Synthesis of *trans*-B/C-Rotenoids: X-Ray and NMR Data for *cis*- and *trans*-Forms of Isorotenone

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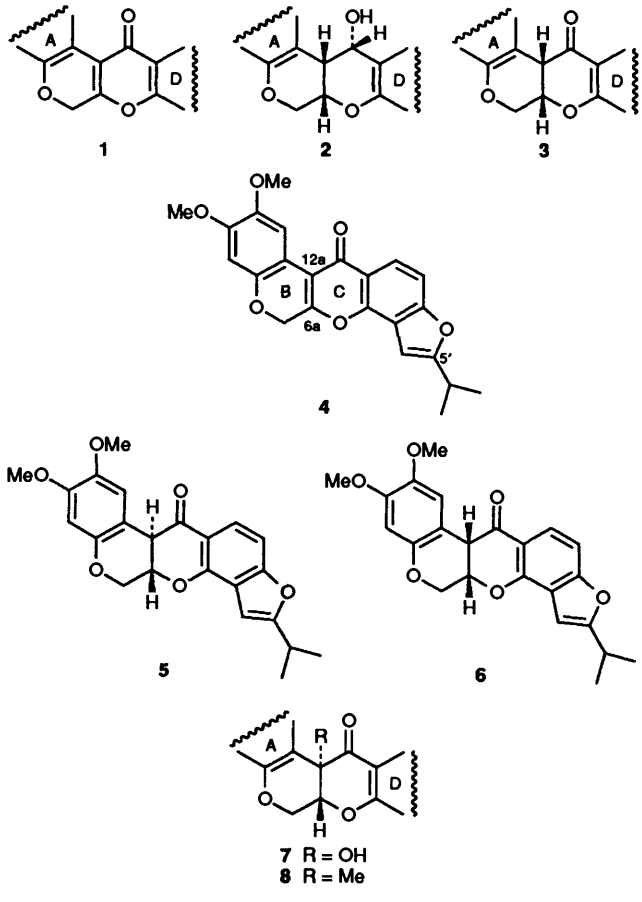
Reduction of 6a,12a-didehydrorotenoids with diisobutylaluminium hydride gives clean 1,4-reduction leading to unstable *trans*-B/C-fusions, not previously known for enolisable rotenoids: they are epimerised to stable *cis*-forms under acid conditions. Applied initially to isorotenone, the method is extended to *trans*-B/C-deguelin, α-toxicarol, the 'core' rotenoid structure and the 6a*S*,12a*R*,5'*R*- and 6a*R*,12a*S*,5'*R*-rotenone stereoisomers. ^{1}H and ^{13}C NMR data are compared for the *cis*- and *trans*-forms and the geometry and conformations of the isorotenones are compared by X-ray analysis, providing insight into the reasons for the instability of the *trans*-forms.

Reduction of the ridge-tile-like *cis*-isorotenone by sodium borohydride occurs from one face to give a *cis*-12α-hydroxy product, whilst the flatter *trans*-structure is attacked from both faces to give *trans*-12α- and 12β-hydroxy products.

A number of synthetic methods for rotenoids converge on a 6a,12a-didehydro compound **1**, requiring further reduction to species **3** to complete the synthesis. A standard procedure has been reduction with sodium borohydride which not only effects 1,4-reduction, but unwanted further reduction of the resulting carbonyl to give a 12-hydroxy derivative **2**. The latter must now be reoxidised by the Oppenauer method [1] or by using reagents such as manganese dioxide [2] or chromium trioxide. Apart from being lengthy, unless care is exercised some of these procedures can overoxidise back to the 6a,12a-didehydro level. Our initial aim was therefore to develop a more selective method for converting 6a,12a-didehydrorotenoids into stable *cis*-rotenoids of the natural series. In the course of the work, a new method for making the hitherto unknown thermodynamically unstable and enolisable *trans*-rotenoids emerged as well.



