%).

Repeated recrystallisation of fraction 17, without change in m. p., gave what was probably *moronol*, m. p. 196—197° (Found: C, 81·4; H, 11·0. $C_{30}H_{48}O_2$ requires C, 81·75; H, 11·0%).

Fraction 12 was heterogeneous since two recrystallisations from chloroform–methanol changed the

specific rotation to +77° (*c*, 0·84). Acetylation gave somewhat impure acetoxymoral, $[\alpha]_D$ +57° (*c*, 1·81) (see below).

Fractions corresponding to 12, 13, 14 and 15 were obtained in a number of other oxidations and, in each case, it was not possible to isolate a pure compound by direct recrystallisation. However acetylation of such fractions by pyridine–acetic anhydride, followed by careful chromatography, gave acetoxymoral, m. p. 250—260°, $[\alpha]_D$ +62° (*c*, 2·60).

Fraction 18 was somewhat impure moradiol.

In one experiment a further unidentified compound was obtained. It was eluted from alumina with 1 : 1 ether–benzene and, after recrystallisation from chloroform–methanol, melted sharply at 168°, $[\alpha]_D$ + 39° (*c*, 3·67) (Found : C, 80·15; H, 11·15%). Further recrystallisation did not alter the m. p. It was not investigated further.

Chromic Acid Oxidation of Moradiol 2-Monoacetate.—The monoacetate (663 mg.) in acetic acid (200 ml.) and ether (500 ml.) was oxidised by dropwise addition of chromic anhydride (150 mg.) in 97% aqueous acetic acid (15 ml.) during 45 minutes. The solution was left for 3 hours and then worked up in the usual way. The crystalline reaction product was chromatographed over 40 g. of alumina, each fraction being crystallised once from chloroform–methanol :

Fraction.	Eluant.	Product.
1	300 Ml., 80 : 20 light petroleum (b. p. 40—60°)–benzene	Trace only
2	200 Ml., 70 : 30 light petroleum (b. p. 40—60°)–benzene	Cryst., m. p. *ca.* 205°
3	100 Ml., ,, ,,	Nil
4	50 Ml., ,, ,,	Cryst., m. p. 255—260°
5	50 Ml., 60 : 40 light petroleum (b. p. 40—60°)–benzene	Cryst., m. p. 255—265°
6	100 Ml., ,, ,,	Cryst., m. p. 255—265°
7	200 Ml., ,, ,,	Cryst., trace
8	200 Ml., 40 : 60 light petroleum (b. p. 40—60°)–benzene	Nil
9	180 Ml., benzene	,,
10	50 Ml., 1 : 1 ether–benzene	,,
11	50 Ml., ,, ,,	Cryst., m. p. 247—248°
12	50 Ml., ,, ,,	Cryst., m. p. 260—270°

Fraction 2 was clearly identical with oleanyl acetate (see below). Fractions 4—6, recrystallised from chloroform–methanol, had $[\alpha]_D$ +63° (*c*, 2·39). After further recrystallisation the m. p. remained indefinite, but the specific rotation remained constant, $[\alpha]_D$ +62° (*c*, 2·68). Clearly these fractions consisted of *acetoxymoral* (Found : C, 80·0, 79·0; H, 10·7, 10·3. $C_{32}H_{50}O_3$ requires C, 79·6; H, 10·45%). The corresponding 2 : 4-*dinitrophenylhydrazone*, prepared in the usual way and purified by chromatography, melted sharply at 243° (Found : N, 8·85. $C_{38}H_{54}O_6N_4$ requires N, 8·45%).

Fraction 11, recrystallised from chloroform–methanol, gave a *substance*, m. p. 247—249°, $[\alpha]_D$ +40° (*c*, 1·30) (Found : C, 79·2; H, 10·65. $C_{32}H_{52}O_3$ requires C, 79·3; H, 10·8%). It was not investigated further. Fraction 12, on recrystallisation from chloroform–methanol, had m. p. 274—279°, $[\alpha]_D$ +12° (*c*, 1·80), and was shown to be somewhat impure starting material.

