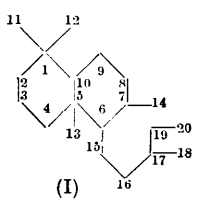
519. *The Constitution of Marrubiin. Part I.**

By Wesley Cocker, B. E. Cross, S. R. Duff, J. T. Edward, and T. F. Holley.

Spectroscopic and oxidative studies of marrubiin, $C_{20}H_{28}O_4$, indicate that it is a furan derivative, having the structure (III).

Marrubiin, a bitter principle of horehound (*Marrubium vulgare* L), is extracted from the herb with acetone in yields of up to 1%, but different samples of the herb produce widely differing amounts, and some samples we examined yielded none. Season of harvesting, conditions of drying, and length of storage have a profound influence upon the yield, and it has been suggested (Rizzocath, Thesis, Univ. Aix-Marseille, 1933) that marrubiin is an artefact arising from an essential oil during the drying of the horehound. It is, of course, important that the herb should be of the correct botanical species. From time to time, *Ballota hirsuta* Bentham has been sold as horehound; this contains little or no marrubiin, which can however be isolated in small yield from *B. foetida* (Balansard, *Compt. rend. Soc. Biol.*, 1934, **117**, 1014). In the present investigation, horehound of Morroccan origin was procured and kindly extracted by Messrs. C. W. Field, Ltd., of Liverpool, to whom we are extremely indebted for valuable co-operation and assistance.

Although earlier workers were engaged on the study of marrubiin, the most significant advances in its chemistry were made by Lawson and Eustice (*J.*, 1939, 587), Holliss, Richards, and Robertson (*Nature*, 1939, **143**, 604), and more recently by Ghigi (*Gazzetta*, 1948, **78**, 865; 1951, **81**, 336). Before Ghigi's publications, it had been shown that marrubiin exhibits the following characteristics and reactions. (*a*) It is a diunsaturated lactone, hydrolysed by alkali to a diunsaturated hydroxy-acid, marrubic acid: reduction of marrubiin and marrubic acid gives the corresponding tetrahydro-compounds. (*b*) It possesses a hydroxyl group incapable of acetylation, but readily removed by dehydrating agents, thus yielding the triunsaturated compound which we now term anhydromarrubiin. For this reason, the hydroxyl is considered to be tertiary. (*c*) The fourth oxygen atom is inert, and probably ethereal. (*d*) On dehydrogenation, marrubiin yields 1 : 2 : 5-trimethylnaphthalene; on the basis of this evidence, and by analogy with the diterpenoid alcohols, Lawson and Eustice (*loc. cit.*) proposed the skeletal structure (I), whilst Holliss, Richards, and Robertson (*loc. cit.*) suggested that marrubiin is a hydroxy-diterpene of the manoyl oxide type. However, marrubiin and its hydro-derivatives differ from manoyl oxide and the closely related sclareol in their inability to form tricyclic derivatives, which can be dehydrogenated to 1 : 2 : 8-trimethylphenanthrene, on treatment with dehydrating agents (cf. Lawson and Eustice, *loc. cit.*; Ruzicka and Janot, *Helv. Chim. Acta*, 1931, **14**, 203; Hosking and Brandt, *Ber.*, 1935, **68**, 37).



(I)

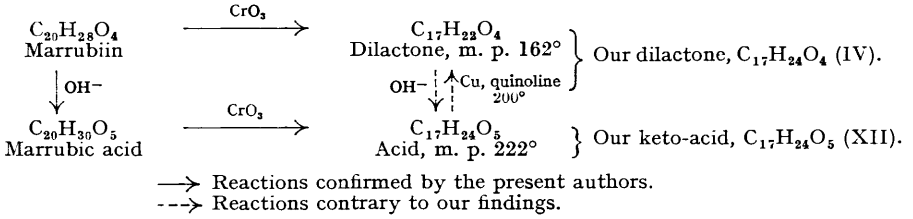
Our investigations, begun in 1949, were well under way when our attention was drawn to the earlier paper by Ghigi (*loc. cit.*). Unfortunately, after some months, a shortage of marrubiin brought our investigations to a standstill until late in 1951. Ghigi's later paper

* Cf. Cocker, Cross, Duff, and Holley, *Chem. and Ind.*, 1952, 827.

contributes little further to the problem, although a number of new derivatives of marrubiin are mentioned. In the earlier paper, she claimed that on oxidation with chromic acid in acetic acid, marrubiin loses three carbon atoms, two double bonds, and the inert oxygen, and yields a lactone, $C_{17}H_{22}O_4$, which can be hydrolysed to an acid, $C_{17}H_{24}O_5$. She claimed further that the latter compound is obtained when marrubic acid is oxidised with either chromic acid or alkaline permanganate (Scheme A). We do not agree with all these conclusions, but we are in agreement that C_{17} compounds are produced in the oxidations and that these compounds yield 1 : 2 : 5-trimethylnaphthalene on dehydrogenation, thus establishing the presence of a bicyclic system in them.

We must now consider what side chain attached to the bicyclic system can lose three carbon atoms, two double bonds, and an inert oxygen atom on oxidation. The light-absorption characteristics and colour reactions of marrubiin strongly suggest that the side chain is a furan, and this would conform with the oxidative evidence.