Results and Discussion

As test substrate for reductions, 6a,12a-didehydroisorotenone **4** was selected.[3,4] It is easily accessible from natural rotenone and the removal of the 5'-chirality of the latter avoids stereochemical complications. Various reagents were tried for the conversion into (±)-isorotenone, and of them, diisobutylaluminium hydride (DIBAL)[5] in toluene–tetrahydrofuran (THF) at −78 °C, followed by warming to room temperature, was found to achieve 1,4-reduction successfully without further 1,2-reduction of the carbonyl. The product of the reaction was unstable in the deuteriochloroform used for recording of the NMR spectra and a product converting into a second compound was indicated. Closer analysis of the spectra suggested that unstable (±)-*trans*-isorotenone **5** might be converting into stable (±)-*cis*-isorotenone **6**. Except for 12a-hydroxyrotenoids (rotenolones)[4] **7** and 12a-methylrotenoids[6] **8**, in which enolisation is blocked, the unstable *trans*-forms of the natural or synthetic enolisable rotenoids have remained unknown for more than 50 years.[7]

Chromatography [TLC; silica, development with hexane–ethyl acetate (7:3)] of the DIBAL reaction product, isolated after quenching with dry methanol, indicated that almost quantitative conversion from didehydroisorotenone into a single product had occurred. The reaction mixture was worked up by being poured into 1 mol dm^{-3} hydrochloric acid and immediately extracted with dichloromethane followed by shaking with calcium carbonate, evaporation, and drying. At this stage TLC revealed that small amounts of *cis*-isorotenone were present. It was possible to purify (±)-*trans*-isorotenone by flash chromatography on silica, and elution with hexane–ethyl acetate (4:1). Under these conditions, though some epimerisation took place on the column, *trans*-isorotenone, the faster eluting component, could be successfully separated from *cis*-material. Final purification was by crystallisation from methanol–acid-free chloroform, when (±)-*trans*-isorotenone **5**, m.p. 160–163 °C, was obtained as needles. Its m.p. was so close to that of (±)-*cis*-isorotenone (162–163 °C) that it was thought that epimerisation might have occurred during the determination. However, a mixed m.p. refuted this, as there was a marked depression.

Table 1 ^{1}H NMR data for pairs of *cis*- and *trans*-B/C-rotenoids: chemical shifts (δ_H)

Compound	Solvent	1-H	2-H	3-H	4-H	6^a-H	6^b-H	6a-H	4′-H	5′/6′-H	7′-H	8′-H	10-H	11-H	12a-H
cis-Isorotenone **6**	[$^2H_6$]acetone	6.71	3.63	3.73	6.46	4.35	4.69	5.28	6.58	3.07	1.31	1.32	7.13	7.78	4.00
trans-Isorotenone **5**	[$^2H_6$]acetone	7.64	3.81	3.79	6.48	4.62	4.31	4.88	6.65	3.14	1.37	1.37	7.24	7.81	4.24
cis-Deguelin	[$^2H_6$]acetone	6.73	3.74	3.64	6.44	4.28	4.66	5.13	6.65	5.71	1.44	1.35	6.45	7.69	3.90
trans-Deguelin **14**	[$^2H_6$]acetone	7.63	3.79	3.81	6.46	4.59	4.24	4.74	6.67	5.79	1.45	1.48	6.54	7.71	4.09
cis-Rotenone	[$^2H_6$]acetone	6.72	3.64	3.74	6.45	4.27	4.59	5.11	2.93 3.29	5.30	4.92 5.07	1.76	6.50	7.77	3.89
trans-Rotenone **12**	[$^2H_6$]acetone	7.62	3.78	3.80	6.46	4.53	4.22	4.74	3.02 3.42	5.42	4.95 5.11	1.79	6.60	7.79	4.09
cis-α-Toxicarol	[$^2H_6$]benzene	7.11	3.20	3.35	6.43	3.40	4.15	3.81	6.60	5.04	1.02	1.16	6.32	12.94	3.37
trans-α-Toxicarol **16**	[$^2H_6$]benzene	7.80	3.31	3.68	6.47	4.12	3.71	3.98	6.56	5.18	1.23	1.24	6.42	12.58	3.28
cis-Core	[$^2H_6$]benzene	7.44		multiplets		3.34	4.12	3.88					mult	8.10	3.45
trans-Core **15**	[$^2H_6$]benzene	8.27		multiplets		4.09	3.65	4.01					mult	8.06	3.29