Morene (*Germanicene*).—Moronal (250 mg.) in the minimum of absolute ethanol was refluxed with several times the theoretical amount of 94% hydrazine for 2 hours. Concentration of the ethanolic solution furnished crude moronal dihydrazone, which (130 mg.) with 94% hydrazine (2 ml.) and a solution of sodium (250 mg.) in ethanol (6 ml.) was heated at 180° overnight. The reaction mixture was worked up in the usual way. Chromatography over alumina furnished in the first light petroleum (b. p. 40—60°) eluate morene (germanicene) (40 mg.); recrystallised from chloroform–methanol, this had m. p. 171—172°, $[\alpha]_D$ +3° (*c*, 0·60) (Found : C, 87·45; H, 12·3. Calc. for $C_{30}H_{50}$: C, 87·75; H, 12·25%).

Morol (*Germanicol*).—The crude hydrazone (80 mg.) of acetoxymoral, prepared as for moronal, was heated for 12 hours at 180° with 94% hydrazine (2 ml.) and a solution of ethanolic sodium ethoxide (250 mg. of sodium in 6 ml. of absolute ethanol). The reaction mixture was worked up in the usual way to give a crude product, m. p. *ca.* 150°. Acetylation with excess of acetic anhydride in pyridine on the water-bath for 2 hours and working up in the usual way gave a crystalline substance, m. p. 259—262°, depressed in m. p. by admixture with moradiol diacetate. Careful chromatography, over 10 g. of alumina, of this and the product of a further Wolff–Kishner reduction, gave the following results, each fraction being crystallised once from chloroform–methanol :

Fraction.	Eluent.	Product.
1	100 Ml., 90 : 10 light petroleum (b. p. 40—60°)–benzene	Plates, m. p. 267—270°
2	150 Ml., ,, ,,	Nil
3	50 Ml., 50 : 50 light petroleum (b. p. 40—60°)–benzene	Plates, m. p. 264—269°

Fraction 1, the main fraction, consisted of moryl acetate and had, on recrystallisation from ethyl acetate–methanol, m. p. 268—270°, $[\alpha]_D$ +18° (*c*, 1·13). It gave no depression in m. p. with an authentic specimen of germanicyl acetate, m. p. 269—271°, $[\alpha]_D$ recorded (Simpson, *J.*, 1944, 283) as +18°.

Fraction 3 gave no depression in m. p. on admixture with morodiol diacetate m. p. 273°.

Alkaline hydrolysis of moryl acetate gave morol (recrystallised from methanol), m. p. 173—175°, $[\alpha]_D$ +4° (*c*, 4·78). There was no depression in m. p. on admixture with an authentic specimen of germanicol, m. p. 171—173°, $[\alpha]_D$ +4° (*c*, 3·24).

Benzoylation of morol by heating with benzoyl chloride in pyridine solution on the steam-bath for

several hours, and then working up in the usual way, afforded moryl benzoate (recrystallised from chloroform–methanol), m. p. 261—262°, $[\alpha]_D$ +38° (*c*, 2·86). Germanicyl benzoate, prepared from germanicol in the same way and recrystallised from chloroform–methanol, had m. p. 261—262°, $[\alpha]_D$ + 37° (*c*, 4·31), and there was no depression in m. p. on admixture with moryl benzoate.

In the Liebermann–Burchard reaction morol behaved in the same way as methyl morolate acetate.

Oxide Fission by Dry Hydrogen Chloride.—(*a*) Methyl morolate acetate oxide (200 mg.) in chloroform (200 ml.) was treated for 30 minutes at room temperature with a stream of dry hydrogen chloride. Removal of the solvent *in vacuo* and recrystallisation of the residue from chloroform–methanol yielded needles (160 mg.), m. p. 220—221°, $[\alpha]_D$ −127° (*c*, 4·59). An authentic specimen of methyl dehydro-oleanolate acetate, prepared according to the procedure of Ruzicka *et al.* (*Helv. Chim. Acta*, 1939, **22**, 788) and recrystallised from chloroform–methanol, had m. p. 220—221°, $[\alpha]_D$ −128° (*c*, 4·49), −127° (*c*, 3·50). Identity was confirmed by a mixed m. p. which showed no depression.

