Regioselective Ether Cleavages of Rotenoids: Spiro-ether Formation and Stereoselective Isotopic Labelling of (*E*)- or (*Z*)-Prenyl Methyl Groups in (6a*S*, 12a*S*)-Rot-2′-enonic Acid

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Treated with boron tribromide (−)-(6a*S*,12a*S*,5′*R*)-rotenone is converted first into a primary allylic bromide by ring-*E* cleavage, then into the 2-de-*O*-methyl and finally the 2,3-dide-*O*-methyl derivatives. With (6a*S*,12a*S*,5′*R*)-6′7′-dihydrorotenone and (6a*S*,12a*S*)-isorotenone, ring-*E* cleavage does not take place. The main reaction is 2-, followed by 2,3-demethylation: this supports a stereospecific pericyclic mechanism for the rotenone ring-*E* cleavage. Treatment of the geometrically pure (*E*)-bromide with cyanoboro-deuteride or -tritide leads to (*E*)-4′-labelled (6a*S*,12a*S*)-rot-2′-enonic acid without reduction of the 12-carbonyl group. By using [7′-^{13}C or -^{14}C]-rotenone, (*E*)-[4′-^{13}C- or -^{14}C-]rot-2′-enonic acid is accessible. Trimethylsilyl iodide can cleave the 2-methoxy-group of rotenone without rupturing ring *E*, and remethylation with [^{2}H]- or [^{3}H]-diazomethane represents a convenient method for preparing a general tracer molecule.

On treatment with sodium hydride, 3-de-*O*-methylisorotenone (but not the 2-isomer) rearranges into a spiro-ether, thus confirming the position of initial de-*O*-methylation as deduced from ^{1}H and ^{13}C n.m.r. data. Because of this rearrangement, methylenation (NaH–CH_2I_2) of 2,3-dide-*O*-methylisorotenone gives mainly the methylenedioxy-spiro-ether, with small yields of methylenedioxy-rotenoid.

Deuteriogenolysis of (−)-rotenone over palladium catalyst in ($^{2}H_5$)pyridine gives (*E*)-[4′-^{2}H]rot-2′-enonic acid, but experiments using [7′-^{13}C]rotenone indicate stereoselectivity rather than stereospecificity, *ca.* 12% of (*Z*)-[5′-^{13}C]- accompanying the major (*E*)-product. A similar specimen of [4′-^{14}C]rotenonic acid has been prepared. A hydrogenolysis route from amorphigenin, *via* [8′-^{2}H]rotenone, to (*Z*)-[5′-^{2}H]rot-2′-enonic acid is described.

(−)-(6a*S*,12a*S*,5′*R*)-Rotenone (1a) contains ten ether C–O linkages, and fissions of the C(6a)–O(7) and O(1′)–C(5′) bonds are involved in a number of its characteristic reactions. Specific ether cleavages open the way to new chemical transformations and offer potentialities for specific isotopic labellings necessary for our biosynthetic work.[1] With this in mind we have engaged in a study of rotenoid ether cleavages initiated by boron halides and other reagents capable of breaking C–O bonds, and of allylic ether hydrogenolysis. The results are reported in this paper.[2]

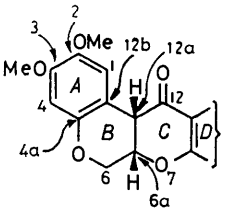
An earlier study[3,4] of the reaction between rotenone and boron tribromide has indicated that in the presence of 1 mol equiv. of reagent the 1′,5′-*seco*-bromide (1b) is formed, whereas use of 2 mol equiv. causes additional demethylation of the C-3 methoxy-group giving (3b); > 3 mol equiv. of reagent de-*O*-methylates at C-2 and C-3, forming (4b). The n.m.r. data[4] given in support of structure (1b) seemed to us to be inconsistent with that formulation, and as the evidence for the position of initial de-*O*-methylation seemed slender, the reaction was re-examined.

Treatment of rotenone with 1 mol equiv. of boron tribromide at −5 to −10 °C for 2 min gave, as reported,[3,4] a 1′,5′-*seco*-bromide (58%), m.p. 152—154 °C as a methanol solvate. ^{1}H N.m.r. signals at δ 3.35 (2 H, d, 1′-H), 5.58 (1 H, t, 2′-vinyl), and 3.87 (2 H, s, 4′-H_2) supported structure (1c) rather than (1b) and ^{13}C n.m.r. signals at δ 41.8 (t, C-4′), 22.3 (t, C-1′), and 14.7 (q, C-5′) with vinyl carbon peaks at δ 127.3 (d, C-2′) and 132.4 (s, C-3′) were compatible only with the primary bromide structure (1c). In order to identify the geometry of the trisubstituted double bond we established the chemical relationship with (6a*S*,12a*S*)-rot-2′-enonic acid (1d), and this allowed us to develop a stereospecific isotopic labelling method for the latter. (6a*S*,12a*S*)-Rot-2′-enonic acid † was first obtained by hydrogenolysis of rotenone[5,6] (see later) and is now a known natural product.[1a] It is the primary product of ring-*D* prenylation in the biosynthesis of rotenoids and in *Amorpha fruticosa* it forms the starting point for the ring-*E* biosynthetic variations.[1]

The two *C*-methyl groups of rot-2′-enonic acid (1d) are readily distinguished by ^{13}C n.m.r.;[7] C-4′ (*trans* to C-1′ resonates at δ 25.7 and C-5′ (*cis* to C-1′) at 17.8 p.p.m. On treating the bromide (1c) with sodium cyanoborohydride in hexamethylphosphoramide,[8] (6a*S*,12a*S*)-rot-2′-enonic acid, m.p. 206—207 °C was obtained directly without reduction of the 12-carbonyl. Using cyanoborodeuteride a monodeuterio-product was obtained, m.p. 206—207 °C, M^+ 397, thus marking the site of the halogen. The ^{13}C n.m.r. spectrum (proton-decoupled) of the monodeuterio-compound showed the signal at δ 17.8 to be the same as in the undeuteriated specimen, but the signal at δ 25.7 took on triplet form. Thus the deuterio-compound (1e) and hence the bromide (1c) are both *E*-compounds. These results have useful consequences for specific labellings required for biosynthetic work, and by using a tritiated reagent ($B^{3}H_3CN$ was made by $B^{1}H_3CN$–$^{3}H_2O$ exchange), (*E*)-[4′-^{3}H]rot-2′-enonic acid was prepared.

† The older stereochemical designation (−)-rot-2′-enonic acid used to indicate its relationship to natural (−)-rotenone, is misleading particularly as it has a positive rotation in chloroform; we now discard this usage. In describing products relating to natural rotenone, the dropping of stereochemical descriptors implies that the remaining configurational elements are the same as in the natural parent.

Furthermore, since the 4′-methyl group of rot-2′-enonic acid (1d) is derived from C-7′ of rotenone, by using the [7′-^{13}C]- and [7′-^{14}C]-rotenone described in the previous paper we have available a route to (*E*)-[4′-^{13}C]- and [4′-^{14}C]rot-2′-enonic acids. A further piece of information which the possession of (*E*)-[4′-^{2}H]rot-2′-



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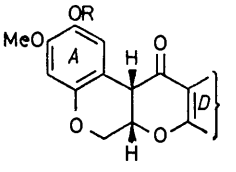

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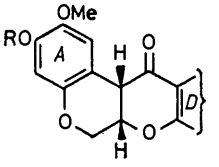
(8) R = Ac



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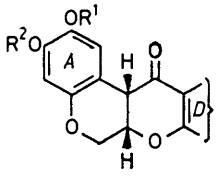
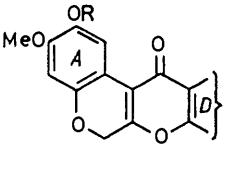


(4) $R^1 = R^2 = H$
(10) $R^1 = R^2 = Ac$



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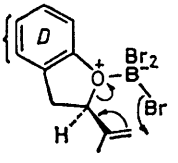

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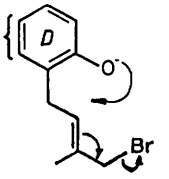
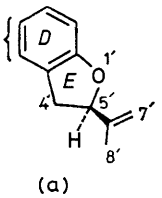


(7)

enonic acid has given us is the assignment of the two *C*-methyl signals in the ^{1}H n.m.r. spectrum of (1d). These occur at δ 1.72 and 1.80 and in the 4′-monodeuteriospecimen the signal at 1.72 is broadened whilst a ^{2}H n.m.r. spectrum shows only a single line with this chemical shift. The signal at δ 1.72 is thus due to the 4′- and that at 1.80 to the 5′-methyl.

The product obtained by treating the bromo-compound (1c) with methanol–water (9 : 1) at 40 °C for 1 h has been given structure (1f).[3] In our hands this treatment gave the methyl ether (1g) arising by methanolysis, and formulated on spectroscopic information. Higher temperatures, longer reaction times, and increases in the water concentration also resulted in formation of (1g). Methylated with diazomethane in ether, (1c) gave the trimethoxy-compound (1h); similar treatment in methanol gave the tetramethoxy-ether (1i).

