

The constituents of olive leaves / by Frederick B. Power and Frank Tutin.

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Publication/Creation

London : Wellcome Chemical Research Laboratories, [1908]

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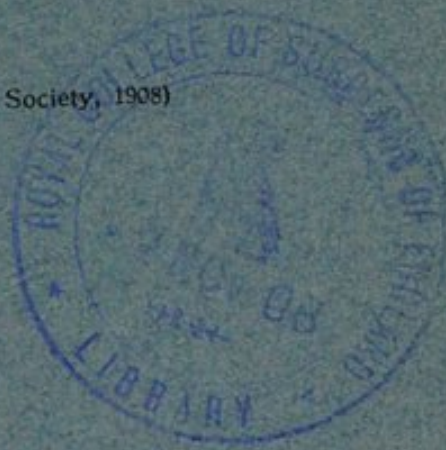
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THE CONSTITUENTS
OF
OLIVE LEAVES

BY
FREDERICK B. POWER, PH.D.
AND
FRANK TUTIN

(From the Transactions of the Chemical Society, 1908)



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LXXXIII.—*The Constituents of Olive Leaves.*

By FREDERICK BELDING POWER and FRANK TUTIN.

THE leaves of the olive tree, *Olea Europaea*, Linné, were employed many years ago as a remedy in intermittent fever, and quite recently have again been brought to notice on account of their reputed therapeutic value as a tonic and febrifuge (*Pharm. J.*, 1906, **77**, 376).

The only recent investigation of these leaves that has been recorded appears to be that of F. Canzoneri (*Gazzetta*, 1906, **36**, ii, 372).

This author extracted the leaves with 95 per cent. alcohol, and observed that the liquid, on cooling, deposited small quantities of crystals, consisting apparently of two wax-like substances melting at $85-100^{\circ}$ and $69-70^{\circ}$ respectively. From the alcoholic liquid, he obtained a white compound, $C_{25}H_{44}O_3$ or $C_{24}H_{42}O_3$ (m. p. $297-298^{\circ}$), soluble in ether; a white, crystalline, resin-like substance (m. p. $253-255^{\circ}$); a substance melting at about 236° ; a crystalline acid (m. p. 165°) insoluble in ether; a substance melting at 180° when crystallised from acetic acid, and at $200-235^{\circ}$ after recrystallisation from alcohol; an acid of dough-like consistence, turning brown on exposure to the air, together with mannitol, gallic and tannic acids.

It is evident that the above-mentioned products were, for the most part, of a very indefinite character, and that they did not represent pure substances. The compound $C_{25}H_{44}O_3$ or $C_{24}H_{42}O_3$, melting at $297-298^{\circ}$, would appear, however, to have been the hydrated form of a substance having the formula $C_{31}H_{50}O_3$ (m. p. $303-304^{\circ}$), which the present authors have isolated and designated *oleanol*, since compounds possessing the formulæ $C_{24}H_{42}O_3$ and $C_{31}H_{50}O_3 \cdot H_2O$ respectively have approximately the same percentage composition.

For the purpose of ascertaining more precisely the nature of the constituents of olive leaves, their complete investigation was undertaken, and the results are embodied in the present communication.

EXPERIMENTAL.

The material employed in this investigation consisted of the leaves of *Olea Europaea*, Linné, which had been freshly gathered for us during the early spring in the neighbourhood of Ventimiglia, on the Italian Riviera.

A portion of the leaves was first tested for the presence of an alkaloid, but with a negative result.

In order to ascertain the general characters of the constituents of the leaves, 50 grams of the ground material were successively extracted in a Soxhlet apparatus with various solvents, when the following amounts of extract, dried at 100° , were obtained:

Petroleum (b. p. $35-50^{\circ}$)	extracted	2.86	per cent.
Ether	"	7.16	"
Chloroform	"	1.00	"
Ethyl acetate	"	6.26	"
Alcohol	"	12.56	"
Total		29.84	"

For the purpose of a complete examination, a quantity (28.35 kilograms) of the ground leaves was extracted by continuous percolation with hot alcohol. One kilogram of the resulting extract,

containing 567 grams of extracted matter, was then mixed with water and distilled with steam, when a trace of an aromatic essential oil was obtained. The mixture contained in the distillation flask was filtered while hot, and a quantity (163.5 grams) of a pulverulent, green resin (A) thus collected, which was washed with boiling water, and dried. The combined filtrate and washings, on cooling, deposited 47 grams of brown, resinous matter (B), which was also collected on a filter, freed from substances soluble in cold water, and dried. The final aqueous filtrate and washings (C), which amounted to about 10 litres, were then concentrated on a water-bath to about one-fourth this volume.