(A) *Scheme of Ghigi's oxidation experiments.*



⟶ Reactions confirmed by the present authors.
⇢ Reactions contrary to our findings.

In the ultra-violet region, marrubiin shows maxima at 2080, 2120, and 2160 Å (log ε 3·75, 3·75, and 3·70 respectively), in substantial agreement with those expected of a substituted furan (cf. Price and Walsh, *Proc. Roy. Soc.*, 1941, *A*, **179**, 201, who give 1996, 2046, and 2110 Å for furan).

The infra-red spectra of marrubiin and furfuryl alcohol and the published data for furan are :

Marrubiin : 753, 768, 815, 874, 985, 1010, 1045, 1066, 1100, 1155, 1200, 1240, 1270, 1370, 1470, 1505, 1560, 1600, 1665, (1740, 1760, 1765),* 2880, 3440 † cm.$^{-1}$ (* lactonic C=O stretching frequency; † OH stretching frequency).

Furan : 740, 864, 990, 1052, 1176, 1249, 1375, 1495 cm.$^{-1}$ (measurements in liquid phase; Pickett, *J. Chem. Phys.*, 1942, **10**, 660); 605, 744, 837, 872, 994, 1067, 1180, 1270, 1340, 1381, 1486, 1579, 1596 cm.$^{-1}$ (measurement in vapour phase; Thompson and Temple, *Trans. Faraday Soc.*, 1945, **41**, 27).

Furfuryl alcohol : 750, 815, 887, 915, 1010, 1055, 1080, 1148, 1190, 1220, 1260, 1360, 1380, 1510, 1565, 1600, 1680, 2860, 3300 * cm.$^{-1}$ (* OH stretching frequency).

The spectrum of marrubiin is very similar to that of furan and furfuryl alcohol. The absence of the strong " aromatic " absorption in the 750, 1150, and 1470 cm.$^{-1}$ region in tetrahydromarrubic acid (and in tetrahydrofurfuryl alcohol) lends support to the postulate of a furan ring in marrubiin.

Examination of the lactonic C=O stretching frequency of marrubiin shows the presence of a butanolide :

Marrubiin : 1740 cm.$^{-1}$ in paraffin paste; 1760 cm.$^{-1}$ in $CHCl_3$; 1765 cm.$^{-1}$ in CS_2.
Anhydromarrubiin : 1760 cm.$^{-1}$ in $CHCl_3$.

Grove and Willis (*J.*, 1951, 877) have shown that butanolides display lactonic C=O stretching frequencies of 1770 cm.$^{-1}$, whilst pentanolides show a corresponding band at 1740 cm.$^{-1}$. It has been found, however, that suspensions in paraffin paste of compounds in which hydrogen bonding is possible, frequently exhibit lower C=O stretching frequencies than are exhibited by solutions of such compounds. We are grateful to Mr. Grove for this information.

Tetrahydromarrubic acid shows strong bands at 3470 (OH) and at 1688 cm.$^{-1}$ (CO_2H).

The chemical properties of marrubiin are in harmony with its butanolide structure. Thus, marrubiin is unreactive towards ammonia, aniline, and phenylhydrazine, and

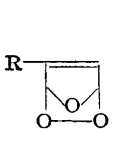
marrubic acid is lactonised by diazomethane. On the other hand, marrubic acid can be acetylated, and must be heated quite strongly before it lactonises, although on long boiling in xylene lactonisation takes place.

So far, we have been unable to obtain much direct chemical evidence in favour of the furan structure. Thus, we have been unable to obtain an adduct with maleic anhydride, nor is the ring hydrogenolysed over copper chromite. Marrubiin, however, smoothly absorbs two mols. of hydrogen over palladised charcoal, but relatively slowly. Hydrogenation over Adams's catalyst is faster, and the rate is in agreement with that expected of a furan (cf. Lawson and Eustice, *loc. cit.*; Jones and Taylor, *Quart. Reviews*, 1950, **4**, 198).

Marrubiin reacts with two mols. of perbenzoic acid, giving a compound which is probably of type (II) (cf. Jones and Taylor, *loc. cit.*), but so far it has proved intractable.

Marrubiin gives a green colour with a mixture of acetic anhydride and sulphuric acid in chloroform, as do furans (Levine and Richman, *Proc. Soc. Exp. Biol., N.Y.*, 1934, **31**, 582), but it fails to give the Shear colour reaction (Levine and Seaman, *Biochem. J.*, 1933, **27**, 2047). Ghigi (1951) claims that a pure coupling product is obtained from the diterpene and *p*-nitrobenzenediazonium chloride. Undoubtedly, a coloured product is obtained but, on an alumina column, we found that it separated into four coloured bands, none of which gave a crystalline product.

When the furan ring is removed by oxidation with chromic acid, we find that a dilactone, $C_{17}H_{24}O_4$, m. p. 160°, $[\alpha]_D$ +30°, is obtained. Evidently the carboxyl group left from the oxidation of the furan ring undergoes ready lactonisation with the tertiary hydroxyl group of marrubiin, which accordingly must be at the γ-position with respect to the point of attachment of the furan ring, as in (III), and the formula of the dilactone must be (IV).