The remarkable ease of this stereospecific allyl ether cleavage of rotenone [(1a) ⟶ (1c)] by boron tribromide suggests that it proceeds by a pericyclic mechanism



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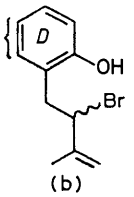

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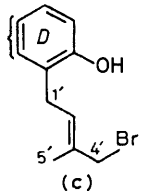

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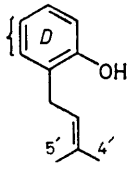

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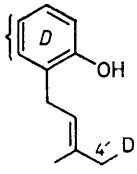

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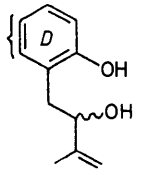

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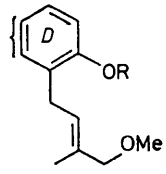
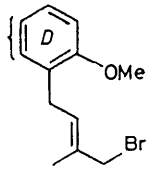


(g) R = H
(i) R = Me



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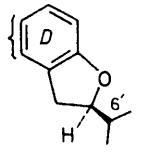

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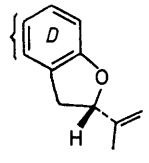

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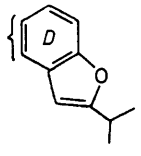

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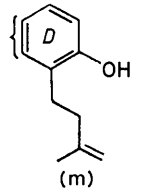

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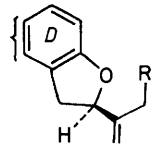
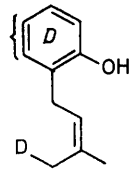


(n) R = OH
(o) R = Br



(p)

[see structure (6)]. It is also effected by the milder boron trichloride, and under the conditions used an excess (3 mol equiv.) of reagent caused little attack on the ring-*A* methoxy-groups. Support for a pericyclic mechanism is given by the fact that 6′,7′-dihydrorotenone (1j) does not undergo O(1′)–C(5′) scission when treated with 1 mol equiv. of boron tribromide: instead, ring-*A* methoxy-cleavage occurs giving (2j). It has been shown that treatment of (1c) with aqueous sodium

hydrogen carbonate [3,4] causes recyclisation to give a mixture of (5′*R*)(natural) (1a) and (5′*S*) (unnatural) (1k) rotenone, the base conditions being insufficient to epimerise the C-6a,C-12a system. This is an important reaction, as to our knowledge it is the only simple method available for epimerising C-5′ in natural rotenone: the transformation must now be represented as an S_N2' reaction [see structure (7)].

As mentioned earlier, the first reported boron-tribromide-induced de-*O*-methylation of rotenone was considered to occur at C-3; [4] this matter is of some importance in identifying the mono-de-*O*-methyl rotenone formed metabolically and photochemically.[4,9] In our laboratory, when rotenone was treated with 2 mol equiv. of boron tribromide a crystalline methanol solvate $C_{22}H_{21}O_6Br$·MeOH, evidently (2c) or (3c) from ^{1}H n.m.r. analysis, was isolated. The compound cyclised in mild alkaline medium giving a mixture of (2a) plus (2k) or (3a) plus (3k) from which (2a) or (3a) having the natural 5′*R*-configuration was isolated by crystallisation: as expected, the n.m.r. spectrum was similar to that of rotenone except for the lost methoxy-group. This system is, however, inconvenient for studying ring-*A* demethylation because the latter is prefaced by the ring-*E* cleavage reaction; thus subsequent recyclisation and separation steps are required. Attention was therefore transferred to (6a*S*,12a*S*)-isorotenone (1l). In this case, treatment with 1 mol equiv. of boron tribromide causes no ring-*E* cleavage, but two products of monodemethylation shown to be (2l) and (3l) are formed in *ca.* 10 : 1 ratio. Excess of reagent leads to the catechol (4l), and by partial remethylation with diazomethane further supplies of the scarcer (3l) can be obtained. Since the major monodemethylated isorotenone, shown later to be (2l), is obtained when the monodemethylated rotenone discussed above is prototropically rearranged with sulphuric–acetic acid, the two series are structurally connected and deductions for one apply also to the other.

TABLE 1

^{1}H N.m.r. chemical shifts of the ring *A* aromatic protons (1- and 4-H) [a,b]

	1-H	4-H
Rotenone (1a)	6.71	6.43
2-De-*O*-methylrotenone (2a)	6.63	6.41
6′,7′-Dihydrorotenone (1j)	6.73	6.44
2-De-*O*-methyl-6′,7′-dihydrorotenone (2j)	6.67	6.43
Isorotenone (1l)	6.70	6.42
2-De-*O*-methylisorotenone (2l)	6.62	6.42
3-De-*O*-methylisorotenone (3l)	6.69	6.38
2,3-Dide-*O*-methylisorotenone (4l)	6.61	6.38
2-*O*-Acetyl-2-de-*O*-methylisorotenone (8l)	6.80	6.58
3-*O*-Acetyl-3-de-*O*-methylisorotenone (9l)	6.75	6.61
2,3-Di-*O*-acetyl-2,3-dide-*O*-methylisorotenone (10l)	6.97	6.75

[a] ($^2H_6$)Acetone calibrated by an internal benzene line. [b] Data in our preliminary communication [2a] relate to $CDCl_3$.

The orientation problem was approached spectroscopically. Table 1 contains ^{1}H n.m.r. data, accurately measured on solutions in ($^2H_6$)acetone and calibrated by internal benzene, for rotenone, isorotenone, and various demethylated and reacetylated derivatives. Demethylation causes a small increase in shielding of the adjacent aromatic proton, allowing the assignments shown. Deshieldings of the adjacent 1-H or 4-H caused by acetylation also generally support these orientations but here there are conformational effects and *m*-shifts. Since in all cases shifts are small, attention was turned to ^{13}C spectra for confirmation. Full ^{13}C assignments are available for rotenone, isorotenone, and other rotenoids and their transformation products [10] and the signals due to the 4a- and 12b-carbon atoms of aromatic

TABLE 2

^{13}C N.m.r. chemical shifts of the ring *A* aromatic carbons (C-4a and 12b) [a]

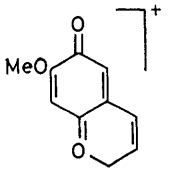
	C-12b	C-4a
Rotenone (1a)	104.9	147.6
	(105.1) [b]	(149.2)
Isorotenone (1l)	104.9	147.6
2-De-*O*-methylisorotenone (2l)	106.1	146.9
	(106.9)	(147.8)
3-De-*O*-methylisorotenone (3l)	104.7	148.1
	(105.4)	(149.2)
2,3-Dide-*O*-methylisorotenone (4l)	(105.8) [a]	(148.1)
2-*O*-Acetyl-2-de-*O*-methylisorotenone (8l)	105.9	151.6
3-*O*-Acetyl-3-de-*O*-methylisorotenone (9l)	112.0	145.9
2,3-Di-*O*-acetyl-2,3-dide-*O*-methylisorotenone (10l)	112.7	151.5

[a] $CDCl_3$. [b] Shifts in parentheses relate to ($^2H_6$)acetone as solvent.

ring *A* are readily located singlets in the off-resonance spectra. Relative to benzene, the shifts at a specific *para*-carbon due to the substituents OMe, OH, and OAc are −7.7, −7.3, and −2.3 p.p.m.[11] The formation of a *p*-acetoxy-group is thus easily discerned by monitoring the shift induced by acetylation, relative to the parent phenol or methoxyphenol. A shift of the C-12b signal from δ 104—106 to 112—113 signifies a *para*-located 3-acetoxy-group, whilst a shift of the C-4a signal from δ 146—148 to near 151.5 signifies a *para*-located 2-acetoxy-group (Table 2). The assignments made agree with those deduced from the ^{1}H spectra.

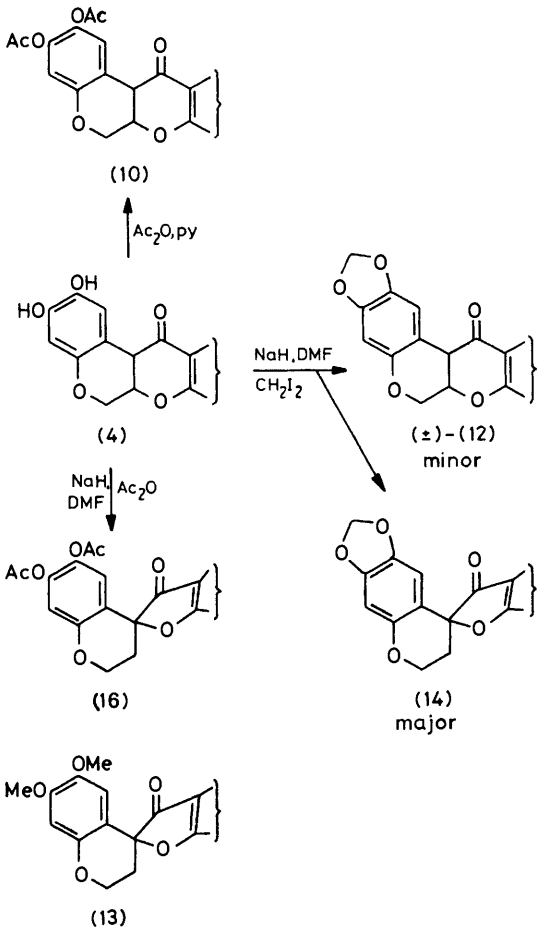
It is thus clear that boron tribromide first de-*O*-methylates rotenone and isorotenone largely at the 2-position rather than the 3- as originally claimed,[4] and apparently demethylation by insect metabolism, or photochemical processes, involves the 2-position. The general preference for 2-de-*O*-methylation may be rationalised by invoking a role for the *para*-5-*O* in increasing the Lewis basicity of the 2-O towards boron tribromide. This easy availability of isorotenone specifically demethylated at the 2-position allows ready introduction of H- or C- isotopes by remethylation giving a molecule suitable for general tracking purposes. Because demethylation is prefaced by ring-*E* cleavage, the situation with rotenone itself is less convenient and we have therefore examined other demethylation agents. Lithium t-butyl thiolate,[12] representative of the thiolate anion group, proved too basic, opening ring *C* by β-elimination and participating in conjugate addition to the newly generated α-unsaturated ketone. On the