Examination of the Green Resin (A).

This resin, which was in the form of a green powder, was mixed with a considerable quantity of purified sawdust, and the mixture subjected to prolonged successive extraction in a large Soxhlet apparatus with the following solvents: petroleum (b. p. 35–50°), ether, chloroform, ethyl acetate, and alcohol.

Petroleum Extract of the Resin.

The material removed by petroleum was dark green, and weighed 41 grams. A portion of it was very sparingly soluble in this solvent. The entire petroleum extract was dissolved in 700 c.c. of warm ether, and the solution cooled, when about 3 grams of a solid substance separated. This was collected on a filter and washed with cold ether, after which it was distilled under a pressure of 15 mm. The distillate, which rapidly solidified, was crystallised from ethyl acetate, when small, glistening, colourless leaflets were obtained, which melted at 74.5°:

0.0872 gave 0.2717 CO₂ and 0.1148 H₂O. C = 85.0; H = 14.6.

C₃₅H₇₂ requires C = 85.4; H = 14.6 per cent.

This substance, therefore, was pentatriacontane.

The ethereal filtrate from the pentatriacontane was shaken with a solution of sodium hydroxide, when a dark green, flocculent product separated, which was collected on a filter and thoroughly washed with ether. The filtered, alkaline, aqueous liquid yielded nothing on acidification. For the examination of the green, flocculent product, it was warmed for some time with dilute aqueous sulphuric acid and chloroform, after which the chloroformic liquid was separated, washed, and evaporated to dryness. The small amount of residue was dissolved in ethyl acetate, and the solution heated with animal charcoal and filtered, when, on allowing the liquid to cool, small, colourless crystals were obtained. These melted quite indefinitely between 230° and

270°, and were found to be a mixture, for, when heated with acetic anhydride, they yielded diacetyloleanol, a substance which will subsequently be described, together with an acetyl derivative melting at 83·5°. The latter substance crystallised from acetic anhydride or ethyl acetate in tufts of small needles, but the amount obtained was not sufficient for analysis.

Isolation of a New Monocarboxylic Acid, $C_{22}H_{45}\cdot CO_2H$.

The ethereal liquid, which had been shaken with sodium hydroxide solution, was washed with water, when a large amount of chlorophyll was removed. This was examined, but nothing definite could be isolated from it. The ethereal liquid was then evaporated, and the residue heated with an alcoholic solution of an excess of potassium hydroxide for one hour in a reflux apparatus. After removing the alcohol, water was added, and the mixture extracted eleven times with ether. During the first extraction, a flocculent precipitate collected in the lower part of the ethereal layer. This was separated, washed, treated with sulphuric acid, and the product extracted with chloroform. On removing the chloroform, a wax-like residue was obtained, which, after treatment with animal charcoal, was crystallised from ethyl acetate, from which it separated in small, colourless, glistening leaflets melting at 68—69°:

0·1047 gave 0·2993 CO_2 and 0·1223 H_2O . $C = 78\cdot0$; $H = 13\cdot0$.

0·0959 „ 0·2738 CO_2 „ 0·1125 H_2O . $C = 77\cdot9$; $H = 13\cdot0$.

$C_{23}H_{46}O_2$ requires $C = 78\cdot0$; $H = 13\cdot0$ per cent.

This substance therefore appears to be a new *monocarboxylic acid*, as no acid possessing the formula $C_{23}H_{46}O_2$ has heretofore been described.

The combined ethereal extracts of the alkaline liquid were washed, dried, and concentrated to a small bulk, when a quantity of crystalline leaflets separated. These were removed, and, on further concentrating the solution, three more successive crops of crystals were obtained from it. The first and second portions of crystalline material, when dry, melted at 68°. They were therefore mixed and distilled in a vacuum, when a distillate was obtained which solidified and, when crystallised from ethyl acetate, separated in handsome, nacreous leaflets melting at 68—68·5°:

0·0862 gave 0·2697 CO_2 and 0·1147 H_2O . $C = 85\cdot3$; $H = 14\cdot8$.

$C_{31}H_{64}$ requires $C = 85\cdot3$; $H = 14\cdot7$ per cent.

The first crops of crystals therefore consisted of hentriacontane.

The third portion of crystalline material, when dissolved in chloro-

form, gave with acetic anhydride and sulphuric acid the colour reaction characteristic of the phytosterols, but it also contained hentriacontane, and no pure substance could be isolated from it.