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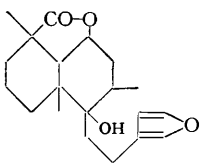

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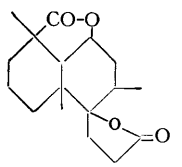

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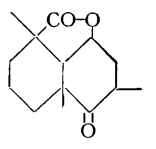


(V)

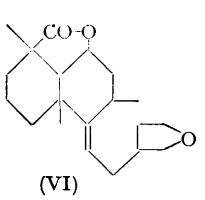
[To economise space, we shall develop our argument by reference to the structure (III) established in the sequel for marrubiin, rather than employ partial formulæ. Our argument involves the reasonable assumption that the skeletal structure of marrubiin is (I), but is otherwise rigorous.] Ghigi obtained by this oxidation a lactone, m. p. 162°, which is almost certainly identical with ours, although she ascribed to it the formula, $C_{17}H_{22}O_4$, and did not recognise it as a dilactone.

Further evidence for the location of the tertiary hydroxyl at $C_{(6)}$ is given by the results of ozonolysis of anhydrotetrahydromarrubiin (Lawson and Eustice, *loc. cit.*) which affords a keto-lactone, $C_{14}H_{20}O_3$, to which we ascribe the formula (V); in this reaction the C_6 side chain, including the tetrahydrofuran ring is removed. The keto-lactone exhibits maximum absorption at 2900 Å (log ε 1·44), and has, amongst others, a band at 1706 cm.$^{-1}$ (cf. Grove and Willis, *loc. cit.*; Fieser and Fieser, "Natural Products Related to Phenanthrene," Reinhold, New York, 1949, p. 203), both indicative of a ketonic group; a band at 1760 cm.$^{-1}$ indicates the presence of a butanolide, namely, that present in marrubiin. The keto-lactone gives an oxime, but fails to give a semicarbazone (cf. Ruegg, Dreiding, Jeger, and Ruzicka, *Helv. Chim. Acta*, 1950, **33**, 889), and is not reduced either with hydrogen and Adams's catalyst or under Clemmensen conditions, indicative of a hindered carbonyl group. The keto-lactone is identical with the unidentified product obtained by Ghigi from the oxidation of anhydromarrubiin with chromic acid. Its formation by ozonolysis indicates that anhydrotetrahydromarrubiin must be (VI), the removal of the tertiary OH at $C_{(6)}$ giving rise to an exocyclic double bond.

The keto-hydroxy-acid afforded by hydrolysis of the lactone ring is stable in boiling water or alcohol, and so the compound cannot be a β-keto-acid. Consequently, of the three possible positions, 1, 5, and 7, for the attachment of the carboxyl group to the perhydronaphthalene nucleus, the last two must be excluded, and so the carboxyl group

of this compound, and of marrubic acid, must be at $C_{(1)}$. A second reason for believing the carboxyl group to be at this position is that otherwise a *gem*-dimethyl group would be found at $C_{(1)}$. This is most unlikely in view of the ready aromatisation to naphthalene derivatives (cf. Clemo and Dickenson, *J.*, 1935, 735; 1937, 255; Cocker, Cross, Fateen, Lipman, Stuart, Thompson, and Whyte, *J.*, 1950, 1781). The formation of 1 : 2 : 5-trimethylnaphthalene could be explained only by rather improbable migrations of alkyl groups from the *gem*-position. On the other hand, presence of the carboxyl group at $C_{(1)}$ should lead to ready aromatisation and loss of carbon dioxide.

Further evidence against the location of the carboxyl at $C_{(7)}$ (though not necessarily at $C_{(5)}$) is given by facts pointing to its attachment to a tertiary carbon atom. Thus, (*a*) heating marrubic acid with concentrated sulphuric acid yields carbon monoxide (cf. Fieser and Fieser, *op. cit.*, p. 52); and (*b*) marrubic acid resists esterification, so that diazomethane lactonises it, even in cold ether, to marrubiin.



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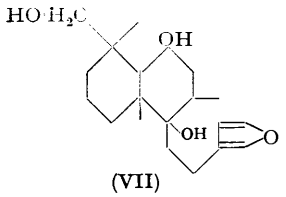

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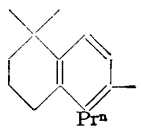


(VIII)

We have attempted to confirm the position of the carboxyl group by the methods applied by Ruzicka and Meyer (*Helv. Chim. Acta*, 1922, **5**, 581) to the resin acids. Reduction of marrubiin or marrubic acid with lithium aluminium hydride yields the triol, marrubenol, $C_{20}H_{32}O_4$ (VII), which is reduced catalytically to a tetrahydro-compound, marrubanol, $C_{20}H_{36}O_4$. This compound can also be obtained by the reduction of tetrahydromarrubiin. Dehydrogenation of marrubenol yields, with difficulty, a small amount of 1 : 2 : 5-trimethylnaphthalene. Attempts to rearrange and dehydrate marrubanol with anhydrous formic acid, polyphosphoric acid, or hydrochloric acid in acetic acid gave intractable gums. Dehydrogenation of these products with sulphur, selenium, or palladised charcoal failed to give the expected 5-ethyl-1 : 2-dimethylnaphthalene. Instead a benzenoid hydrocarbon, probably (VIII), was obtained.