NMR data were obtained in [$^2H_6$]acetone since solution in chloroform resulted in epimerisation to *cis*-isorotenone (depending on the acidity, this was noticeable by NMR and TLC after a few minutes). In our earlier work on rotenolones, isorotenolones, and their methyl ethers,[8] it was shown that the C-1 proton of the dimethoxylated ring A in the *trans*-B/C series was deshielded, being approximately in plane with the 12-carbonyl [$\delta(CHCl_3)$ 7.6–8.0]. On the other hand, in the *cis*-series the C-1 proton lies near the nodal shielding surface of the carbonyl [$\delta(CHCl_3)$ 6.4–6.8]. These large shifts provide a simple diagnostic tool. Whereas in (±)-*cis*-isorotenone 1-H resonated at δ([$^2H_6$]acetone) 6.71, the new (±)-*trans*-isomer from DIBAL reduction showed the resonance at δ([$^2H_6$]acetone) 7.64. Fig. 1 shows an expansion of NMR data for the four-proton system around the B/C ring junction of the *cis*-isomer and Fig. 2 shows comparative data for the *trans*-isomer. The coupling constant for the 6a,12a protons is 4.0 Hz for the former and 12.7 Hz for the latter. Full assignments for the ^1H and ^{13}C spectra,[9,10] assisted by COSY and hetero-COSY experiments, are to be found in Tables 1–3. IR data for the two isomers are very similar, with small differences in the fingerprint region. UV data also show only small differences in wavelength maxima and extinction coefficients.

In view of the importance of molecular shape to biochemical activity we have undertaken X-ray structure analyses of the two forms of (±)-isorotenone. The (±)-*trans*-isomer **5** crystallised in the triclinic system, contained two symmetry-related molecules per unit cell in space group $P\bar{1}$, and was refined to an *R*-value of 3.84%. It is shown in two projections in Figs. 3*a* and 3*b*. Considerable difficulties attended the growing of a suitable crystal of the (±)-*cis*-isomer **6** but success was eventually attained by the use of vapour diffusion with hexane as the solvent of low solubility and ethyl acetate as that of good solubility: the growing time was one month. This isomer also crystallised in the triclinic system, with four molecules per unit cell, of which there are two crystallographically independent molecules in space group $P\bar{1}$. The structure was refined to an *R*-value of 8.14% and one of the crystallographically independent molecules is shown in two projections in Figs. 4*a* and 4*b*.

The two crystallographically independent molecules in the (±)-*cis*-isorotenone **6** structure show very similar molecular geometry and conformation. Assessment of the major change in passing from the planar (±)-*trans*-structure to the (±)-*cis* one can be conveniently quantified in terms of the dihedral angle between the aromatic rings A and D: this is 158° for the *trans*- and 99° and 101° for the crystallographically independent *cis*-molecules. The results are also of interest in connection with the problem of the greater stability of the *cis*-series of rotenoids relative to the *trans*. It has been suggested earlier[11,12] that a major factor in this is the increased compression between the 12-carbonyl oxygen and the C-1 hydrogen in the *trans*- relative to the *cis*-forms. The X-ray structures show a non-bonded contact of 2.40 Å in the former and 2.48 and 2.56 Å for the two forms of the latter [in (−)-*cis*-bromodihydrorotenone it is 2.46 Å].[13] The strain of the shortened contact in the *trans*-structure is relieved by expansion of the sp^3 C-1a–C-12a–C-12 angle to 118.4°: it is 111.4° and 111.8° in the two *cis*-forms [and 111.4° in (−)-*cis*-bromodihydrorotenone].[13] In the *trans*-structure the sp^2 C-12a–C-12–carbonyl-O angle is expanded to 124.9°: it is 119.4° and 120.0° in the two *cis*-forms (and 117.2° in bromodihydrorotenone). However, the bond angles at C-1a and C-1 (hydrogen located and refined) do not deviate appreciably from the sp^2 hybridisation value of 120°.