Isolation of a New Crystalline Alcohol, Oleasterol, $C_{20}H_{33}\cdot OH$.

The last portion of crystals which separated from the above-described ethereal liquid was recrystallised several times from ethyl acetate when tufts of needles were obtained which melted at 172° . It was then crystallised twice from benzene, from which it finally separated in small tufts of fine needles melting at 174° :

0.0969 gave 0.2938 CO_2 and 0.1023 H_2O . $C = 82.7$; $H = 11.7$.

$C_{20}H_{34}O$ requires $C = 82.8$; $H = 11.7$ per cent.

This substance is evidently an alcohol related to the phytosterols, but is not identical with any compound previously described. It is therefore proposed to designate it *oleasterol*.

If a small quantity of oleasterol is dissolved in chloroform, a little acetic anhydride added, and, after first boiling the solution and then cooling it, a drop of concentrated sulphuric acid introduced, a slight yellow colour is first developed, which rapidly changes to pink. The tint then deepens to an intense magenta colour, which is accompanied by a brilliant yellowish-bronze fluorescence, subsequently changes to violet, and, after about one hour, the liquid becomes dull blackish-brown. If the reaction is conducted in acetic anhydride solution, no chloroform being used, the same colours are produced, but the changes are more rapid. When concentrated sulphuric acid is added to a solution of oleasterol in chloroform, the acid layer becomes yellow and, finally, of a deep orange colour. Solid oleasterol, when brought in contact with a drop of concentrated sulphuric acid, develops a bright orange and, finally, a vermillion-red colour, but again becomes colourless on the addition of water. Oleasterol is isomeric with several alcohols which possess properties similar to those of the phytosterols, namely, cinchol, cupreol, quebrachol, and rhamnol (compare Beilstein's *Handbuch*, 3rd edit., II., 1068, and *Proc. Amer. Pharm. Assoc.*, 1904, 52, 296), but melts at a higher temperature than any of these compounds, and differs from the last-named one, with which it has been directly compared, both in appearance and in the colour reactions yielded by it.

On removing the solvent from the original ethereal mother liquors from which the oleasterol had been separated, a residue was obtained which amounted to about 4 grams. This was distilled under 12 mm. pressure, when it passed over for the most part between 300° and 365° without decomposition, but a portion distilled above this temperature. The distilled product was dissolved in boiling ethyl acetate, when, on

cooling the solution, very hard, crystalline, wart-like masses separated, which melted at 200—204°. After several recrystallisations from ethyl acetate and, finally, from benzene, the melting point of the substance remained constant at 209—210°:

0.0864 gave 0.2560 CO₂ and 0.0896 H₂O. C = 80.8; H = 11.5.

C₂₇H₄₆O₂ requires C = 80.6; H = 11.4 per cent.

This substance was identical with a new compound, *homo-olestranol*, which was subsequently isolated in larger quantity from the ether extract of the resin.

The alkaline, aqueous liquid, from which the above-described alcohols and hydrocarbon had been removed by means of ether, was acidified with sulphuric acid and distilled with steam, but no volatile product was obtained. The acid liquid was therefore extracted with ether, the ethereal solution concentrated to a small bulk, and a considerable volume of light petroleum added. This caused the precipitation of the greater portion of the dissolved material in the form of an amorphous, dark green powder, from which nothing crystalline could be obtained. The clear petroleum liquid was decanted from the precipitate and evaporated, when a residue was obtained which was distilled under diminished pressure. The distilled product amounted to about 4 grams, and yielded a small quantity of a mixture of fatty acids melting at 46—49°, but the greater portion consisted of an oily liquid, which was unsaturated, and probably contained oleic acid.

Ethereal Extract of the Resin.

This extract amounted to 87 grams. As originally obtained, it consisted of a dark green, ethereal solution containing a quantity of an amorphous, green, sparingly soluble powder. The latter was collected on a filter and washed well with ether, after which it weighed 33 grams. On concentrating the ethereal filtrate to about 500 c.c., a further quantity (20 grams) of a similar amorphous powder was obtained.

Isolation of a New Crystalline Substance, Oleanol, C₃₁H₄₈O(OH)₂.