other hand the methionine–methanesulphonic acid system [13] was too acidic and caused prototropic shift involving ring-*E* giving 2- and 2,3-dide-*O*-methylisorotenone among the products. However, treatment of rotenone with trimethylsilyl iodide [14] (1.3 mol equiv.; 60 °C), a reagent with strong oxygenophilic silicon and soft nucleophilic iodine in combination, gave 2-de-*O*-methylrotenone (2a) directly in 25—30% yield thus making the labelling of rotenone direct and simple. Remethylation using deuteriated and tritiated diazomethane (made by equilibrating the latter with ^2H_2O or ^3H_2O) gave rotenone containing a deuteriated * and a tritiated 2-methoxy-group.



(11)

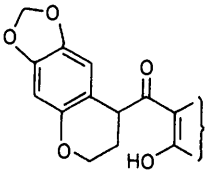
The accessibility of 2,3-dide-*O*-methylisorotenone (4l) led us to attempt methylenation to form a methylenedioxy-rotenoid of the isomilletone (12) series, and in the process abundant chemical confirmation of the spectroscopic demethylation assignments became available (Scheme 1). Treatment with sodium hydride and diiodomethane in DMF at 20 °C gave (12l) in low yield, having $\nu_{max.}$ 1 680 cm^{-1} and the expected ^1H n.m.r. and mass-spectral fragmentation properties. The isomeric major product differed from (12l) in that the mass spectral fragment carrying the *A*/*B* ring residue (*m*/*z* 176) was less abundant than the M^+ 378; in the spectrum of (12l) *m*/*z* 176 is the base peak. The major compound had $\nu_{max.}$ 1 710 cm^{-1} and this reminded us of a spiro-compound (13l) ($\nu_{max.}$ 1 709 cm^{-1}) which we had encountered in earlier work.[16] U.v. data confirmed the close similarity of chromophores and, excluding the methoxy- and methylenedioxy-appendages there were close n.m.r. parallels leading to structure (14l) for the new and major product of the di-iodomethane reaction. In confirmation, zinc dust and alkali gave a compound of the rotenol type (15l) by cleavage of the spiro-C–O bond: such a product from (13l) is known.[16] The latter spiro-compound is formed by radical processes,[16] but in the present case only base-catalysed initiation is necessary for spiro-compound formation. Thus treatment of (4l) with NaH—DMF followed by quenching with an acetylating mixture gave (16l). Mechanistically (Scheme 2) the reaction is viewed as enolate anion formation at C–12a, and β-elimination to give (18). Prototropic shift leads to the quinone methide (19), which undergoes



SCHEME 1 Reactions of a 2,3-dihydroxyrotenoid

addition using the ring-*D* phenolate anion to give the spiro-compound (16l) after acetylation.

The implication of this mechanism is a specific requirement for a free phenolic 3-hydroxy-group. Treatment of the 2- and the 3-monode-*O*-methylated isorotenones



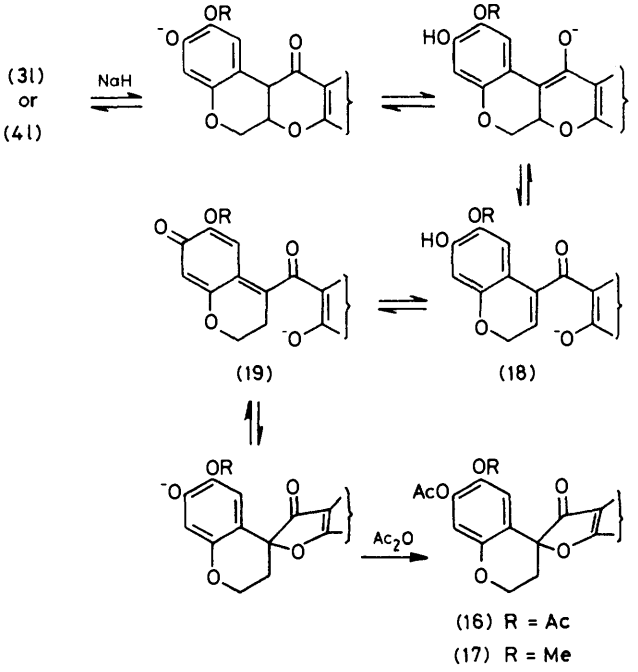
(15)

(identified by spectral means), with sodium hydride followed by acetylation gave the 3-acetoxy-spiro-derivative (17l) from the latter and (±)-2-*O*-acetylisorotenone (±)-(8l) from the former. Spectroscopic and chemical assignments are thus in harmony.

In earlier work [5] we have examined the allylic hydrogenolysis of (—)-rotenone over palladium in pyridine or ethanolic sodium acetate. The products are rot-2′-enonic acid (1d) together with rot-3′-enonic acid (1m) which can be separated by preparative layer chroma-

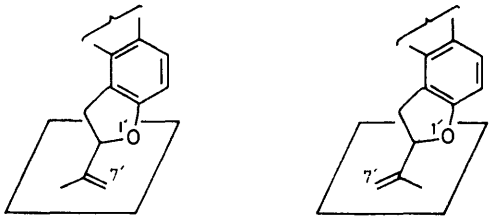
* Reed and Wilson [15] showed that the major fragmentation of rotenone is formation of a retro-Diels–Alder product which loses a methyl to give a species *m*/*z* 177: this loss is represented as being of the 3-methyl. Our 2-deuteriated specimen of rotenone shows conclusively that it is the 2-methyl that is lost, giving (11) as the fragment.

tography or, more efficiently, by preparative h.p.l.c. For the purpose of examining the stereoselectivity of this reaction we have carried out the corresponding deuteriolysis using deuterium gas over palladium in ($^2H_5$)pyridine. The ^1H, ^2H, and ^{13}C n.m.r. spectra of the [^2H]rot-2′-enonic acid thus obtained were closely similar



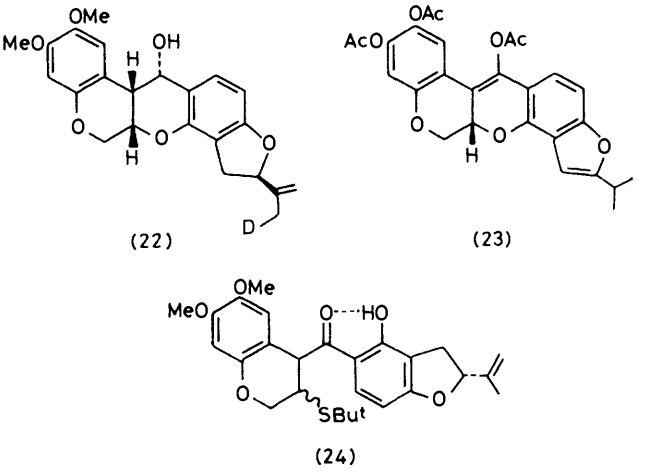
SCHEME 2 Anionic formation of spiro-compounds from 3-hydroxylated rotenoids

to those of the (*E*)-[4′-^2H]-labelled specimen (1e) obtained from treating (1c) with cyanoborodeuteride. This strong selectivity suggests that there is a preference for orientation (20) over orientation (21) on the catalyst. Unfortunately the quality of the ^2H spectrum was inadequate for quantitative evaluation, the resonance line being inherently broadened and the signal : noise ratio



(20) (21)

being rather low. Hydrogenation of rotenone was therefore repeated using [7′-^{13}C]-material prepared earlier.[17] ^{13}C-Labelled rotenonic acid (1d) was isolated as before and the C-4′ and C-5′ signals in the enriched and natural abundance specimens (spectra proton-decoupled under identical conditions) were compared in turn using 20 sharp signals, and averaging the fractional enhancements. Five pairs of spectra were used and the mean was taken. This treatment showed that whilst the hydrogenolysis was highly selective, it was not stereospecific, 88% of the ^{13}C-residing in C-4′ and 12% in C-5′. (In the 5 pairs of spectra the maximum variations encountered at C-5′ were within ±4 on either side of the value given.) A similar sample of predominantly [4′-^{14}C]rot-2-enonic acid was made at the same time from [7′-^{14}C]rotenone. These stereoselectively labelled samples are easily produced from 7′-labelled rotenone



and have sufficient label discrimination between the 4′- and 5′-methyl groups for use (with correction for labelling bias) in biosynthetic experiments. [^{13}C]Rot-3′-enonic acid, isolated as the second product in the hydrogenolysis, showed the expected augmentation of the 4′-vinyl resonance at 110.1 p.p.m., in accordance with 1,2-allylic cleavage of O(1)′–C(5′) during the hydrogenolysis.