(*b*) Methyl morolate benzoate oxide (220 mg.) in chloroform (20 ml.) was treated as under (*a*) above. The product (150 mg.), recrystallised from chloroform–methanol, had m. p. 282—283°, $[\alpha]_D$ −95° (*c*, 3·35). There was no depression in m. p. on admixture with authentic *methyl dehydro-oleanolate benzoate* (recrystallised from chloroform–methanol), m. p. 281—282°, $[\alpha]_D$ −93° (*c*, 4·12) (Found: C, 79·4; H, 9·3. $C_{38}H_{52}O_4$ requires C, 79·65; H, 9·15%). The latter was prepared by heating the corresponding alcohol (see below) in pyridine with excess of benzoyl chloride on the steam-bath for 3 hours and then working up the product in the usual way.

A portion of the benzoate, prepared from the oxide, was hydrolysed by alkali to the corresponding alcohol, m. p. 168° (from methanol), $[\alpha]_D$ −137° (*c*, 3·95). There was no depression in m. p. on admixture with methyl dehydro-oleanolate (recrystallised from methanol), m. p. 168°, $[\alpha]_D$ −139° (*c*, 5·16), prepared by alkaline hydrolysis of the acetate.

Oxide Fission by Aqueous-methanolic Sulphuric Acid.—Methyl morolate acetate oxide (106 mg.) was refluxed for 24 hours with 95% methanol (50 ml.) to which 2N-aqueous sulphuric acid (5 ml.) had been added. The solution was evaporated to two-thirds of its bulk, poured into water (200 ml.), and extracted with ether. The product, recrystallised from aqueous acetone, afforded pure *methyl iso-dehydro-oleanolate* (55 mg.), m. p. 179—180°, $[\alpha]_D$ +214° (*c*, 2·50) ($\lambda_{max.}$ 237 mμ.; ε = 10,200) (Found: C, 79·5; H, 10·7. $C_{31}H_{48}O_3$ requires C, 79·45; H, 10·3%).

The mother-liquors from a similar preparation were evaporated to dryness and the residue acetylated by dissolving it in pyridine, adding excess of acetic anhydride, and leaving the mixture for 2 days at room temperature. After being worked up in the usual way the reaction product was chromatographed over alumina, to give pure *methyl iso-dehydro-oleanolate acetate* which, recrystallised from ethyl acetate–methanol, had m. p. 195°, $[\alpha]_D$ +209° (*c*, 3·35) (Found: C, 77·9; H, 10·05. $C_{33}H_{50}O_4$ requires C, 77·6; H, 9·85%).

The above-mentioned acetate methyl ester (600 mg.) was heated with 20 ml. of 85% ethanol containing 20% of potassium hydroxide at 170° for 6 hours. After working up of the acid fraction in the usual way and recrystallisation from aqueous methanol, *iso-dehydro-oleanolic acid*, m. p. 232—233° (decomp.), $[\alpha]_D$ +224° [$\lambda_{max.}$ 229 and 238 mμ.; ε (both) = 8200], was obtained (Found: C, 77·9; H, 10·45. $C_{30}H_{46}O_3$, $0{\cdot}5CH_3{\cdot}OH$ requires C, 77·8; H, 10·3%).

The pure acetate methyl ester (40 mg.) in acetic acid (25 ml.) was hydrogenated over platinum oxide catalyst (20 mg.) for 24 hours, until there was no further absorption. Working up the product in the usual way and recrystallisation from chloroform–methanol gave crystals, m. p. 229—232°, not depressed by admixture with a specimen of authentic methyl olean-13(18)-enolate acetate, m. p. 230—231°, prepared according to the method of Jeger, Norymberski, and Ruzicka (*Helv. Chim. Acta*, 1944, **27**, 1532).

Methyl *iso*dehydro-oleanolate acetate (100 mg.) in chloroform (20 ml.) was treated with a stream of dry hydrogen chloride for 15 minutes at room temperature. Working up in the usual way and recrystallisation from chloroform–methanol afforded, in almost quantitative yield, methyl dehydro-oleanolate acetate, m. p. 219—221°, not depressed by admixture with an authentic specimen m. p. 220—221°.