The above-mentioned portions of solid matter were separately examined, but were found to be identical. The material was dissolved in hot alcohol, and the solution decolorised by means of animal charcoal, after which it deposited a colourless substance in tufts of extremely fine, silky needles. This substance, when not quite pure, tends to separate in an amorphous condition, and, in these circumstances, can most readily be obtained crystalline by allowing its solution in boiling alcohol to evaporate. By successively concentrating the mother liquors from this crystalline substance, further quantities of it

were obtained, and also some portions of amorphous material which could not be caused to crystallise from any solvent. It was at first thought that these amorphous products might contain a second substance, but on acetylation they gave an almost quantitative yield of the diacetyl derivative of the crystalline substance, with only a little amorphous matter. As first obtained, the crystalline substance melted at about 301° , but on recrystallisation the melting point was raised to $303\text{--}304^{\circ}$, when it remained constant, no decomposition occurring. After drying at 110° , it was analysed:

0.1127 gave 0.3271 CO_2 and 0.1088 H_2O . $\text{C} = 79.1$; $\text{H} = 10.7$.

0.1135 „ 0.3291 CO_2 „ 0.1083 H_2O . $\text{C} = 79.1$; $\text{H} = 10.6$.

$\text{C}_{31}\text{H}_{50}\text{O}_3$ requires $\text{C} = 79.1$; $\text{H} = 10.6$ per cent.

No substance of the formula $\text{C}_{31}\text{H}_{50}\text{O}_3$ has previously been described, and it is therefore proposed to designate the new compound *oleanol*.

Oleanol is not very soluble in any of the usual organic solvents, but is moderately so in boiling chloroform, from which it separates in an amorphous condition. When crystallised from ordinary alcohol, it contains one molecule of water of crystallisation, as shown by the following analysis:

0.3466 of air-dried substance, on heating at 110° , lost 0.0130 H_2O .

$\text{H}_2\text{O} = 3.7$

0.1136 of air-dried substance gave 0.3152 CO_2 and 0.1107 H_2O .

$\text{C} = 75.7$; $\text{H} = 10.8$.

$\text{C}_{31}\text{H}_{50}\text{O}_3 \cdot \text{H}_2\text{O}$ requires $\text{H}_2\text{O} = 3.7$; $\text{C} = 76.2$; $\text{H} = 10.7$ per cent.

Two of the oxygen atoms contained in oleanol are, as shown below, present in the form of hydroxyl groups, and one of the latter possesses phenolic properties. In consequence of this fact, the substance reacts with sodium carbonate and hydroxide, giving an insoluble, amorphous *sodium* derivative. The third oxygen atom present in oleanol probably occurs in an ether linking, since the compound does not react with hydroxylamine. It was ascertained, however, by means of Perkin's modification of Zeisel's method, that no methoxyl or ethoxyl group was present. When oleanol is gently warmed with concentrated sulphuric acid, a deep orange colour is produced, and, when dissolved in chloroform, it does not decolorise a solution of bromine in the same solvent.

A determination of the specific rotatory power of oleanol gave the following result:

0.3032, dissolved in 25 c.c. of chloroform at 30° , gave $\alpha_D + 1^{\circ}54'$ in a 2-dm. tube, whence $[\alpha]_D + 78.3^{\circ}$.

Diacetyloleanol, $\text{C}_{31}\text{H}_{48}\text{O}_3(\text{CO} \cdot \text{CH}_3)_2$.—A quantity of oleanol was dissolved in acetic anhydride, and the solution boiled for some time.

It was then allowed to cool, when the acetyl derivative separated in colourless, glistening leaflets :

0.0994 gave 0.2749 CO_2 and 0.0881 H_2O . $\text{C} = 75.4$; $\text{H} = 9.8$.

$\text{C}_{31}\text{H}_{48}\text{O}_3(\text{CO}\cdot\text{CH}_3)_2$ requires $\text{C} = 75.8$; $\text{H} = 9.7$ per cent.

Diacetyloleanol is very readily soluble in chloroform, only moderately so in ethyl acetate, and nearly insoluble in cold alcohol. It is recrystallised most conveniently from acetic anhydride.

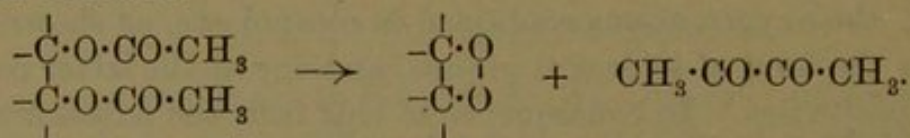
When diacetyloleanol is heated, fusion takes place at about 208° , the exact temperature depending on the rate of heating, gas is then evolved, and the liquid quickly re-solidifies. The solid so obtained is insoluble in alcohol, and only sparingly soluble in ethyl acetate or acetic anhydride, and is not acted on by the latter. It is very readily soluble in chloroform or benzene, but crystallises from a mixture of one part of the latter and two parts of petroleum (b. p. $90-120^\circ$) in small tufts of colourless needles, which do not melt at a temperature of 310° . This substance is unaffected by alkalis, and, when dissolved in chloroform, slowly decolorises a solution of bromine in the same solvent. On analysis, it yielded the following results :

0.1128 gave 0.3280 CO_2 and 0.1063 H_2O . $\text{C} = 79.3$; $\text{H} = 10.5$.