To complement the preparation of [4′-^2H]rot-2-enonic acid above, we have also devised a synthesis of the [5′-^2H]-isomer. Amorphigenin (1n)[18] was converted into 8′-bromorotenone (1o) by treatment with phosphorus tribromide and the bromo-compound was reduced with lithium aluminium deuteride to give (22).[19] This [8′-^2H]benzylic alcohol was oxidised by the Oppenauer method to provide [8′-^2H]rotenone. Hydrogenolysis and chromatography now gave [5′-^2H]rot-2′-enonic acid (1p). The sample showed one ^2H n.m.r. signal (broad) at δ 1.83 and within the sensitivity of the spectrum available no signal was resolved at δ 1.72. Its origins *via* hydrogenolysis lead us to suspect, however, that there is probably 4′-labelled contamination (*ca.* 10—20%).

EXPERIMENTAL

For general procedures see preceding paper. Methanol solvates were reprecipitated from benzene–light petroleum before n.m.r. measurement.

Reaction of Rotenone with Boron Tribromide.—(a) Rotenone (1.0 g, 2.54 mmol) in dry dichloromethane (6 cm^3) was added with stirring to boron tribromide (0.242 cm^3, 640 mg, 2.55 mmol) in dry dichloromethane (6 cm^3) kept below —5 °C. The mixture was stirred for 2 min, then rapidly

evaporated to dryness (ambient temperature), and the residue cooled in ice was treated with anhydrous methanol (15 cm³). The product was set aside at 4 °C to yield crystals; further crystallisation from methanol–dichloromethane afforded *4'-bromorot-2'-enonic acid* (1c) (700 mg, 58%), m.p. 152—154 °C, $[\alpha]_D^{20}$ + 27.2° (*c* 0.72, $CHCl_3$) {lit.,[3] [claimed as (1b)] m.p. 152—154 °C, $[\alpha]_D^{20}$ + 30.8° (*c* 2.0, $CHCl_3$)} (Found: C, 56.4; H, 5.3. $C_{23}H_{23}BrO_6$ requires C, 56.8; H, 5.3%); $\lambda_{max.}$ 220 (4.41), 235 (4.21), 291 (4.22), and 316 nm (3.92); $\nu_{max.}$ (KBr) 3 500, 1 666, 1 651, 1 594, 840, and 821 cm^{-1}; δ_H 1.90 (3 H, s, 5'-H_3), 3.35 (2 H, d, *J* 7 Hz, 1'-H_2), 3.68 and 3.75 (both 3 H, s. OMe), 3.87 (2 H, s, 4'-H_2), 4.16 (*J* 12 Hz, 6-H_a), 4.57 (1 H, dd, $J_{6.6}$ 12, $J_{6.6a}$ 3 Hz, 6-H_b), 4.86 (1 H, m, 6a-H), 5.58 (1 H, t, *J* 8 Hz, 2'-H), 6.39 (1 H, s, 4-H), 6.43 (1 H, d, *J* 9 Hz, 10-H), 6.70 (1 H, s, 1-H), 6.86 (1 H, s, OH), 7.62 (1 H, d, *J* 9 Hz, 11-H), δ_C 14.7 (q, C-5'), 22.3 (t, C-1'), 41.8 (t, C-4'), 127.2 (d, C-2'), and 132.4 (s, C-3').

The experiment (*a*) was repeated with 1.28 g (5.11 mmol) of boron tribromide. Product isolation in the same manner afforded *2-de-O-methyl-4'-bromorot-2'-enonic acid* (2c) (730 mg, 63%), m.p. 112—115 °C (from methanol–dichloromethane); $[\alpha]_D^{20}$ + 48.5° (*c* 0.45, acetone) {lit.,[4] m.p. 115—125 °C, $[\alpha]_D^{20}$ + 45.0° (acetone)} (Found: C, 55.65; H, 5.2. $C_{22}H_{20}BrO_6.CH_3OH$ requires C, 56.1; H, 4.85%); $\lambda_{max.}$ 219 (4.45), 234 (4.29), 291 (4.23), and 317 nm (3.91); $\nu_{max.}$ (KBr) 1 658, 1 595, 1 514, 839, and 819 cm^{-1}; δ[($^2H_6$)-acetone] 1.91 (3 H, s, 5'-H_3), 2.83 (1 H, s, OH), 3.28 (2 H, d, *J* 6 Hz, 1'-H_2), 3.71 (3 H, s, OMe), 3.97 (2 H, s, 4'-H_2), 4.23 (1 H, d, *J* 12 Hz, 6-H_a), 4.55 (1 H, dd, $J_{6.6}$ 12, $J_{6.6a}$ 3 Hz, 6-H_b), 4.98 (1 H, m, 6a-H), 5.64 (1 H, t, *J* 7 Hz, 2'-H), 6.38 (1 H, s, 4-H), 6.52 (1 H, d, *J* 9 Hz, 10-H), 6.71 (1 H, s, 1-H), 6.91 (1 H, s, OH), and 7.54 (1 H, d, *J* 9 Hz, 11-H).

Reaction of 4'-Bromorot-2'-enonic Acid with Aqueous Methanol.—4'-Bromorot-2'-enonic acid (400 mg) was stirred with methanol (9 cm³) and water (1 cm³) at 65—70 °C for 1 h. The solution was concentrated and set aside at 4 °C. The product was collected and recrystallised from methanol to yield *4'-methoxyrot-2'-enonic acid* (1g) (133 mg, 37%), m.p. 135—136 °C, $[\alpha]_D^{20}$ + 12.2° (*c* 0.29, $CHCl_3$) (Found: C, 65.6; H, 6.4. $C_{24}H_{26}O_7.CH_3OH$ requires C, 65.5; H, 6.55%); $\lambda_{max.}$ 219 (4.41), 234 (4.21), 291 (4.22), and 319 nm (3.88); $\nu_{max.}$ (KBr) 3 410, 1 675, 1 599, 1 518, 838, and 818 cm^{-1}; δ 1.80 (3 H, s, 5'-H_3), 3.22 (3 H, s, 4'-OMe), 3.34 (2 H, d, *J* 7 Hz, 1'-H_2), 3.70 and 3.75 (both 3 H, s, OMe), 4.14 (1 H, d, *J* 12 Hz, 6-H_a), 4.54 (1 H, dd, $J_{6.6}$ 12, $J_{6.6a}$ 3 Hz, 6-H_b), 4.86 (1 H, m, 6a-H), 5.44 (1 H, d, *J* 7 Hz, 2'-H), 6.34 (1 H, s, 4-H), 6.36 (1 H, d, *J* 9 Hz, 10-H), 6.69 (1 H, s, 1-H), 7.07 (1 H, 9-OH), and 7.55 (1 H, d, *J* 9 Hz, 11-H).

Rot-2'-enonic Acid from 4'-Bromorot-2'-enonic Acid.—(*a*) Sodium cyanoborohydride (100 mg, 1.6 mmol) was added to 4'-bromorot-2'-enonic acid (200 mg, 0.42 mmol) dissolved in hexamethylphosphoramide (4 cm³) and the mixture was stirred at 40—45 °C for 40 min. It was then diluted with brine and extracted with ether. The extracts were washed, dried, and evaporated: the residue was separated by p.l.c. (chloroform–methanol, 98 : 2). The major band, after extraction (chloroform) and crystallisation, afforded rot-2'-enonic acid (80 mg, 48%), m.p. 205—206 °C (from methanol), mixed m.p. 205—206 °C with an authentic specimen (m.p. 206—207 °C), and with an i.r. spectrum identical with that of the authentic compound.

(*b*) The reaction was repeated in the same manner, but using sodium cyanoborodeuteride. *4'-Deuteriorot-2'-enonic acid* (113 mg, 57%), m.p. 206—207 °C, was obtained, *m/z* 397 (M^+, 9%).