Oxidation of Methyl Morolate Acetate by Selenium Dioxide.—To the acetate methyl ester (500 mg.) in acetic acid (50 ml.) selenium dioxide (250 mg.) was added and the solution refluxed for 6 hours. Worked up in the usual way the reaction product was obtained as an oil. It was chromatographed over 30 g. of alumina, each solid fraction being crystallised once from chloroform–methanol:

Fraction.	Eluant.	Product.
1	200 Ml., 75 : 25 light petroleum (b. p. 40—60°)–benzene	Nil
2	50 Ml., 50 : 50 light petroleum (b. p. 40—60°)–benzene	Cryst., m. p. 192°
3	50 Ml., ,, ,,	Cryst., m. p. 193°
4	100 Ml., ,, ,,	Trace
5	50 Ml., benzene	Cryst., m. p. 249—252° (decomp.)
6	50 Ml., benzene	Trace
7	50 Ml., 5 : 95 ether–benzene	Trace, amorphous

Fractions 2 and 3, combined and recrystallised from chloroform–methanol, gave methyl *iso*dehydro-oleanolate acetate, m. p. 193—194°, $[\alpha]_D$ +210° (*c*, 3·35), not depressed on admixture with an authentic specimen m. p. 195°.

Fraction 5 gave a 30° depression in m. p. on admixture with methyl diketo-oleandienolate acetate (m. p. 245—246°, $[\alpha]_D$ −146°) prepared according to the directions of Ruzicka *et al.* (*Helv. Chim. Acta*, 1939, **22**, 788). It was not investigated further.

Pyrolysis of Morolic Acid.—Morolic acid (2 g.) was heated at the m. p. in a fused mixed-nitrite bath.

As the acid melted, carbon dioxide was evolved; there was no further evolution of gas on continued heating. After 5 minutes the melt was rapidly cooled, whereupon it crystallised. One crystallisation from chloroform–methanol gave needles (1·65 g.), m. p. 222—224°, $[\alpha]_D$ +56° (*c*, 5·76). There was no depression in m. p. on admixture with an authentic specimen of oleanol (recrystallised from chloroform–methanol), m. p. 221—224°, $[\alpha]_D$ +56° (*c*, 1·49), prepared by pyrolysis of oleanolic acid according to the directions of Winterstein and Stein (*Z. physiol. Chem.*, 1931, **202**, 222).

The product from the pyrolysis of the morolic acid was refluxed with excess of acetic anhydride for 30 minutes. Working up in the usual way and recrystallisation from chloroform–methanol afforded the acetate, m. p. 201—203°, $[\alpha]_D$ +48° (*c*, 2·51). There was no depression in m. p. on admixture with oleanyl acetate, m. p. 201—204°, $[\alpha]_D$ +47° (*c*, 3·03), prepared in the same way from authentic oleanol.

*Pyrolysis of Olean-*13(18)*-enolic Acid.*—Methyl olean-13(18)-enolate acetate was heated at 180° in 85% ethanol (15 ml.) containing 20% of potassium hydroxide for 8 hours. Working up in the usual way furnished, in the acid fraction, *olean-*13(18)*-enolic acid* which, recrystallised from methanol, had m. p. 252—254° (decomp.) (Found: C, 75·35; H, 11·3. $C_{30}H_{48}O_3,CH_3{\cdot}OH$ requires C, 76·2; H, 10·7%). When heated at 270° for 1 minute, the acid (50 mg.) melted with effervescence. Recrystallisation of the cooled crystalline melt from methanol furnished crystals (35 mg.), m. p. 220—222° not depressed on admixture with authentic oleanol of m. p. 222—224°. Olean-13(18)-enolic acid was recovered unchanged under conditions adequate for the formation of a bromo-lactone from oleanolic acid (Winterstein and Stein, *Z. physiol. Chem.*, 1931, **199**, 56, 64).

Pyrolysis of iso*Dehydro-oleanolic Acid.*—The acid (350 mg.) was heated at 230° for 5 minutes and then at 240° for 5 minutes. The cooled melt was chromatographed over alumina, to furnish three fractions of *norolean-*12 : 18(17)*-dienol* which, recrystallised from methanol, had m. p. 189—191°, $[\alpha]_D$ +79° (*c*, 3·72) (Found: C, 84·7; H, 11·4. $C_{29}H_{46}O$ requires C, 84·8; H, 11·3%). Acetylation on the steam-bath for 2 hours with an excess of acetic anhydride in pyridine and then working up in the usual way gave *norolean-*12 : 18(17)*-dienol acetate* which, recrystallised from ethyl acetate–methanol, had m. p. 187—188°, $[\alpha]_D$ +66° (*c*, 3·66) ($\lambda_{max.}$ 238 and 244 mμ.; ε = 16,700 and 18,800 respectively) (Found: C, 81·3; H, 10·65. $C_{31}H_{48}O_2$ requires C, 82·05; H, 10·65%).