0.1049 „ 0.3058 CO_2 „ 0.0990 H_2O . $\text{C} = 79.5$; $\text{H} = 10.5$.

$\text{C}_{31}\text{H}_{48}\text{O}_3$ requires $\text{C} = 79.4$; $\text{H} = 10.3$ per cent.

This result indicates that, on heating diacetyloleanol, both the acetyl groups are removed, but, as no water is present during the operation, they are not eliminated by hydrolysis. The change might be represented as follows :



This view is in harmony with the fact that the high melting product contains no hydroxyl group, and the loss of weight suffered by the original acetyl derivative on being heated is also in agreement with it :

0.1648 of diacetyloleanol, on heating at 230° , lost 0.0254 $\text{CH}_3\cdot\text{CO}$.

$\text{CH}_3\cdot\text{CO} = 15.4$.

$\text{C}_{31}\text{H}_{48}\text{O}_3(\text{CO}\cdot\text{CH}_3)_2$ requires $\text{CH}_3\cdot\text{CO} = 15.5$ per cent.

Monoacetyloleanol, $\text{C}_{31}\text{H}_{48}\text{O}_2(\text{OH})\cdot\text{CO}\cdot\text{CH}_3$.—A quantity of diacetyloleanol was dissolved in 85 per cent. alcohol, and the solution boiled for forty-five minutes. On cooling the liquid, a product was obtained which crystallised in needles melting at 258° . This substance was much more soluble in alcohol than diacetyloleanol, and was evidently a *monoacetyloleanol*, since it readily yielded the diacetyl derivative on treatment with acetic anhydride. After drying at 110° , it was analysed :

0.1055 gave 0.2978 CO_2 and 0.0980 H_2O . $\text{C} = 77.0$; $\text{H} = 10.3$.

$\text{C}_{33}\text{H}_{52}\text{O}_4$ requires $\text{C} = 77.3$; $\text{H} = 10.2$ per cent.

Oleanol possesses some phenolic properties, as indicated by the fact that it yields a sodium derivative. The hydroxyl group contained in the above-described monoacetyloleanol is, however, devoid of such properties, since the compound does not react with cold alkalis.

Monomethyloleanol, $\text{C}_{31}\text{H}_{48}\text{O}(\text{OH})\cdot\text{O}\cdot\text{CH}_3$.—Five grams of oleanol were dissolved in absolute alcohol, and a slight excess of both sodium ethoxide and methyl iodide added. The mixture was then heated for one and a-half hours in a flask attached to a reflux condenser, after which the greater part of the solvent was removed. The addition of water then caused the precipitation of a *monomethyloleanol*, which crystallised from alcohol in long, thin, quite colourless needles. This compound, when dried in the air at the ordinary temperature or at 90° , melted at about 145° and then re-solidified, after which it fused at 194 — 195° . It also melted sharply at the latter temperature when dried at 120° . After drying at 125° , it gave on analysis the following results:

0.1325 gave 0.3828 CO_2 and 0.1290 H_2O . $\text{C} = 78.8$; $\text{H} = 10.8$.

0.3144 gave, by Perkin's modification of Zeisel's method, 0.1500 AgI . $\text{O}\cdot\text{CH}_3 = 6.3$.

$\text{C}_{32}\text{H}_{52}\text{O}_3$ requires $\text{C} = 79.3$; $\text{H} = 10.7$; $\text{O}\cdot\text{CH}_3 = 6.4$ per cent.

The air-dried preparation was then analysed:

0.0977 gave 0.2788 CO_2 and 0.0960 H_2O . $\text{C} = 77.8$; $\text{H} = 10.9$.

0.3169, on heating at 125° , lost 0.0139 $\text{C}_2\text{H}_5\cdot\text{OH}$. $\text{C}_2\text{H}_5\cdot\text{OH} = 4.4$.
 $\text{C}_{31}\text{H}_{48}\text{O}(\text{OH})\cdot\text{O}\cdot\text{CH}_3, \frac{1}{2}\text{C}_2\text{H}_5\cdot\text{OH}$ requires $\text{C} = 78.1$; $\text{H} = 10.8$;
 $\text{C}_2\text{H}_5\cdot\text{OH} = 4.5$ per cent.