(5'R)-2-*De*-O-*methylrotenone*.—Rotenone (3.0 g, 7.62 mmol) in dry dichloromethane (15 cm³) was added to boron tribromide (1.45 cm³, 15.33 mmol) in dichloromethane (15 cm³), kept below −5 °C. After 5 min the solution was evaporated. The residue, cooled in ice, was treated in sequence with methanol (30 cm³), acetone (90 cm³), and saturated aqueous sodium hydrogen carbonate (120 cm³). The mixture was stirred at room temperature for 2 h, diluted with water, neutralised (10% hydrochloric acid), and extracted with chloroform. The extracts were washed, dried, and evaporated. The residue was purified by p.l.c. (chloroform–methanol, 95 : 5). The faster-running of the two main bands was isolated, and the product repeatedly crystallised from benzene to yield *2-de-O-methylrotenone* (2a) (494 mg, 16.5%), m.p. 179—181 °C, $[\alpha]_D^{23}$ − 55.4° (*c* 0.23, $CHCl_3$) {lit.,[4] (for claimed 3-de-*O*-methyl), m.p. 177—182 °C, $[\alpha]_D^{20}$ − 54.0° ($CHCl_3$)} [Found: C, 71.35; H, 5.85. $C_{22}H_{20}O_6.(C_6H_6)_{0.5}$ requires C, 71.6; H, 5.5%]; $\lambda_{max.}$ 219 (4.41), 237 (4.01), 241 (4.06), and 294 nm (4.19); $\nu_{max.}$ (KBr) 1 667, 1 611, 1 515, 837, and 822 cm^{-1}, δ 1.74 (3 H, s, 8'-H_3), 3.07 (1 H, dd, 4'-H_a), 3.14 (1 H, dd, $J_{4',4'}$ 16 Hz, 4'-H_b), 3.77 (3 H, s, OMe), 4.13 (1 H, d, 6-Ha), 4.56 (1 H, dd, $J_{6.6}$ 12, $J_{6.6a}$ 3 Hz, 6-H_b), 4.91 and 5.05 (both 1 H, br, s, 7'-H_2), 5.18 (1 H, t, *J* 9 Hz, 5'-H), 6.40 (1 H, s, 4-H), 6.45 (1 H, d, *J* 9 Hz, 10-H), 6.79 (1 H, s, 1-H), 7.79 (1 H, d, *J* 9 Hz, 11-H). Treatment of this product (10 mg) with an excess of ethereal diazomethane gave rotenone (6 mg, 59%), m.p. and mixed m.p. 162—163 °C (with authentic rotenone, m.p. 163 °C). Acetylation of 2-de-*O*-methylrotenone in an excess of acetic anhydride–pyridine (1 : 1) at room temperature gave, after the standard isolation procedure, the 2-*O-acetyl derivative* (8a) (51%), m.p. 204—205 °C, $[\alpha]_D^{23}$ − 127.9° (*c* 0.08, $CHCl_3$) {lit.,[4] (claimed as 3-*O*-acetyl), m.p. 205 °C, $[\alpha]_D^{20}$ − 129.2° ($CHCl_3$)} [Found: C, 67.3; H, 5.35. $C_{24}H_{22}O_7.(CH_3OH)_{0.5}$ requires C, 67.1; H, 5.45%]; $\lambda_{max.}$ 219 (4.47), 235 (4.12), 243 (4.11), 292 (4.21), and 298 nm (4.15); $\nu_{max.}$ (KBr) 1 756, 1 683, 1 606, 1 512, 831, and 808 cm^{-1}; δ 1.77 (3 H, s, 8'-H_3), 2.22 (3 H, s, COMe), 3.08 (1 H, dd, 4'-H_a), 3.17 (1 H, dd, $J_{4',4'}$ 16 Hz, 4'-H_b), 3.74 (3 H, s, OMe), 3.82 (1 H, d, *J* 4 Hz, 12a-H), 4.19 (1 H, d, *J* 12 Hz, 6-Ha), 4.63 (1 H, dd, $J_{6.6}$ 12, $J_{6.6a}$ 3 Hz, 6-H_b), 4.91 (1 H, m, 6a-H), 4.92 and 5.08 (both 1 H, br, s, 7'-H_2), 5.25 (1 H, t, *J* 9 Hz, 5'-H), 6.43 (1 H, s, 4-H), 6.50 (1 H, d, *J* 9 Hz, 10-H), 6.89 (1 H, s, 1-H), and 7.82 (1 H, d, *J* 9 Hz, 11-H).

2-O-*Acetyl-2-de-*O-*methyl*-6a,12a-*didehydrorotenone* (5a).—The preceding acetate (60 mg) was refluxed in acetone (4 cm³) with activated manganese dioxide (1.0 g) for 1 h. The mixture was filtered, the residues were washed with hot acetone, and the combined acetone solutions evaporated to dryness. The residue crystallised from chloroform–methanol to afford *compound* (5a) (25 mg, 43%), m.p. 238—240 °C, $[\alpha]_D^{16}$ − 5.4° (*c* 0.35, $CHCl_3$) (Found: C, 68.65; H, 4.65. $C_{24}H_{20}O_7$ requires C, 68.55; H, 4.75%); $\lambda_{max.}$ 229 (4.38), 247 (4.40), 279 (4.42), and 303 nm (4.30); $\nu_{max.}$ (KBr) 1 764, 1 643, 1 634, 1 610, 1 514, 915, 806, and 780 cm^{-1}; δ [($^2H_6$)acetone] 1.81 (3 H, s, 8'-H_3), 2.28 (3 H, s, COMe), 3.31 (1 H, dd, 4'-H_a), 3.39 (1 H, dd, 4'-H_b), 3.80 (3 H, s, OMe), 5.00 (2 H, s, 6-H_2), 4.97 and 5.13 (both 1 H, br, s, 7'-H_2), 5.39 (1 H, t, *J* 8 Hz, 5'-H), 6.51 (1 H, s, 4-H), 6.90 (1 H, d, *J* 9 Hz, 10-H), 8.09 (1 H, d, *J* 9 Hz, 11-H), and 8.52 (1 H, s, 1-H).

8'-*Bromorotenone* (1o).—Amorphigenin (1n) (88 mg; isolated from *Amorpha fruticosa*) was refluxed in chloroform (2 cm³) and benzene (2 cm³) with phosphorus tribromide (0.3 cm³) for 3.5 h. The mixture was poured into water.

The organic products were isolated through chloroform extraction and purified by p.l.c. (chloroform–propan-2-ol, 20 : 1). The band, after extraction, afforded *compound* (1o) (48 mg, 47%), m.p. 192—193 °C (Found: C, 57.75; H, 4.3. $C_{23}H_{21}BrO_6$ requires C, 58.35; H, 4.45%); *m/z* 474/472 (M^+); $\lambda_{max.}$ 234 (4.21), 243 (4.10), 290 (4.15), and 320 nm (3.95); $\nu_{max.}$ (KBr) 1 670, 1 605, 1 515, and 825 cm^{-1}; δ (60 MHz) 3.26 (2 H, m, 4′-H), 3.73 and 3.77 (each 3 H, s, OMe), 4.03 (2 H, s, 8′-H_2), 4.11 (1 H, d, 6-Ha), 4.59 (1 H, dd, $J_{6.6}$ 12, $J_{6.6a}$ 3 Hz, 6-Hb), 4.90 (1 H, t, 6a-H), 5.37 (2 H, s, 7′-H_2), 5.43 (1 H, t, *J* 10 Hz, 5′-H), 6.41 (1 H, s, 4-H), 6.48 (1 H, d, *J* 9 Hz, 10-H), 6.73 (1 H, s, 1-H), and 7.80 (1 H, d, *J* 9 Hz, 11-H).

Deuteriolysis of Rotenone.—A standard Brown hydrogenator was employed, deuterium gas being generated from addition of sodium borodeuteride in deuterium oxide to deuterium chloride–deuterium oxide. The apparatus was connected to a vacuum/air manifold, permitting (*a*) evacuation of the system before generation of gas, and (*b*) maintenance of pressure balance during generation of gas. Natural rotenone (394 mg, 1 mmol) in ($^2H_5$)pyridine (1.5 cm^3) was deuteriated over palladium–barium sulphate (50 mg), until 1 mmol of deuterium was absorbed. The solution was filtered, diluted with water, and extracted with ether. The extracts were washed with aqueous M-sodium hydroxide, and the washings were immediately acidified. The precipitated phenols were collected in ether. After drying, the ethereal solution was evaporated and the residue separated by p.l.c. (four elutions, $CHCl_3$) into two main components. The higher R_F band afforded [4′-^{2}H]rot-2′-enonic acid (1e) (68 mg, 17%), m.p. 206 °C (from ethanol), *m/z* 397 (M^+). The i.r. spectrum showed a band at 862 cm^{-1} not present in that of the non-deuterio-compound. The band of lower R_F gave [2′-^{2}H]rot-3′- enonic acid [*cf.* (1m)] (65 mg, 14%), m.p. 186 °C (from ethanol), *m/z* 397 (M^+), with additional i.r. vibrations at 1 025 and 852 cm^{-1}.

9-O-*Methyl*-4′-*bromorot*-2′-*enonic Acid* (1h).—4′-Bromorot-2′-enonic acid (5.9 g) in acetone (100 cm^3) was treated with diazomethane (mol excess) in benzene, and the solution stirred at *ca.* 17 °C for 7 h. Surplus diazomethane was destroyed with acetic acid, and the residue obtained by evaporation was chromatographed on silica (Woelm). Elution with chloroform gave *compound* (1h) (4.3 g), m.p. 135—137 °C (from methanol–chloroform), $[\alpha]_D^{22.5}$ + 31° (*c* 0.03, $CHCl_3$) (Found: M^+, 488.086; C, 58.65; H, 5.4. $C_{24}H_{25}BrO_6$ requires *M*, 488.084; C, 58.9; H, 5.15%); $\lambda_{max.}$ 233 (4.37), 240 (4.31), 287 (4.26), and 321 nm (3.77); $\nu_{max.}$ 1 665, 1 600, 1 520, 1 470, 1 450, and 1 350 cm^{-1}; δ 1.92 (3 H, s, 5′-H_3), 3.34 (2 H, d, *J* 8 Hz, 1′-H_2), 3.77, 3.82, and 3.86 (each 3 H, s, OMe), 4.14 (1 H, d, *J* 12 Hz, 6-Ha), 4.60 (1 H, dd, *J* 4, 12 Hz, 6-Hb), 4.88 (1 H, br, t, *J* 4 Hz, 6a-H), 5.58 (1 H, t, *J* 8 Hz, 2′-H), 6.43 (1 H, s, 4-H), 6.56 (1 H, d, *J* 8 Hz, 10-H), 6.74 (1 H, s, 1-H), and 7.82 (1 H, d, *J* 8 Hz, 11-H).