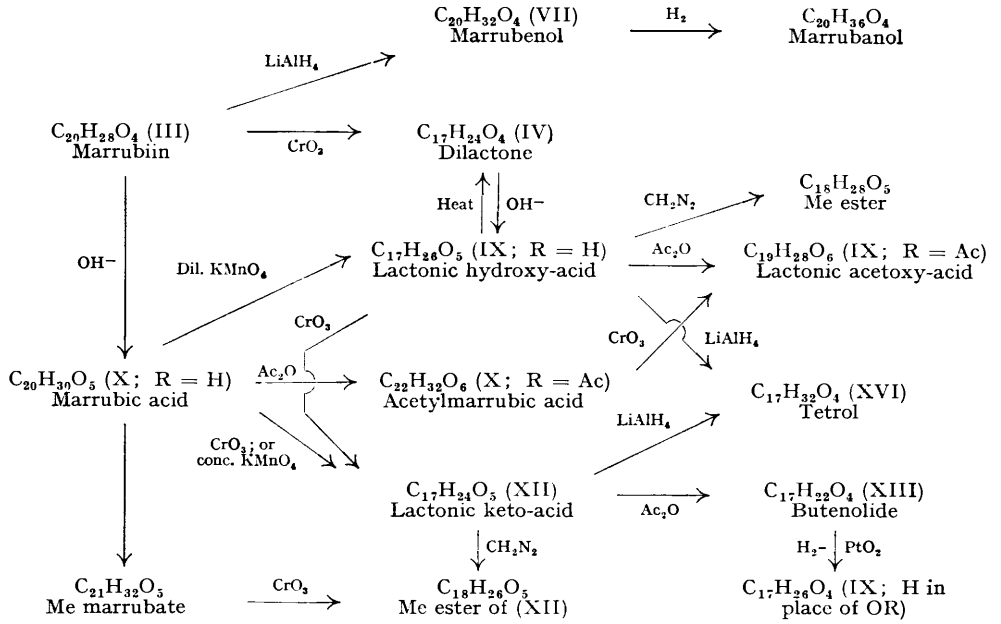
When the dilactone (IV), $C_{17}H_{24}O_4$, is hydrolysed, one lactone ring is opened and a lactonic hydroxy-acid, $C_{17}H_{26}O_5$, m. p. 205°, $[\alpha]_D^{16}$ −15°, showing the characteristic absorption by hydroxyl at 3300 cm.$^{-1}$, is obtained. This acid neutralises one equivalent of alkali in the cold, but by hydrolytic titration consumes two equivalents, showing it to be a lactonic acid. Ghigi claimed that hydrolysis of her lactone, $C_{17}H_{22}O_4$, gave an acid, $C_{17}H_{24}O_5$, m. p. 222°, also afforded by the oxidation of marrubic acid with chromic acid. As we show below, this oxidation yields a lactonic keto-acid, m. p. 220°, $[\alpha]_D^{23}$ +88·7°. The melting point of the lactonic hydroxy-acid is markedly influenced by rate of heating, and we have obtained melting points above 210° by rapid heating. It is possible that the closeness of the melting points and analytical values led Ghigi to confuse these two acids.

The lactone ring opened by the hydrolysis is the one originally present in marrubiin, so the acid must have the structure (IX; R = H). This follows from the facts, (*a*) that it also arises in low yield from the oxidation of marrubic acid (X; R = H) with dilute permanganate, and (*b*) that its acetyl derivative (IX; R = Ac) is obtained on oxidation of acetylmarrubic acid, which must have formula (X; R = Ac) since the tertiary hydroxyl is incapable of being acetylated (Lawson and Eustice, *loc. cit.*). Obviously in both of these reactions a dicarboxylic acid intermediate (XI; R = H and Ac) is involved, which undergoes ready lactonisation between the 6-hydroxyl and the 17-carboxyl group noted previously. When the hydroxy-acid (IX; R = H) is heated with acetic anhydride, formation of the second lactone ring proceeds, yielding the dilactone (IV).

When marrubic acid is oxidised with chromic acid, or with concentrated permanganate solution, or when the lactonic hydroxy-acid (IX; R = H) is oxidised with chromic acid,

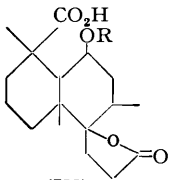
the lactonic keto-acid, $C_{17}H_{24}O_5$ (XII), m. p. 220°, $[\alpha]_D$ +88·7°, is formed. Ghigi obtained an uncharacterised acid, $C_{17}H_{24}O_5$, from the oxidation of marrubic acid. The presence in the acid of a six-membered ring ketone is shown by an absorption band at 1736 cm.$^{-1}$; the butanolide C=O gives rise to a band at 1758 cm.$^{-1}$, and the carboxyl C=O to one at 1672 cm.$^{-1}$; a strong band at 1460 cm.$^{-1}$ may be due to the carboxylate ion (cf. Thompson, *J.*,

(B) *Our oxidation experiments.*



1948, 328). In the ultra-violet the acid exhibits a maximum at 2580 Å (log ε, 1·69); this is an unusually short wave-length and high intensity for an isolated keto-group, but no other reasonable absorbing system can be ascribed to it; possibly the neighbouring carboxyl group, rigidly anchored close to it, has a profound effect on the electronic mobility. That the keto-group is strongly hindered is shown by the formation of an oxime, but not a semicarbazone.