Acetylmethyloleanol, $\text{C}_{31}\text{H}_{48}\text{O}_2(\text{O}\cdot\text{CH}_3)\cdot\text{CO}\cdot\text{CH}_3$.—Monomethyloleanol was dissolved in acetic anhydride, and the mixture boiled for half an hour. The solution was then allowed to cool, when *acetylmethyloleanol* separated in long, highly lustrous leaflets, which melted at 215.5° :

0.1041 gave 0.2960 CO_2 and 0.0978 H_2O . $\text{C} = 77.5$; $\text{H} = 10.4$.

$\text{C}_{34}\text{H}_{54}\text{O}_4$ requires $\text{C} = 77.6$; $\text{H} = 10.3$ per cent.

0.2132, on hydrolysis, neutralised 4.4 c.c. $N/10$ NaOH .

$\text{C}_{31}\text{H}_{48}\text{O}_2(\text{O}\cdot\text{CH}_3)\cdot\text{CO}\cdot\text{CH}_3$ requires 4.1 c.c.

Acetylmethyloleanol can be recrystallised from ordinary alcohol, but, when boiled for some time with this solvent, the acetyl group is eliminated and monomethyloleanol is regenerated.

The original ethereal filtrate from the crude oleanol was shaken with a solution of ammonium carbonate, but this treatment removed only small amounts of amorphous products. It was then shaken with several successive portions of a solution of sodium carbonate, when a

considerable quantity of a green precipitate was formed. This precipitate was separated from the ethereal liquid and from the alkaline, aqueous solution, when, on examination, it was found to consist almost entirely of sodium oleanol, together with chlorophyll. The alkaline, aqueous liquid was acidified, but it yielded only a small amount of tarry matter. The ethereal liquid, when free from substances which reacted with sodium carbonate, was shaken with a solution of sodium hydroxide, but this dissolved nothing. It was then washed with water, when a large amount of chlorophyll was removed, dried, and the greater part of the ether evaporated.

Isolation of Two New Crystalline Alcohols.

Olestranol, $C_{25}H_{42}O_2$, and Homo-olestranol, $C_{27}H_{46}O_2$.

On allowing the concentrated ethereal liquid, obtained as above described, to stand, it deposited about 3 grams of a white solid. This was dissolved in hot ethyl acetate, from which, on cooling, it separated in hard, wart-like masses, which melted at 217° . After recrystallisation from benzene, it melted at $217-218^{\circ}$ and was analysed:

0.0998 gave 0.2928 CO_2 and 0.1009 H_2O . $C = 80.0$; $H = 11.2$.

0.0813 „ 0.2389 CO_2 „ 0.0845 H_2O . $C = 80.1$; $H = 11.4$.

$C_{25}H_{42}O_2$ requires $C = 80.2$; $H = 11.2$ per cent.

This substance, so far as can be ascertained, is not identical with any compound previously described. It is proposed therefore to designate it *olestranol*, since it is an alcohol.

Olestranyl acetate and *benzoate* were prepared, but were found to be uncrystallisable syrups. The latter compound was very viscid, and dissolved readily in ethyl acetate and in chloroform, but was almost insoluble in alcohol. When dissolved in chloroform and treated with a solution of bromine in the same solvent, olestranol readily gives a *bromide*, which is precipitated as a viscid oil. This oil did not become crystalline on standing, and when warmed with any solvent, in order to bring it into solution, it evolved hydrogen bromide, yielding a product which was readily soluble in chloroform, but uncrystallisable. Olestranol exhibits colour reactions which, so far as could be observed, are indistinguishable from those shown by oleasterol (p. 895). Its formula, $C_{25}H_{42}O_2$, indicates it to be a lower homologue of a hydroxyphytosterol, and its properties are in accordance with this view, for, although some of the colours which are produced on treating it with acetic anhydride and sulphuric acid differ from those given under similar conditions by the phytosterols, the general character of the colour reactions is analogous to that yielded by the latter compounds.

The original ethereal filtrate from the olestranol was green, owing to the presence of chlorophyll. It was therefore boiled with an alcoholic solution of potassium hydroxide, the greater part of the solvent removed, water added, and the mixture extracted six times with ether. On removing the solvent, several grams of a light-coloured product were obtained. This was repeatedly crystallised from ethyl acetate, from which it separated in hard, wart-like masses, identical in appearance with the nodular masses of olestranol obtained under similar conditions. This preparation, however, melted at a temperature lower than that at which olestranol fuses, namely, at 210° , and repeated crystallisations from ethyl acetate and from benzene failed to raise the melting point of the material :

0.1120 gave 0.3314 CO_2 and 0.1152 H_2O . $\text{C} = 80.7$; $\text{H} = 11.4$.