A similar reaction using ether–methanol as solvent gave two products, separated by h.p.l.c. (chloroform–propan-2-ol, 100 : 1), one of which was the ether (1h). The second product was amorphous but gave only one peak on h.p.l.c. (solvent as above): it proved to be 9-O-*methyl*-4′-*methoxyrot*-2′-*enonic acid* (1i) (Found: M^+, 440.185. $C_{25}H_{28}O_7$ requires *M*, 440.184); $\lambda_{max.}$ 219infl (4.43), 232infl (4.28), 290 (4.22), and 320infl nm (3.77); $\nu_{max.}$ 1 675 and 1 605 cm^{-1}; *m/z* 440 (M^+, 23), 217 (5), 208 (10), 193 (10), 192 (100), 191 (18), and 177 (8).

4′-*Chlororot*-2′-*enonic Acid.*—Rotenone (1.0 g) in dry dichloromethane (6 cm^3) was added to boron trichloride (443 mm^3) in dichloromethane (6 cm^3) kept at −5 to −10 °C. The solution was stirred for 5 min at ambient temperature, then evaporated, and the residue was washed with dry methanol (10 cm^3). Crystallisation from methanol–dichloromethane gave the *chloro-derivative* (0.55 g), m.p. 159—160 °C, $[\alpha]_D^{23}$ + 26.5 (*c* 0.18, $CHCl_3$) (Found: C, 62.15; H, 5.2. $C_{23}H_{23}ClO_6{\cdot}CH_3OH$ requires C, 62.3; H, 5.85%); $\nu_{max.}$ (KBr) 1 660, 1 600, 1 450, and 1 355 cm^{-1}; δ 1.88 (3 H, s, 5′-H_3), 3.34 (2 H, d, *J* 8 Hz, 1′-H_2), 3.49 (3 H, s, HOCH_3), 3.76 and 3.81 (each 3 H, s, OMe), 3.94 (2 H, s, 4′-H_2), 4.11 (1 H, d, *J* 12 Hz, 6-Ha), 4.49 (1 H, dd, *J* 3.5 and 12 Hz, 6-Hb), 4.83 (1 H, 6a-H), 5.46 (1 H, t, *J* 8 Hz, 2′-H), 6.38 (1 H, s, 4-H), 6.42 (1 H, d, *J* 9 Hz, 10-H), 6.67 (1 H, s, 1-H), 7.33 (1 H, s, 9-OH), and 7.59 (1 H, d, *J* 9 Hz, 11-H).

2-*De*-O-*methyldihydrorotenone* (2j).—Dihydrorotenone (m.p. 208—209 °C; from hydrogenation of rotenone in ethyl acetate over platinum) (1.5 g) in dry dichloromethane (20 cm^3) was treated, at −5 to −10 °C and with stirring, with boron tribromide (450 mm^3) in dichloromethane (6 cm^3). After 5 min the solution was evaporated, and methanol and water were added in turn. The organic products were collected in chloroform and chromatographed on silica (Woelm). Elution with chloroform gave unchanged dihydrorotenone (0.42 g), m.p. and mixed m.p. 208—209 °C followed by *compound* (2j) (0.245 g), m.p. 165—166 °C (from ethanol), $[\alpha]_D^{20}$ − 66° (*c* 0.2, $CHCl_3$) (Found: M^+, 382.143; C, 69.05; H, 6.0. $C_{22}H_{22}O_6$ requires *M*, 382.142; C, 69.1; H, 5.8%); $\lambda_{max.}$ 238 (4.08), 243i (4.06), and 297 nm (4.22); $\lambda_{max.}$ (alkaline ethanol) 239 (4.01), 245infl (4.01), 299 (4.01), and 307 nm (4.00); $\nu_{max.}$ 3 555, 1 670, and 1 615 cm^{-1}; δ(CD_3-$COCD_3$) 1.06 and 1.12 (each 3 H, d, *J* 7 Hz, 7′-H_3, 8′-H_3), 1.94 (1 H, $J_{6',7'}$=$J_{6',8'}$=7 Hz, 6′-H), 3.03 (2 H, 4′-H_2), 3.79 (3 H, s, OMe), 3.83 (1 H, d, *J* 4 Hz, 12a-H), 4.22 (1 H, d, *J* 12 Hz, 6-H_a), 4.8 (2 H, 6-H_b, 5′-H), 5.07 (1 H, m, 6a-H), 6.40 (1 H, s, 4-H), 6.42 (1 H, d, *J* 9 Hz, 10-H), 6.64 (1 H, s, 1-H), 6.96 (1 H, s, OH), and 7.70 (1 H, d, *J* 9 Hz, 11-H).

De-O-*methylation of Isorotenone*—(6a*S*,12a*S*)-Isorotenone [m.p. 181—182.5 °C; prepared by sulphuric–acetic acid isomerisation of (−)-rotenone] [19] (1.5 g, 3.80 mmol) in dry dichloromethane (10 cm^3) was treated with boron tribromide (350 mm^3, 3.68 mmol) in dichloromethane at −5 to −10 °C, with stirring, for 3 min. After evaporation and quenching with methanol (10 cm^3), water (60 cm^3) was added. The organic products were collected in chloroform, and chromatographed on silica (Woelm) using gradient elution with chloroform and methanol (0→9%). Products were eluted in the following order.

(i) Isorotenone (330 mg), m.p. 181—182 °C (from chloroform–methanol).

(ii) 3-*De*-O-*methylisorotenone* (3l) (50 mg), m.p. 174—175 °C (from ethanol), $[\alpha]_D^{26}$ − 66° (*c* 0.2, $CHCl_3$) (Found: M^+, 380.127; C, 69.5; H, 5.1. $C_{22}H_{20}O_6$ requires *M*, 380.126; C, 69.45; H, 5.3%); $\lambda_{max.}$ 243 (4.66), 247infl (4.64), 262infl (4.22), 280 (4.09), and 330 nm (3.78); $\lambda_{max.}$ (alkaline ethanol) 245 (4.59), 282 (4.20), and 336 nm (3.93); $\nu_{max.}$ 3 350, 1 673, 1 615, 1 545, and 1 470 cm^{-1}; δ(CD_3COCD_3) 1.33 (6 H, d, *J* 7 Hz, 7′-H_3, 8′-H_3), 3.05 (1 H, septet, *J* 7 Hz, 6′-H), 3.68 (3 H, s, OMe), 3.97 (1 H, d, *J* 4 Hz, 12a-H), 4.30 (1 H, d, *J* 12 Hz, 6-Ha), 4.65 (1 H, dd, *J* 3.5 and 12 Hz, 6-H_b), 5.24 (1 H, 6a-H), 6.38 (1 H, s, 4-H), 6.55 (1 H, s, 4′-H), 6.69 (1 H, s, 1-H), 7.11 (1 H, d, *J* 9 Hz, 10-H), 7.50 (1 H, s, OH), and 7.77 (1 H, d, *J* 9 Hz, 11-H).

On treatment with acetic anhydride-pyridine at room

temperature (1 h), 3-de-*O*-methylisorotenone gave the *acetate* (9l), m.p. 177—178 °C, after chromatography on silica (chloroform) and crystallisation from ethanol; $[\alpha]_D^{26}$ − 78° (*c* 0.2, $CHCl_3$) (Found: M^+, 422.138; C, 68.5; H, 5.45. $C_{24}H_{22}O_7$ requires *M*, 422.137; C, 68.25; H, 5.25%); $\nu_{max.}$ (KBr) 1 760 and 1 680 cm^{-1}; δ(CD_3COCD_3) 2.21 (3 H, s, OAc), 6.61 (1 H, s, 4-H), and 6.75 (1 H, s, 1-H).

(iii) 2-*De*-O-*methylisorotenone* (2l) (540 mg), m.p. 130.5—131 °C (from benzene) (as a benzene solvate), $[\alpha]_D^{26}$ + 48° (*c* 0.27, $CHCl_3$) (Found: M^+, 380.125; C, 71.75; H, 5.5. ($C_{22}H_{20}O_6 \cdot 0.5C_6H_6$ requires *M*, 380.126; C, 71.6; H, 5.55%); $\lambda_{max.}$ 243 (4.64), 247infl (4.62), 262infl (4.18), 279 (4.03), 300infl (3.89), and 328 nm (3.70); $\lambda_{max.}$ (alkaline ethanol) 243 (4.54), 249infl (4.50), 260infl (4.19), 281 (4.11), and 310infl nm (3.81); $\nu_{max.}$ 3 557, 1 675, 1 618, 1 595, and 1 510 cm^{-1}; δ (CD_3COCD_3) 1.32 (6 H, d, *J* 7 Hz, 7′-H_3, 8′-H_3), 3.05 (1 H, septet, *J* 7 Hz, 6′H), 3.74 (3 H, s, OMe), 3.95 (1 H, d, *J* 4 Hz, 12a-H), 4.30 (1 H, d, *J* 12 Hz, 6-Ha), 4.66 (1 H, dd, *J* 3.5 and 12 Hz, 6-Hb), 5.22 (1 H, 6a-H), 6.42 (1 H, s, 4-H), 6.55 (1 H, s, 4′-H), 6.62 (1 H, s, 1-H), 7.07 (1 H, s, OH), 7.11 (1 H, d, *J* 8 Hz, 10-H), 7.35 (3 H, s, $0.5C_6H_6$), and 7.74 (1 H, d, *J* 8 Hz, 11-H). It formed an O-*acetate* (8l), when treated as above, m.p. 146—147 °C (prisms) or 164.5—165 °C (needles) (from di-isopropyl ether), $[\alpha]_D^{24}$ − 3° (*c* 0.25, $CHCl_3$) (Found: M^+, 422.138; C, 68.35; H, 5.3. $C_{24}H_{22}O_7$ requires *M*, 422.137; C, 68.25; H, 5.25%); $\nu_{max.}$ (KBr) 1 760 and 1 680 cm^{-1}; δ(CD_3COCD_3) 2.17 (3 H, s, OAc), 6.58 (1 H, s, 4-H), and 6.80 (1 H, s, 1-H).