A specimen of norechinocystadienol, kindly supplied by Professor C. R. Noller (Stanford), had m. p. 189—191°, not depressed on admixture with norolean-12 : 18(17)-dienol, also of m. p. 189—191°. Acetylation of norechinocystadienol by treatment with an excess of acetic anhydride in pyridine at room temperature for 24 hours and then working up in the usual way gave the acetate which, recrystallised from methanol, had m. p. 175—178°. After three further recrystallisations the m. p. was 178—180° after sintering at 172° and was not depressed on admixture with norolean-12 : 18(17)-dienol acetate. Fractional crystallisation from aqueous methanol of the more soluble portion of the norechinocystadienol acetate finally gave an acetate, m. p. 186—188°, mixed m. p. with norolean-12 : 18(17)-dienol acetate also 186—188°.

Norolean-12 : 18(17)-dienol (100 mg.) was heated on the steam-bath with excess of benzoyl chloride in pyridine for 2 hours. Working up in the usual way afforded *norolean-*12 : 18(17)*-dienol benzoate*; recrystallised from chloroform–methanol, this had m. p. 227—229°, $[\alpha]_D$ +80° (*c*, 1·50) (Found: C, 83·95; H, 9·85. $C_{36}H_{50}O_2$ requires C, 84·0; H, 9·8%).

Reduction of Moradiol Diacetate Oxide by Lithium Aluminium Hydride.—Moradiol diacetate oxide (1·6 g.) in dry ether (700 ml.) was treated with a large excess of lithium aluminium hydride in ether, and the mixture was refluxed for 20 hours. The excess of hydride was destroyed by aqueous acetic acid, and the ethereal layer was separated, washed, and evaporated, yielding a crystalline residue. This sintered at 180° and melted with decomposition at 200—208°. It was acetylated by heating it in pyridine on the steam-bath for 1·5 hours with an excess of acetic anhydride. The crude acetate, worked up as usual, was chromatographed over alumina, each fraction being crystallised once from chloroform–methanol:

Fraction.	Eluant (50 ml.).	Product.
1	Benzene	*ca.* 100 Mg., m. p. 215—218°
2	,,	*ca.* 250 Mg., m. p. 212—216
3	,,	*ca.* 100 Mg., m. p. 184—188
4	,,	*ca.* 50 Mg., m. p. 233—238
5	95 : 5 Benzene–ether	*ca.* 20 Mg., m. p. 243—246

Further elution with benzene–ether and chloroform–methanol afforded only traces of oils.

Fractions 1 and 2, combined and recrystallised several times from chloroform–methanol, furnished fine needles (260 mg.) of *norolean-*16 : 18*-dienol acetate*, m. p. 220—222°, $[\alpha]_D$ −19° (*c*, 2·95) ($\lambda_{max.}$ 241 mμ.; ε = 17,200) (Found: C, 81·9; H, 10·8. $C_{31}H_{48}O_2$ requires C, 82·05; H, 10·65%). The Liebermann–Burchard reaction was red changing to yellow.

Fractions 4 and 5, together with their respective mother-liquors, were combined and re-chromatographed over alumina, each fraction being crystallised once from chloroform–methanol:

Fraction.	Eluant (50 ml.).	Product, m. p.	Fraction.	Eluant (50 ml.).	Product, m. p.
6	1 : 1 Light petroleum (b. p. 40—60°)–benzene	212—215°	10	1 : 1 Light petroleum (b. p. 40—60°)–benzene	243—245°
7	,, ,,	212—218	11	,, ,,	245—247
8	,, ,,	190—210	12	Benzene (200 ml.)	Trace
9	,, ,,	243—245 (main fraction)			

Fractions 9—11 inclusive were combined and recrystallised from chloroform–methanol. The *acetate* (p. 265) had the constant m. p. 248—250°, $[\alpha]_D$ −2° (*c*, 2·28) (Found: C, 79·6; H, 10·5. $C_{32}H_{50}O_3$ requires C, 79·6; H, 10·45%). It gave a yellow colour with tetranitromethane. In the Liebermann–Burchard reaction the colour sequence was red changing to deep reddish-violet.