$\text{C}_{27}\text{H}_{46}\text{O}_2$ requires $\text{C} = 80.6$; $\text{H} = 11.4$ per cent.

This compound, melting at 210° , is therefore identical with the substance possessing the same formula and melting point which was isolated in smaller quantity from the petroleum extract of the resin. Its formula, $\text{C}_{27}\text{H}_{46}\text{O}_2$, is that of a hydroxyphytosterol, and, since it is not identical with any compound previously described and is apparently a homologue of olestranol, differing from the latter by the increment of the elements C_2H_4 , the name *homo-olestranol* may appropriately be assigned to it.

One compound, possessing properties similar to those of homo-olestranol, has previously been isolated from the seeds of *Casimiroa edulis*, La Llave (Fam. *Rutaceae*), by W. Bickern (*Arch. Pharm.*, 1903, 241, 173). This substance, which has been designated casimirol, has been assigned the formula $\text{C}_{27}\text{H}_{48}\text{O}_2$, and is stated to melt at 207° . It was therefore at first thought that these two substances might be identical, but a comparison of their respective colour reactions rendered it evident that this was not the case.

Homo-olestranol exhibits colour reactions which, like those of olestranol, are identical with those yielded by oleasterol. It is apparently somewhat more readily soluble in ether than its lower homologue, and, as seen from the fact that it was also obtained from the previously-described extract of the resin, it is very slightly soluble in petroleum, whilst olestranol appears to be insoluble in this solvent. With the exception of these slight differences, the two homologous hydroxyphytosterols could only be distinguished by their melting points or by analysis, as their behaviour on acetylation, benzylation, and bromination is identical. Homo-olestranol does not yield the colour reaction characteristic of the so-called "oxycholesterin" of Lifschütz (*Ber.*, 1908, 41, 252), which, however, does not appear to have been analysed.

A determination of the specific rotatory power of homo-olestranol yielded the following result:

0.7216, dissolved in 25 c.c. of chloroform, gave $\alpha_D + 4^{\circ}6'$ in a 2-dcm. tube, whence $[\alpha]_D + 71.0^{\circ}$.

The amount of olestranol available did not permit of a similar determination being made with that compound.

Chloroform, Ethyl Acetate, and Alcohol Extracts of the Resin.

The chloroform extract of the resin amounted to 9 grams. It was exhaustively examined, but nothing crystalline could be obtained from it.

The portion of the resin extracted by ethyl acetate amounted to only 2 grams. It consisted of tarry matter, together with a trace of *d*-mannitol, which was subsequently isolated in larger amount from the aqueous liquid.

The remainder of the resin, which was soluble only in alcohol, formed a brown powder, and amounted to 23 grams. Nothing crystalline could be separated from it, and, in view of the possibility of its containing some glucosidic material, it was freed from any traces of sugar and submitted to the action of sulphuric acid in dilute alcohol, but was unaltered by this treatment.

Examination of the Brown Resin (B).

This product, obtained as previously described, amounted to 47 grams. It was mixed with purified sawdust, and extracted with the same solvents as were employed in the case of the green resin. The petroleum and ethereal extracts thus obtained were small in amount, and consisted chiefly of the material which formed the corresponding extracts from the resin previously examined. The remaining extracts, of which that obtained by ethyl acetate was the largest, were all brown, resinous products, from which nothing crystalline could be isolated.

Examination of the Aqueous Liquid (C).

After concentrating this liquid, it was extracted many times with ether, but this removed only a small amount of amorphous material, containing but a trace of crystalline substance. An excess of a solution of basic lead acetate was then added, when a bulky, yellow precipitate was produced, which was removed by filtration and thoroughly washed with boiling water. This precipitate was suspended in water, decomposed by hydrogen sulphide, and the mixture filtered. The filtrate was concentrated and allowed to cool, when a quantity of brown, viscid, resinous matter separated. The clear liquid was then ex-

tracted with amyl alcohol, when a further quantity of brown, viscid material was removed. These resinous products possessed a bitter, astringent taste, and, when dissolved in boiling water, readily reduced Fehling's solution, but nothing crystalline could be obtained from them. The brown, aqueous liquid which had been extracted with amyl alcohol gave a deep black colour with ferric chloride, and evidently contained a considerable amount of tannin. When treated with a solution of barium hydroxide, it gave a yellow precipitate, which rapidly darkened in the air, but nothing could be isolated from it.