(iv) 2,3-*Dide*-O-*methylisorotenone* (4l) (440 mg), m.p. 210 °C (decomp.) (from ethanol), $[\alpha]_D^{20}$ − 22° (*c* 0.13, Me_2CO) (Found: M^+, 366.110; C, 68.6; H, 5.2. $C_{21}H_{18}O_6$ requires *M*, 366.110; C, 68.85; H, 4.95%); $\lambda_{max.}$ 243 (4.70), 247infl (4.67), 261infl (4.19), 278 (4.01), 299infl (4.81), and 327 nm (3.54); $\nu_{max.}$ (KBr) 1 660, 1 605, 1 590, and 1 525 cm^{-1}; δ (CD_3COCD_3), 1.32 (6 H, d, *J* 7 Hz, 7′-H_3 and 8′-H_3), 3.05 (1 H, septet, *J* 7 Hz, 6′-H), 3.92 (1 H, d, *J* 4 Hz, 12a-H), 4.27 (1 H, d, *J* 12 Hz, 6-Ha), 4.63 (1 H, dd, *J* 3.5 and 12 Hz, 6-Hb), 6.38 (1 H, d, 4-H), 6.54 (1 H, s, 4′-H), 6.61 (1 H, s, 1-H), 7.12 (1 H, d, *J* 9 Hz, 10-H), and 7.77 (1 H, d, *J* 9 Hz, 11-H). This product predominated when (−)-isorotenone was treated with 2 mol equiv. of boron tribromide.

(±)-2,3-*Dide*-O-*methylisorotenone*, *Rac*-(4l).—(±)-Isorotenone [from base catalysed racemisation of (−)-isorotenone] [19] (8 g) was treated with boron tribromide (2 cm^3) in the above manner. Product isolation in the same fashion afforded the racemic *compound* (4l), crystallising from acetone–ethanol, m.p. 210 °C (decomp.) (Found: C, 68.55; H, 4.9. $C_{21}H_{18}O_6$ requires C, 68.85; H, 4.95%). It gave a *diacetate*, rac-(10l), m.p. 196—198 °C (from ethanol–chloroform), on treatment with acetic anhydride–pyridine as above (Found: M^+, 450.134; C, 66.9; H, 5.05. $C_{25}H_{22}O_8$ requires *M*, 450.132; C, 66.5; H, 4.95%). When the acetylation was allowed to continue for 4 h, the major product (isolated by p.l.c. on silica HF_{254}, chloroform–propan-2-ol, 25 : 1) was (±)-2,3-*di*-O-*acetyl*-2,3-*dide*-O-*methylisorotenone enol acetate*, (23) (21%), m.p. 210—215 °C (from chloroform–ethanol) (Found: M^+, 492.145; C, 65.6; H, 4.7%. $C_{27}H_{24}O_9$ requires *M*, 492.142; C, 65.85; H, 4.9%); $\lambda_{max.}$ 255 (4.51), 263 (4.45), 274 (4.38), 298infl (3.85), 309 (4.00), 320infl (4.05), 335infl (4.22), 352 (4.28), and 367-infl nm (4.19), $\nu_{max.}$ 1 786 cm^{-1}; δ 1.33 (6 H, d, *J* 7 Hz, 7′-H_3, 8′-H_3), 2.29 (6 H, s, 2 × OAc), 2.41 (3 H, s, OAc), 3.01 (1 H, septet, *J* 7 Hz, 6′-H), 4.5 (2 H, m, 6-H_2), 5.5 (1 H, m, 6a-H), 6.40 (1 H, s, 4′-H), 6.76 (1 H, s, 4-H), 6.93 (1 H, d, *J* 8.5 Hz, 10-H), 7.00 (1 H, d, *J* 8.5 Hz, 11-H), and 7.78 (1 H, s, 1-H).

Methylation of (6aS,12aS)-2,3-*Dide*-O-*methylisorotenone*.—Didemethylisorotenone (0.4 g) in acetone (25 cm^3) was treated with ethereal diazomethane, with t.l.c. monitoring using authentic samples of methyl ethers for comparison. When substantial monomethylation had taken place, the solvents were evaporated off and the product was separated on a silica gel column [chloroform–methanol (0→9%)] to yield 3-de-*O*-methylisorotenone (3l) (120 mg) and 2-de-*O*-methylisorotenone (2l) (150 mg), both with u.v. and ^1H n.m.r. spectra identical with those of authentic specimens.

2-*De*-O-*methylisorotenone from* 2-*De*-O-*methylrotenone*.—2-De-*O*-methylrotenone (45 mg) was stirred in acetic acid–sulphuric acid (4 : 3; 2 cm^3) for 1 h at 18 °C. Addition of water precipitated a solid which was collected, dried, and recrystallised from benzene to yield 2-de-*O*-methylisorotenone (30 mg), m.p. and mixed m.p. with authentic material 130—131 °C, and with i.r. and ^1H n.m.r. spectra identical with authentic spectra.

Treatment of (−)-*Rotenone with Methionine and Methanesulphonic Acid*.—(−)-Rotenone (250 mg), (±)-methionine (280 mg), and methanesulphonic acid (1.65 cm^3) were stirred together at ambient temperature for 22 h. The precipitate formed on dilution with water was collected, washed, dried, and extracted with acetone. The extracts were evaporated and the residue was separated by p.l.c. (chloroform–propan-2-ol, 25 : 1), to yield 2-de-*O*-methylisorotenone, m.p. 130.5—131.5 °C (from benzene), and 2,3-dide-*O*-methylisorotenone, m.p. 206—209 °C (decomp.) (from ethanol). Both products were identified by spectroscopic comparison with authentic samples.

Treatment of (−)-*Rotenone with Lithium Thio-t-butoxide*.—To a solution of (−)-rotenone (1.0 g, 2.54 mmol) in hexamethylphosphoramide (5 cm^3) was added lithium thio-t-butoxide in the same solvent (prepared [12] from lithium hydride and 2-methylpropanethiol) (3.5 cm^3 of 1.36 M-solution; 4.76 mmol). The mixture was stirred at 70 °C for 5 h and then poured into aqueous ammonium chloride. The product was extracted with chloroform, and the extracts were washed with dilute hydrochloric acid and water, dried, and evaporated. Chromatography of the residue on silica (Woelm) with chloroform elution gave a *sulphide* thought to be (24) as a non-crystalline mixture of diastereoisomers (Found: M^+, 484.193. $C_{29}H_{32}O_6S$ requires *M*, 484.192); $\nu_{max.}$ 1 635 and 1 610 cm^{-1}.

Reaction of (−)-*Rotenone with Trimethylsilyl Iodide*.—Trimethylsilyl iodide (0.85 cm^3, 5.4 mmol) was added to rotenone (1.97 g, 5.0 mmol) in dry chloroform, and the reaction mixture was stirred at 50 °C under argon for 20 h. It was then filtered and quenched with methanol (25 cm^3). After 20 h more, the solvents were evaporated off and the residue was chromatographed on silica (Woelm) using gradient elution (chloroform–methanol, 0→7%). A mixture of rotenone and 6a,12a-didehydrorotenone was eluted first, followed by 2-de-*O*-methylrotenone (516 mg, 27%) from benzene, identified by mixed m.p. and spectroscopic comparisons with an authentic sample.

[2-*methoxy*-$^2H_3$]*Rotenone*.—Ethereal diazomethane was transferred in a nitrogen stream to a deuterium oxide–tetrahydrofuran mixture at 0 °C. This solution was then added to 2-de-*O*-methylrotenone (40 mg) in tetrahydrofuran (0.2 cm^3) containing deuterium oxide (0.05 cm^3). When t.l.c. analysis showed the reaction to be complete, the solvents were replaced by chloroform and the solution was

filtered through a short silica column. Evaporation of the eluate and crystallisation of the residue from chloroform–ethanol gave [2-*methoxy*-$^2H_3$]rotenone, m.p. 161—163 °C (Found: M^+, 397.160. $C_{23}H_{19}D_3O_6$ requires M, 397.161); the ^1H n.m.r. spectrum was that of rotenone, but lacked one OMe resonance.