Alkaline hydrolysis of norolean-16 : 18-dienol acetate in the usual way afforded *norolean*-16 : 18-*dienol* which, recrystallised from chloroform–methanol, had m. p. 181—183°, $[\alpha]_D$ −45° (*c*, 1·84) (Found: C, 84·0; H, 11·4. $C_{29}H_{46}O$ requires C, 84·8; H, 11·3%).

Norolean-16 : 18-dienol acetate (50 mg.) in chloroform (20 ml.) was treated at room temperature with a stream of dry hydrogen chloride. Removal of the solvent *in vacuo* and recrystallisation of the residue from chloroform–methanol furnished norolean-12 : 18(17)-dienol acetate, m. p. 187—189°, $[\alpha]_D$ +68° (*c*, 3·41), not depressed by admixture with an authentic specimen.

The acetate (10 mg.) of m. p. 248—250°, $[\alpha]_D$ −2°, in chloroform (5 ml.) was treated with a stream of dry hydrogen chloride for 15 minutes. Removal of the solvent *in vacuo* and recrystallisation of the residue from methanol gave norolean-12 : 18(17)-dienol acetate, m. p. 188—189°, undepressed in m. p. on admixture with an authentic specimen.

50 Mg. each of norolean-16 : 18-dienol acetate and oleanolic acid were fused together over a free flame. The melt was kept at 250° for 15 minutes. After cooling the product was chromatographed over alumina and recrystallised from methanol, to give nearly 40 mg. of unchanged norolean-16 : 18-dienol acetate, m. p. 217—219°, undepressed on admixture with the starting material.

Reduction of Methyl Morolate Acetate Oxide by Lithium Aluminium Hydride.—The oxide (1·0 g.) in dry ether (1·7 l.) was treated with an excess of lithium aluminium hydride in ether, and the solution refluxed on the steam-bath for 20 hours. The product was worked up as for the reduction of the moradiol diacetate oxide except that acetylation was carried out at room temperature for 24 hours. Chromatography over alumina gave the following fractions, each recrystallised once from chloroform–methanol:

Fraction.	Eluant (70 ml.).	Product, m. p.	Fraction.	Eluant (70 ml.).	Product, m. p.
1	1 : 1 Light petroleum (b. p. 40—60°)–benzene	219—221°	5	1 : 1 Light petroleum (b. p. 40—60°)–benzene	195—235°
2	,, ,,	214—218	6	95 : 5 Benzene–ether	235—245
3	,, ,,	170—183	7	—	250—253
4	,, ,,	173—176	8	90 : 10 Benzene–ether	252—254

Fraction 2, recrystallised, had m. p. 219—221°. Combined with fraction 1 and recrystallised once more it gave 310 mg. of norolean-16 : 18-dienol acetate, m. p. 220—222°, not depressed on admixture with the preparation from moradiol diacetate oxide.

Fractions 7 and 8, recrystallised from chloroform–methanol, had m. p. 254—256°, not depressed by admixture with moradiol diacetate oxide.

Fractions 3, 4, and 5 were combined and carefully re-chromatographed. It was not possible, however, to isolate any of the compound of m. p. 248—250°, $[\alpha]_D$ −2° (see above), and all fractions of the chromatogram gave large depressions in m. p. on admixture with this compound.