That the carboxyl group of (XII) is the original carboxyl group of marrubic acid is shown by the fact that methyl marrubate affords on oxidation a methyl ester identical with that formed by esterification of (XII).



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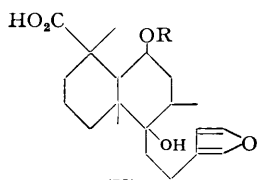

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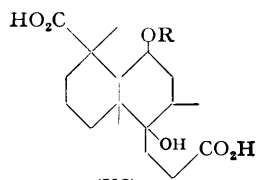
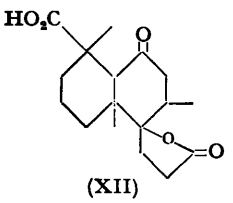


(XI)

Cyclisation of (XII) with acetic anhydride affords a butenolide (XIII), $C_{17}H_{22}O_4$, m. p. 148—150°, $[\alpha]_D^{17}$ −132°. Ghigi, however, claimed that the product of oxidation of marrubic acid with chromic acid is cyclised to a lactone identical with that obtained from the oxidation of marrubiin. The latter product is, in fact, the dilactone (IV), completely different from the butenolide (see Scheme B). That our cyclisation product is a βγ-butenolide is shown by its properties: it rapidly decolorises bromine and permanganate solutions; its solution in warm water has pH 3—4 and after 10 hours' boiling in water the lactone and keto-acid are present in solution in the ratio 2 : 1; it is completely

hydrolysed by boiling 2% aqueous methanolic hydrochloric acid; it gives a keto-anilide when kept in aniline (cf. Kuehl, Linstead, and Orkin, *J.*, 1950, 2213); and it is reduced over Adams's catalyst to a saturated acid, $C_{17}H_{26}O_4$, to which we assign structure (IX; H in place of OR). The last reaction, involving hydrogenolysis of the lactone, is characteristic of βγ-butenolides (cf. Jacobs and Scott, *J. Biol. Chem.*, 1930, **87**, 601; 1931, **93**, 139). The βγ-butenolide shows no tendency to isomerise to the αβ-form, evidence supporting the thesis that the lactonic CO group is attached to a tertiary carbon atom as in (XIII).

The proof that the βγ-butenolide, $C_{17}H_{24}O_5$, has the structure (XIII) is clearly fundamental in deciding the formula for marrubiin. A butanolide of carbon skeleton (I) and having the lactone C=O attached at $C_{(1)}$, can have its other oxygen atom only at $C_{(3)}$ or $C_{(9)}$ as in (III) and (XIV) respectively. Attachment to $C_{(3)}$, resulting in the structure (XIV) for marrubiin, is sterically possible; but it would give rise to the sterically impossible structure (XV) for the butenolide mentioned above. The latter must consequently have the structure (XIII),* and the corresponding keto-acid the structure (XII). Furthermore, it can readily be seen that a pentanolide structure for marrubiin, which would locate the oxygen of the lactone ring at $C_{(4)}$ or $C_{(8)}$, is impossible because the formation of a pentenolide from the keto-acid, $C_{17}H_{24}O_5$, would be stereochemically impossible.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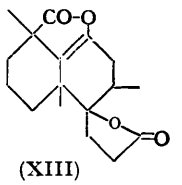
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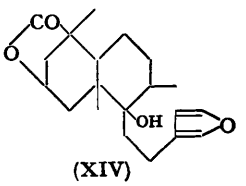
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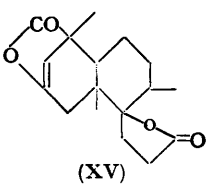
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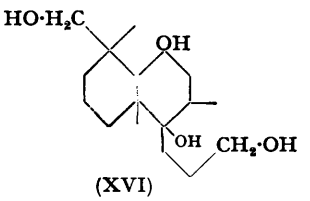
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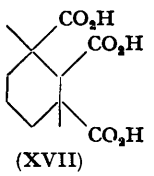
(XVII)

Reduction of either of the C_{17}-acids yields the same tetrahydroxy-compound (XVI), $C_{17}H_{32}O_4$, a result to be expected of acids bearing the relationship expressed above. It was heated with various dehydrating agents, but no crystalline product could be isolated. The product from an attempted rearrangement with hydrochloric acid in acetic acid was, therefore, dehydrogenated with palladised charcoal to a hydrocarbon, $C_{16}H_{24}$. Its spectrum displayed maxima at (2210), 2680, and (2890) Å (log ε 3·99, 2·92, 2·75), similar to the spectrum of a tetra-alkyl substituted benzene, *e.g.*, prehnitene (cf. Amer. Petroleum Inst. Res. Project No. 44, Nat. Bur. Standards, Washington), and is probably (VIII). Such a compound would be expected to resist aromatisation (cf. Clemo and Dickenson, *loc. cit.*; Cocker *et al.*, *loc. cit.*).

Further work is in progress and attempts are being made to degrade marrubiin to the tricarboxylic acid (XVII), already obtained from abietic acid. This would also enable the stereochemistry of the first ring to be elucidated; work on these lines is being pursued.