Base-catalysed Methylenation of 2,3-*Dide*-O-*methylisorotenone*.—Dide-*O*-methylisorotenone (200 mg, 0.55 mmol), methylene dibromide (0.4 cm³), and dimethylformamide (10 cm³) were stirred under nitrogen at 24 °C; sodium hydride (28 mg, 1.17mmol) was added in portions, and the reaction was continued for 10 h then quenched with water. The products were collected by ether extraction and separated by p.l.c. on silica HF_{254} (2 developments with hexane–ethyl acetate, 2 : 1). In this way were obtained (i) the (±)-*methylenedioxy-spiro-ketone* (14l) (66 mg, 32%), m.p. 125—128 °C (from methanol) (Found: M^+, 378.110; C, 69.65; H, 5.0. $C_{22}H_{18}O_6$ requires M, 378.110; C, 69.85; H, 4.8%), $\lambda_{max.}$ 241 (4.73), 246infl (4.66), 261 (4.08), 281 (3.98), 305 (4.01), and 331 nm (3.76); $\nu_{max.}$ (KBr) 1 710, 1 630, and 1 590 cm^{-1}; δ 1.38 (6 H, d, *J* 7 Hz, 7′-H_3, 8′-H_3), 2.08 (1 H, m, 6a-H_a),* 2.41 (1 H, m, 6a-H_b),* 3.10 (1 H, septet, 6′-H), *ca.* 4.4 (2 H, m, 6-H_2), 5.83 (2 H, s, OCH_2O), 6.24, 6.45, and 6.57 (each 1 H, s, 1,4, and 4′-H), 7.17 (1 H, d, *J* 9 Hz, 10-H), and 7.57 (1 H, d, *J* 9 Hz, 11-H); (ii) the *methylenedioxy-rotenoid* (12l) (8 mg, 4%), m.p. 182—183 °C (from methanol) (Found: M^+, 378.110; C, 70.0; H, 5.05. $C_{22}H_{18}O_6$ requires M, 378.110; C, 69.85; H, 4.8%); $\lambda_{max.}$ 242 (5.00), 261infl (4.49), 279 (4.36), 303 (4.29), and 325 nm (4.06); $\nu_{max.}$ (KBr) 1 680, 1 615, and 1 590 cm^{-1}; δ 1.32 (6 H, d, *J* 7 Hz, 7′-H_3, 8′-H_3), 3.08 (1 H, septet, *J* 7 Hz, 6′-H), 3.88 (1 H, d, *J* 4 Hz, 12a-H), 4.22 (1 H, d, *J* 12 Hz, 6-H_a), 4.89 (1 H, dd, *J* 3.5 and 12 Hz, 6-H_b), 5.03 (1 H, m, 6a-H), 5.83 (2 H, OCH_2O), 6.42 (1 H, s, 4-H), 6.52 (1 H, dd, *J* 1,1 Hz, 4′-H), 6.75 (1 H, d, *J* 1 Hz, 1-H), 7.05 (1 H, dd, *J* 1 and 9 Hz, 10-H), and 7.81 (1 H, d, *J* 9 Hz, 11-H).

Reduction of the Spiro-ketone (14l) *with Zinc*.—The ketone (200 mg) in ethanol (8 cm³) and aqueous 40% potassium hydroxide (2 cm³) was refluxed with zinc dust (800 mg) for 2 h. The filtered mixture was acidified and the precipitate collected in ether. The extracts were dried and evaporated. The residue crystallised from methanol to yield the *phenolic ketone* (15l) (120 mg), m.p. 85—86 °C (Found: M^+, 380.127; C, 69.45; H, 5.3. $C_{22}H_{20}O_6$ requires M, 380.126; C, 69.25; H, 5.3%); $\lambda_{max.}$ 235infl (4.45), 243 (4.59), 251 (4.57), 262infl (4.02), 284 (3.97), 300 (3.80), and 340 nm (3.40), $\lambda_{max.}$ (alkaline ethanol) 240infl (4.22), 244 (4.23), 264 (4.13), 277infl (3.94), 298infl (3.72), and 273 nm (3.66); $\nu_{max.}$ 3 050, 1 635, and 1 475 cm^{-1}; δ 1.39 (6 H, d, *J* 7 Hz, 7′-H_3, 8′-H_3), 2.27 (2 H, m, 6a-H_2), 3.04* (1 H, septet, 6′-H), 4.15 (2 H, m, 6-H_2), 4.71 (1 H, t, 12a-H), 5.80 (2 H, OCH_2O), 6.30, 6.39, and 6.55 (each 1 H, s, 1-H, 4-H, 4′-H), 6.73 and 6.95 (each 1 H, d, *J* 9 Hz, 10-H, 11-H), and 13.19 (1 H, s, OH).

Treatment of Phenolic Ring A *Rotenoids with Sodium Hydride*.—(*a*) Sodium hydride (56 mg) was added in portions, over 1 h, to dide-*O*-methylisorotenone (0.4 g) in dimethylformamide (20 cm³), and the mixture was stirred at room temperature for 5 h under nitrogen. It was then poured into sulphuric acid. The organic products were collected by ether extraction and acetylated using pyridine–acetic anhydride (3 : 2; 10 cm³) for 1 h at 18 °C. Isolation of the product in the usual manner gave the *spiro-diacetate* (16l) (0.35 g), m.p. 161.5—163 °C (from ethanol) (Found: C, 66.6; H, 5.05. $C_{25}H_{22}O_8$ requires C, 66.65; H, 4.95%); $\nu_{max.}$ 242 (4.68), 246infl (3.98), 283 (4.03), and 334 nm (3.68); $\nu_{max.}$ (KBr) 1 770, 1 706, 1 630, 1 590, and 1 500 cm^{-1}; δ 1.39 (6 H, d, *J* 8 Hz, 7′-H_3, 8′-H_3), 2.17 and 2.29 (each 3 H, s, OAc), 3.11 (1 H, septet, *J* 8 Hz, 6′-H), 4.54 (2 H, m, 6-H_2), 6.55 (1 H, s, 4′-H), 6.68 and 6.84 (each 1 H, s, 1-H, 4-H), 7.23 (1 H, d, *J* 9 Hz, 10-H), and 7.57 (1 H, d, *J* 9 Hz, 11-H).

(*b*) Under the conditions just described (6a*S*,12a*S*)-2-de-*O*-methylisorotenone (100 mg) with sodium hydride (6.3 mg) in dimethylformamide (5 cm³) gave (±)-2-*O*-*acetyl*-2-*de*-*O*-*methylisorotenone* (8l) (46 mg), m.p. 201.5—202 °C (from ethanol) (Found: M^+, 422.137; C, 68.4; H, 5.4. $C_{24}H_{22}O_7$ requires M, 422.137; C, 68.25; H, 5.25%). The ^1H n.m.r. spectrum was identical with that of the optically active specimen (above).

(*c*) In a similar reaction, (6a*S*,12a*S*)-3-de-*O*-methylisorotenone (73 mg) and sodium hydride (4.6 mg) in dimethylformamide (3 cm³) gave the (±)-3-*O*-*acetyl*-*de*-*O*-*methyl*-*spiro-ketone* (17l) (30 mg), m.p. 174—175 °C (from methanol) (Found: M^+, 422.134; C, 68.0; H, 5.3. $C_{24}H_{22}O_7$ requires M, 422.137; C, 68.25; H, 5.25%), $\nu_{max.}$ (KBr) 1 765, 1 710, 1 630, and 1 595 cm^{-1}; δ 1.36 (6 H, d, *J* 7 Hz, 7′-H_3, 8′-H_3), 2.07 (1 H, m, 6a-H_a), 2.27 (3 H, s, OAc), 2.42 (1 H, m, 6a-H_b), 3.09 (1 H, septet, *J* 7 Hz, 6′-H), 3.54 (3 H, s, OMe), 4.40 (2 H, m, 6-H_2), 6.29 and 6.63 (each 1 H, s, 1-H, 4-H), 6.52 (1 H, s, 4′-H), 7.22 (1 H, d, *J* 9 Hz, 10-H), and 7.55 (1 H, d, *J* 9 Hz, 11-H).

[8′-^{2}H]*Rotenone*.—8′-Bromorotenone (731 mg) in anhydrous tetrahydrofuran (20 cm³) was added dropwise, during 45 min, to a slurry of lithium aluminium deuteride (600 mg) in tetrahydrofuran (20 cm³) under dry nitrogen. The mixture was stirred for 3.5 h at room temperature, then sufficient aqueous ammonium chloride was added to destroy the excess of reagent. The product was filtered and the precipitate washed with tetrahydrofuran. The combined organic solutions were diluted with water and extracted with chloroform. The extracts were dried and evaporated to yield a white foam. The latter was dissolved in a mixture of acetone and benzene with a catalytic quantity of aluminium isopropoxide, and heated to 65 °C. T.l.c. analysis indicated partial conversion into rotenone after 3 h. The product was separated from starting material by p.l.c. (chloroform–propan-2-ol, 20 : 1) and the recovered alcohol was recycled twice more, to yield [8′-^{2}H]rotenone, m.p. 165 °C from ethyl acetate–light petroleum (77%). The u.v., mass, i.r., and ^1H n.m.r. spectra differed only from those of natural rotenone in characteristics relating to the [8′-^{2}H] substitution.

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* Rotenoid numbering retained.

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