Norolean-16 : 18-*dienol Acetate from Siaresinolic Acid.*—2-Hydroxy-19-keto-olean-13(18)-en-17-oic acid (1·1 g.), m. p. 290—300° (decomp.), prepared from siaresinolic acid (obtained from Siamese gum benzoin according to the procedure of Winterstein and Egli, *Z. physiol. Chem.*, 1931, **202**, 207) by the method of Bilham, Kon, and Ross (*J.*, 1942, 540), was heated for 1 minute in an air-bath at 310°. On cooling, a brittle resin was obtained. This was dissolved in pyridine and heated on the steam-bath for 1 hour with an excess of acetic anhydride. After working up in the usual way the product was chromatographed over alumina. Elution of the column with benzene furnished 2-*acetoxynorolean*-17-*en*-19-*one* (250 mg.) which, recrystallised from methanol, had m. p. 232—233°, $[\alpha]_D$ +137° (*c*, 3·11), +139° (*c*, 1·31) ($\lambda_{max.}$ 248 mμ.; ε = 8400) (Found: C, 79·8; H, 10·8. $C_{31}H_{48}O_3$ requires C, 79·45; H, 10·3%).

This nor-ketone (100 mg.) in dry ether (100 ml.) was reduced by an excess of lithium aluminium hydride in ether, the solution being refluxed for 2 hours. The excess of the reducing agent was destroyed by aqueous acetic acid, and the ethereal layer separated and washed with sodium hydrogen carbonate solution. Evaporation gave a residue which, recrystallised, after azeotropic drying with benzene, from chloroform–light petroleum (b. p. 40—60°), melted unsharply at *ca.* 201° (decomp.) and had $[\alpha]_D$ +18° (*c*, 0·51). On further recrystallisation from the same solvent mixture the m. p. remained indefinite and finally fell to 170—180° (decomp.).

The reduction product (50 mg.) was heated in pyridine on the water-bath with excess of acetic anhydride for 2 hours. After working up in the usual way and one recrystallisation from methanol, needles were obtained, having $[\alpha]_D$ −17° (*c*, 1·98), m. p. 216—218°, not depressed on admixture with norolean-16 : 18-dienol acetate. The acetylated product was chromatographed over alumina, each fraction being crystallised once from methanol:

Fraction.	Eluant.	Product, m. p.
1	25 Ml., 75 : 25 light petroleum (b. p. 40—60°)–benzene	218—220°
2	50 Ml., ,, ,,	220—222 (main fraction)
3	50 Ml., 50 : 50 light petroleum (b. p. 40—60°)–benzene	217—219
4	100 Ml., ,, ,,	216—218 (trace)
5	100 Ml., Benzene	Nil

Fraction 2 weighed 32 mg. and gave no depression in m. p. on admixture with norolean-16 : 18-dienol acetate, m. p. 220—222°, prepared from morolic acid. Fractions 1, 3, and 4 combined weighed

8 mg. Fraction 2 was subjected to alkaline hydrolysis in the usual way. The reaction product, recrystallised from methanol, had m. p. 181—183° and gave no depression on admixture with norolean-16 : 18-dienol of the same m. p.

The experiment described above was repeated with 100 mg. of 2-acetoxynorolean-17-en-19-one, but the reduction product was acetylated at room temperature for 24 hours. The results were identical in all respects with those just recorded.

Alkaline hydrolysis of 2-acetoxynorolean-17-en-19-one (70 mg.) under the usual conditions furnished 2-*hydroxyolean*-17-*en*-19-*one* which, recrystallised from methanol, had m.p. 258° (50 mg.), $[\alpha]_D$ +143° (*c*, 3·77) (Found : C, 82·05; H, 11·2. $C_{29}H_{46}O_2$ requires C, 81·65; H, 10·85%).

Action of Perbenzoic Acid on Morolic Acid Acetate.—Morolic acid acetate consumed one equivalent of oxidant on treatment with excess of perbenzoic acid (cf. p. 258). Working up the product of this reaction proved unexpectedly complicated and we defer a full exposition of the experimental results until the constitution of one of the reaction products, originally found in the acid fraction, has been established.

Morolic acid acetate (1·1 g.) in chloroform (100 ml.) was treated with 1·145N-chloroformic perbenzoic acid (10 ml.) and left at 0° for 27 hours. It was then mixed with ether and water, and the acid fraction extracted with concentrated potassium hydroxide solution. The neutral fraction, washed with aqueous hydrochloric acid and then evaporated, was recrystallised from chloroform–methanol, to give norolean-16 : 18-dienol acetate (*ca.* 500 mg.), m. p. and mixed m. p. with an authentic specimen 220—222°.