Experimental

Extraction of Marrubiin (III) (cf. Mercier and Mercier, *Compt. rend.*, 1932, **195**, 1102; Ghigi, 1948, *loc. cit.*).—Powdered horehound (10 lb.) was extracted in a continuously operating apparatus with cold acetone (14 l.) for 48 hr.; the acetone was concentrated to 1 l., and the solution was filtered. The filtrate was evaporated to dryness and the residue set aside with

* The exocyclic Δ^8-butenolide is also sterically possible: a decision on the exact position of the double bond awaits further evidence.

ether (1 l.) for 15 hr., thus removing waxes and chlorophyll and leaving a residue of impure marrubiin (45 g.). This was dissolved in 5% alcoholic sodium hydroxide (300 c.c.), boiled for 10 min., and poured into a large excess of water. The precipitated marrubiin (30 g.) was collected and crystallised from alcohol as prisms (25 g.), m. p. 160°, $[\alpha]_D^{20}$ +35·8° (*c*, 3·1 in $CHCl_3$).

Marrubic acid (X; R = H) (Lawson and Eustice, *loc. cit.*) had $[\alpha]_D^{15}$ +8·81° (*c*, 1·01 in MeOH).

Oxidation of Marrubiin with Chromic Acid to the Dilactone, $C_{17}H_{24}O_4$ (IV).—Carried out essentially in accordance with the procedure of Ghigi (*loc. cit.*), this afforded the dilactone, m. p. 160°, $[\alpha]_D^{16}$ +30° (*c*, 1·2 in $CHCl_3$) (Found: C, 69·2; H, 8·3. Calc. for $C_{17}H_{24}O_4$: C, 69·9; H, 8·2. Calc. for $C_{17}H_{22}O_4$: C, 70·4; H, 7·7%). Ghigi reported m. p. 162° for a compound $C_{17}H_{22}O_4$.

Ozonolysis of Marrubiin (cf. Holliss, Richards, and Robertson, *loc. cit.*).—Marrubiin (5 g.) in acetic acid (100 c.c.) was ozonised with 5% ozone, and the solvent then removed in a vacuum. The oily product, dissolved in methyl alcohol, deposited a dilactone (0·9 g.), m. p. 157—159°, undepressed by the previous compound.

Lactonic Hydroxy-acid, $C_{17}H_{26}O_5$ (IX; R = H).—(a) *By hydrolysis of the dilactone*, $C_{17}H_{24}O_4$. The dilactone (0·7 g.) was refluxed in 10% methyl-alcoholic potassium hydroxide (10 c.c.) for 4·5 hr. After removal of the alcohol, the *hydroxy-acid* was liberated with hydrochloric acid, removed with chloroform, and crystallised from ethyl acetate as plates, m. p. 205—207°, $[\alpha]_D^{16}$ −15·0° (*c*, 1·56 in $CHCl_3$) (Found: C, 65·6; H, 8·3%; equiv., 338. $C_{17}H_{26}O_5$ requires C, 65·8; H, 8·3%; equiv., 310). Hydrolytic titration gave an equivalent wt. 141. The acid was relactonised by heating it with acetic anhydride and sodium acetate on the water-bath, and purifying the product on an alumina column (benzene). The *methyl* ester * of the hydroxy-acid, prepared in ether with diazomethane, crystallised from alcohol in plates, m. p. 154°, $[\alpha]_D^{19}$ −18·8° (*c*, 0·7 in $CHCl_3$) (Found: C, 66·3; H, 8·5; active H, 0·31. $C_{18}H_{28}O_5$ requires C, 66·7; H, 8·6; 1 H, 0·31%).

(b) *By oxidation of marrubic acid with dilute aqueous permanganate solution.* Marrubic acid (15 g.), dissolved in N-sodium hydroxide (45 c.c.) and diluted to 750 c.c., was treated dropwise with 5% aqueous potassium permanganate (182 c.c.), and the mixture was stirred for 1 hr. It was heated on the water-bath for 10 min., then filtered, the filtrate extracted with ether, and the aqueous layer treated with sulphur dioxide to remove marrubic acid. This was collected, and the filtrate was acidified with hydrochloric acid and extracted with ether from which the acid (3 g.) was obtained.

Dehydrogenation of the Lactonic Hydroxy-acid.—The acid (2·5 g.) was heated with palladised charcoal (2·5 g.) at 260° for 48 hr. A pungent acetic acid-like smell was noticed. From the reaction a colourless oil (0·2 g.), b. p. 120°/22 mm., was obtained. It yielded a picrate, m. p. 139—140°, and trinitrobenzene adduct, m. p. 159—160°, undepressed by the picrate and trinitrobenzene adduct, respectively, of 1 : 2 : 5-trimethylnaphthalene.

Acetoxy-lactonic Acid, $C_{19}H_{28}O_6$ (IX; R = Ac).—(a) *By acetylation of the lactonic hydroxy-acid*, $C_{17}H_{26}O_5$. Acetylation with pyridine–acetyl chloride for 14 hr. at room temperature afforded the *acetoxy-acid*, crystallising from *cyclo*hexanone as plates, m. p. 246°, $[\alpha]_D^{12}$ −15·1° (*c*, 0·74 in $CHCl_3$) (Found: C, 64·6; H, 7·9. $C_{19}H_{28}O_6$ requires C, 64·8; H, 8·0%). The *methyl* ester of the acetoxy-acid, prepared with diazomethane, crystallised from methyl alcohol as plates, m. p. 133° (Found: C, 65·4; H, 8·7. $C_{20}H_{30}O_6$ requires C, 65·6; H, 8·2%).