The original aqueous filtrate and washings from the precipitate produced by basic lead acetate were freed from a further small quantity of brown material by a second treatment with this reagent then deprived of lead by means of hydrogen sulphide, again filtered, and concentrated under diminished pressure. On allowing the liquid to stand, it deposited a large amount (60 grams) of needle-shaped crystals, which, after recrystallisation from dilute alcohol, melted at 165° :

0.1183 gave 0.1717 CO_2 and 0.0830 H_2O . $\text{C} = 39.6$; $\text{H} = 7.8$.

$\text{C}_6\text{H}_{14}\text{O}_6$ requires $\text{C} = 39.6$; $\text{H} = 7.7$ per cent.

This substance was evidently *d*-mannitol, since it yielded a dextro-rotatory hexa-acetyl derivative, which melted at 122 — 123° .

The filtrate from the crude mannitol deposited further quantities of the latter on concentration, but no other solid substance could be separated from it. It contained a considerable amount of sugar, for, when warmed with a solution of phenylhydrazine acetate, it readily yielded *d*-phenylglucosazone, which, when heated somewhat rapidly, melted at 217° . In view of the possibility of this final aqueous liquid also containing a glucoside, it was heated for some time with dilute sulphuric acid, but this treatment resulted only in the production of a quantity of resin, together with a little furfural.

Summary.

The results of this investigation may be summarised as follows:

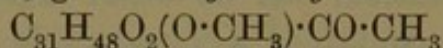
The air-dried olive leaves employed, on percolation with boiling alcohol, yielded about 30 per cent. of their weight of extracted material, of which nearly 63 per cent. was soluble in cold water. A further 8.7 per cent. could be dissolved by boiling water, whilst the remainder, amounting to 28.3 per cent., was quite insoluble in this solvent. The alcoholic extract, when distilled with steam, yielded a trace of essential oil.

The portion of the alcoholic extract which was soluble in cold water contained a large quantity of a sugar which yielded *d*-phenylglucosazone

(m. p. 217°), together with tannic matter, brown, amorphous products, and an amount of *d*-mannitol equivalent to about 3.4 per cent. of the weight of the air-dried leaves.

The portion of the alcoholic extract which was dissolved by boiling water, but separated on cooling, consisted of amorphous resins.

The material insoluble in water contained, together with chlorophyll and some resinous matter, small amounts of the hydrocarbons hentriacontane, $C_{31}H_{64}$ (m. p. $68-69^{\circ}$), and pentatriacontane, $C_{35}H_{72}$ (m. p. 74.5°), a little of a mixture of fatty acids, and a new *monocarboxylic acid*, $C_{22}H_{45}\cdot CO_2H$ (m. p. $68-69^{\circ}$). In addition to these, it yielded three new crystalline alcohols, which appear to be related to the phytosterols, namely, *oleasterol*, $C_{20}H_{34}O$ (m. p. 174°), which crystallises in tufts of colourless needles; *olestranol*, $C_{25}H_{42}O_2$ (m. p. $217-218^{\circ}$), and *homo-olestranol*, $C_{27}H_{46}O_2$ (m. p. 210° ; $[\alpha]_D + 71^{\circ}$). The two last-mentioned alcohols crystallise in hard, wart-like masses, and appear to be *hydroxyphytosterols*. Furthermore, a new colourless, crystalline substance, *oleanol*, $C_{31}H_{48}O(OH)_2\cdot H_2O$ m. p. $303-304^{\circ}$; $[\alpha]_D^{30} + 78.3^{\circ}$, was obtained in an amount equivalent to nearly 3.4 per cent. of the weight of the air-dried leaves. Oleanol contains two hydroxyl groups, one of which possesses phenolic properties. When treated with sodium ethoxide and methyl iodide, it yields a *monomethyloleanol*, $C_{31}H_{48}O(OH)\cdot O\cdot CH_3$ (m. p. $194-195^{\circ}$), and this, on acetylation, gives *acetylmethyloleanol*,



(m. p. 215.5°). *Diacetyloleanol*, $C_{31}H_{48}O_3(CO\cdot CH_3)_2$, fuses at about 208° , and suffers a change which is accompanied by the evolution of gas and the formation of a crystalline substance, $C_{31}H_{48}O_3$, which does not melt at 310° . *Monoacetyloleanol*, $C_{31}H_{48}O_2(OH)\cdot CO\cdot CH_3$ (m. p. 258°), was prepared by heating the diacetyl derivative with aqueous alcohol.

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