2 : 28-*Dihydroxyolean*-13(18)-*ene*.—During the experiments described in this paper 2 : 28-dihydroxyolean-13(18)-ene and its diacetate were prepared for comparative purposes. It is our understanding that these two compounds were prepared by a different route by Professor E. R. H. Jones, F.R.S., at an earlier date and that their properties will be fully reported by him in a subsequent paper.

2 : 28-Dihydroxyolean-13(18)-ene, prepared by lithium aluminium hydride reduction of methyl olean-13(18)-enolate acetate and recrystallised from methanol (after purification through the diacetate, see below), had m. p. 269°, $[\alpha]_D$ −45° (*c*, 0·60) (Found : C, 78·6; H, 11·55. Calc. for $C_{30}H_{50}O_2,CH_3{\cdot}OH$: C, 78·4; H, 11·45%).

2 : 28-Diacetoxyolean-13(18)-ene, recrystallised from chloroform–methanol, had m. p. 194—195°, $[\alpha]_D$ −52° (*c*, 6·36), −55° (*c*, 5·47) (Found : C, 77·9; H, 10·35. Calc. for $C_{34}H_{54}O_4$: C, 77·5; H, 10·35%).

Pyrolysis of Oleanolic Lactone.—The lactone (200 mg.), prepared by a method which will be reported later, was heated at 300°, raised to 350° during 5 minutes. The resin obtained on cooling was acetylated with an excess of boiling acetic anhydride, and the product chromatographed over alumina. 90 : 10 Light petroleum (b. p. 40—60°)–benzene eluted 30 mg. of oleanol acetate, m. p. 197—201°. Alkaline hydrolysis afforded oleanol which, recrystallised from methanol, had m. p. 222—224°, not depressed on admixture with oleanol obtained by the pyrolysis of oleanolic acid.

Action of Hydrogen Chloride on Various Substances.—The following general procedure was adopted. The compound (200 mg.) in chloroform (20 ml.) was treated with a stream of dry hydrogen chloride for 30 minutes. The solvent was removed *in vacuo* and the residue recrystallised from chloroform–methanol or methanol. Morolic acid, morolic acid acetate, methyl olean-13(18)-enolate acetate, methyl oleanolate acetate, and oleanol were each recovered unchanged in almost quantitative yield.

Absorption Spectra of Steroidal Dienes.—(*a*) Dehydro-α-ergostenyl acetate, as prepared by Barton and Cox (*J.*, 1949, 214), had $\lambda_{max.}$ 248 mμ., ε = 19,800.

(*b*) For the absorption spectrum of ergosteryl D acetate see Barton and Cox, *J.*, 1949, 219.

(*c*) Ergosteryl B_3 acetate was prepared in the following way, which gives a purer product than that described in the literature (cf. Windaus *et al.*, *Annalen*, 1931, **488**, 91; Dithmar and Achtermann, *Z. physiol. Chem.*, 1932, **205**, 55). Ergosteryl acetate (3·0 g.) in chloroform (100 ml.) was treated with dry hydrogen chloride gas at −30° for 2 hours. The solution was poured, at once, into an excess of ice-cold sodium hydrogen carbonate solution. Extraction with ether, washing with water, and evaporation *in vacuo* gave a crude acetate, m. p. 123—125°. Several recrystallisations raised the m. p. to a constant value of 140—141°; $[\alpha]_D$ was then −221° (*c*, 0·70). Careful fractionation of this material revealed that it was pure ergosteryl B_3 acetate.

A better yield of the B_3 sterol acetate is obtained if purification is effected through the benzoate. Purified by recrystallisation from ethyl acetate–ethanol *ergosteryl* B_3 *benzoate* had m. p. 158—159°, $[\alpha]_D$ −187° (*c*, 2·24) (Found : C, 83·8; H, 9·35. $C_{35}H_{48}O_2$ requires C, 84·0; H, 9·6%). Alkaline hydrolysis and working up in the usual way afforded ergosterol B_3 which, recrystallised from ethyl acetate–methanol, had m. p. 139°, $[\alpha]_D$ −238° (*c*, 3·12). Acetylation in the usual way in pyridine solution gave ergosteryl B_3 acetate with the physical constants {$[\alpha]_D$ −220° (*c*, 2·13)} recorded above. It showed $\lambda_{max.}$ 242 mμ., ε = 9900.

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