(b) *By oxidation of acetylmarrubic acid with chromic acid.* Acetylmarrubic acid, m. p. 112°, $[\alpha]_D^{15}$ −20·7° (*c*, 0·8 in MeOH) (6 g.), in acetic acid (30 c.c.) was treated with chromic acid (15 g.) in water (20 c.c.) and acetic acid (80 c.c.), added slowly at room temperature. After 72 hr. the acetic acid was removed under reduced pressure, and the residue treated with hydrochloric acid and extracted several times with ether. The acetoxy-acid removed from the ethereal solution by extraction with carbonate solution was identical with the product described above (m. p. and mixed m. p.) (Found: C, 64·8; H, 8·1%).

Lactonic Keto-acid, $C_{17}H_{24}O_5$ (XII).—This compound was prepared by the oxidation of marrubic acid (cf. Ghigi, *loc. cit.*) or the lactonic hydroxy-acid, $C_{17}H_{26}O_5$, with chromic acid in acetic acid. It had m. p. 222—223°, $[\alpha]_D^{23\cdot5}$ +88·7° (*c*, 2·4 in $CHCl_3$) (Found: C, 66·0; H, 7·55. $C_{17}H_{24}O_5$ requires C, 66·2; H, 7·8%). Its *methyl* ester, had m. p. 164°, not 154° as described by Ghigi (Found: C, 67·4; H, 8·2; OMe, 10·75. $C_{18}H_{26}O_5$ requires C, 67·1; H,

* This is probably the ester obtained by Ghigi on the oxidation of marrubic acid and esterification of the product.

8·1; OMe, 9·7%). Its *oxime* had m. p. between 260° and 280° (decomp.) according to rate of heating (Found: N, 4·45. $C_{17}H_{25}O_5N$ requires N, 4·3%). It gave no ferric colour. The *oxime* of the methyl ester had m. p. 187—189° (Found: C, 63·9; H, 7·9. $C_{18}H_{27}O_5N$ requires C, 64·1; H, 8·0%).

Methyl Ester of the Lactonic Keto-acid obtained by Oxidation of Methyl Marrubate.—Methyl marrubate was obtained as follows. A mixture of marrubic acid (2 g.) and alcohol (30 c.c.), containing potassium hydroxide (0·32 g.), was evaporated to dryness, and the residue was heated with methyl iodide (30 c.c.) and anhydrous potassium carbonate (2·0 g.) in a sealed tube at 100° for 3 hr. and at 150° for a further 1 hr. The product crystallised from light petroleum as plates (1 g.), m. p. 84—85° (cf. Hole *et al.*, *loc. cit.*), $[\alpha]_D^{15}$ 0° (*c*, 0·8 in MeOH). The ester (2·7 g.) in acetic acid (20 c.c.) was treated with chromic acid (6·5 g.) in water (10 c.c.) and acetic acid (50 c.c.), added dropwise with stirring, at room temperature. The mixture was left for 72 hr., the acetic acid removed in a vacuum, and the residue treated with 18% hydrochloric acid (50 c.c.) and then extracted several times with ether. The extract, washed first with sodium carbonate solution and then with water, yielded a solid which, crystallised (0·8 g.) from alcohol, had m. p. 162°, undepressed by a specimen of the methyl ester of the lactonic keto-acid mentioned above.

Preparation of the Unsaturated Lactone, $C_{17}H_{22}O_4$ (XIII), *of the Lactonic Keto-acid.*—The lactonic keto-acid was refluxed in acetic anhydride–sodium acetate for 1·5 hr. The unsaturated *lactone,* isolated by the usual methods after removal of the acetic anhydride at reduced pressure, crystallised from aqueous alcohol as needles, m. p. 148—150°, $[\alpha]_D^{17}$ −132° (*c*, 0·5 in $CHCl_3$) (Found: C, 70·3; H, 7·4. $C_{17}H_{22}O_4$ requires C, 70·3; H, 7·6%). It readily absorbed bromine and decolorised methanolic permanganate. Treated with aniline in the cold for 36 hr. it gave an *anilide* (needles from aqueous methyl alcohol), m. p. 196° (Found: C, 71·8; H, 7·6. $C_{23}H_{29}O_4N$ requires C, 72·1; H, 7·6%). When 0·1 g. of lactone was boiled with 20 c.c. of water for 10 hr. it yielded unchanged lactone (0·05 g.), and keto-acid (0·025 g.). Reduced in acetic acid (0·5 g. in 30 c.c.), over Adams's catalyst (0·1 g.), the lactone gave a semi-solid product which was extracted with dilute sodium carbonate, yielding unchanged unsaturated lactone (0·2 g.) and a saturated *acid* (0·15 g.), m. p. 262—264° (needles from dilute methyl alcohol) (Found: C, 69·4; H, 8·9. $C_{17}H_{26}O_4$ requires C, 69·4; H, 8·8%).

Ozonolysis of Anhydrotetrahydromarrubiin.—This compound (Lawson and Eustice, *loc. cit.*), $[\alpha]_D^{21}$ +51·4° (*c*, 2·0 in MeOH), from 10 g. of marrubiin, was ozonised in ethyl acetate at −5°, and the product was hydrogenated over palladised charcoal. A gum was obtained which on refluxing with ether yielded a *keto-lactone* (V) (1·3 g.), and this crystallised from methyl alcohol as needles, m. p. 195°, $[\alpha]_D^{15}$ +118·6° (*c*, 1·06 in $CHCl_3$) (Found: C, 70·7; H, 8·5. $C_{14}H_{20}O_3$ requires C, 71·2; H, 8·5%). Its *oxime* (feathery needles from dilute alcohol) had m. p. 182—183° (Found: N, 5·20. $C_{14}H_{21}O_3N$ requires N, 5·6%) and gave no ferric colour. On hydrolysis with 10% methyl-alcoholic potassium hydroxide, the keto-lactone yielded an *acid,* m. p. 172—174° (needles from methyl alcohol) $[\alpha]_D^{22}$ −30·9° (*c*, 2·0 in MeOH) (Found: C, 66·1; H, 8·5. $C_{14}H_{22}O_4$ requires C, 66·1; H, 8·7%).

Oxidation of Anhydromarrubiin.—The crude anhydro-compound (10 g.), prepared by Lawson and Eustice's method (*loc. cit.*), was warmed on the water-bath with chromic oxide (20 g.) in acetic acid (100 c.c.) and gave the above keto-lactone (0·4 g.), m. p. and mixed m. p. 195°.

Dehydrogenation of the Keto-lactone.—This compound (0·55 g.) was heated for 20 hr. at 300—320° with selenium (1·2 g.) and yielded an oil (0·06 g.; b. p. 110°/2 mm.), which failed to give a picrate.

Marrubenol (VII).—Marrubiin (20 g.) in a Soxhlet thimble was refluxed (18 hr.) with lithium aluminium hydride (12 g.) in dry ether (200 c.c.). The product was decomposed in the usual way. The *triol* (19 g.) crystallised from dilute alcohol in needles, m.p. 138°, $[\alpha]_D^{15}$ +19·9° (*c*, 1·1 in MeOH) (Found: C, 71·2; H, 9·4. $C_{20}H_{32}O_4$ requires C, 71·4; H, 9·5%).

Marrubanol.—The preceding compound (10·5 g.) was reduced in acetic acid (200 c.c.) over Adams's catalyst (0·5 g.). The product crystallised from dilute alcohol as rhombs (4 g.), m. p. 175°, $[\alpha]_D^{20}$ +15·15° (*c*, 2·0 in MeOH) (Found: C, 70·8; H, 11·2. $C_{20}H_{36}O_4$ requires C, 70·6; H, 10·6%).

Attempted Dehydration of Marrubanol, and Dehydrogenation of the Product.—Marrubanol (7·6 g.) was heated with anhydrous formic acid (27 g.) for 4 hr., and the mixture poured into water. The yellow oil was collected (4·8 g.) and distilled, yielding two fractions: (*a*) (1·2 g.), b. p. 154°/2 mm. (Found: C, 79·4; H, 10·6. $C_{20}H_{32}O_2$ requires C, 78·9; H, 10·5%); and (*b*) (1·6 g.) (Found: C, 77·2; H, 9·9%). Fraction (*a*) was heated with palladised charcoal (1·2 g.) at 22° for 19 hr., and at 260° for a further 29 hr. The *product* consisted of an oil (0·4 g.; b. p.

119°/2 mm.) (Found: C, 86·6; H, 11·1. $C_{16}H_{24}$ requires C, 88·8; H, 11·2%). It failed to give a picrate.

Reduction of the C_{17} Acids and Esters with Lithium Aluminium Hydride.—The acids (6 g.), or equivalent quantities of ester, were refluxed in a Soxhlet apparatus with lithium aluminium hydride (3·2 g.) in ether (550 c.c.) for 4 days, and the product worked up in the usual way. A gum (2·5 g.), which slowly solidified, was obtained; the *tetrol* (XVI), crystallised first from acetone and then methyl alcohol, had m. p. 193—194°, $[\alpha]_D^{14}$ +19·8° (*c*, 1·2 in MeOH) (Found: C, 67·6, 67·9; H, 10·8, 10·7. $C_{17}H_{32}O_4$ requires C, 68·0; H, 10·7%) and was very sparingly soluble in ether.

Dehydration of the Alcohol and Aromatisation of the Product.—The alcohol (2·4 g.), dissolved in a mixture of glacial acetic acid (25 c.c.) and concentrated hydrochloric acid (25 c.c.), was set aside overnight. The ice-cold solution was then saturated with hydrogen chloride, set aside for 1 hr., and finally heated on the water-bath for 3·5 hr. The mixture was poured into water and extracted with ether, and the extract washed with sodium carbonate solution. Removal of the ether yielded an intractable gum which was dehydrogenated with palladised charcoal (3 g.) for 48 hr. at 250—260°. The product (0·5 g.) was fractionated over sodium, yielding a colourless oil, b. p. 125°/12 mm. (Found: C, 88·6; H, 11·3. $C_{16}H_{24}$ requires C, 88·9; H, 11·1%). This is probably the *compound* (VIII).

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