The stereochemistry of the B/C-ring junction also influences the conformation of the B and C rings.[11,12] Each of these rings contains an aromatic bond and on this account would be expected to adopt the half-chair cyclohexene conformation with the two aromatic carbons and the two attached carbons coplanar: the remaining two atoms would be equally disposed above and below this mean plane. However, ring C also contains a carbonyl carbon (C-12) and if this is brought into full conjugation with the aromatic ring D (*i.e.*, planarity) an ideal

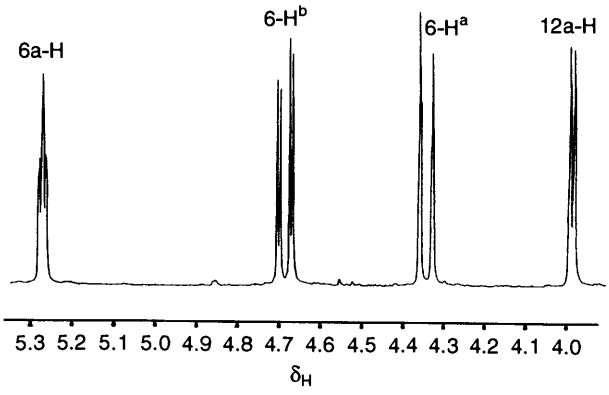


Fig. 1 Expansion of the ^1H NMR spectrum of (±)-*cis*-B/C-isorotenone **6**

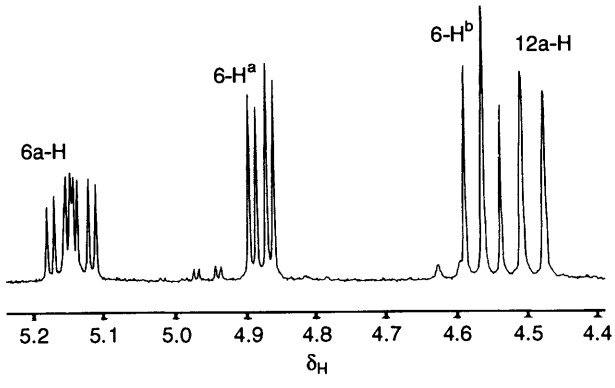


Fig. 2 Expansion of the ^1H NMR spectrum of (±)-*trans*-B/C-isorotenone **5**

Table 2 ^1H NMR data for pairs of *cis*- and *trans*-B/C-rotenoids: coupling constants (*J*/Hz).

Compound	Solvent	$J_{6\text{-H}^a,6a}$	$J_{6\text{-H}^b,6a}$	$J_{6\text{-H}^a,6\text{-H}^b}$	$J_{6a,12a}$	$J_{10,11}$
cis-Isorotenone **6**	[$^2H_6$]acetone	*a*	3.0	12.3	4.0	8.7
trans-Isorotenone **5**	[$^2H_6$]acetone	4.2	10.1	10.0	12.7	8.7
cis-Deguelin	[$^2H_6$]acetone	*a*	2.9	12.1	3.9	8.8
trans-Deguelin **14**	[$^2H_6$]acetone	3.9	10.0	9.8	12.6	8.5
cis-Rotenone	[$^2H_6$]acetone	*a*	2.9	12.3	3.9	8.5
trans-Rotenone **12**	[$^2H_6$]acetone	4.3	10.5	9.9	12.8	8.5
cis-α-Toxicarol	[$^2H_6$]benzene	*a*	3.1	12.1	3.1	*b*
trans-α-Toxicarol **16**	[$^2H_6$]benzene	4.1	10.5	9.6	12.5	*b*
cis-Core	[$^2H_6$]benzene	*a*	3.0	12.2	3.8	7.8
trans-Core **15**	[$^2H_6$]benzene	4.2	10.3	9.7	12.7	7.8

[a] Not observed. [b] Not applicable.

Table 3 ^{13}C NMR data for pairs of *cis*- and *trans*-B/C-rotenoids: chemical shifts δ_C

	[$^2H_6$]benzene		[$^2H_6$]acetone					
Carbon	*trans*-Core **15**	*cis*-Core	*trans*-Rotenone **12**	*cis*-Rotenone	*trans*-Deguelin **14**	*cis*-Deguelin	*trans*-Isorotenone **5**	*cis*-Isorotenone **6**
1	131.7	128.0	115.7	112.6	114.6	112.5	115.7	112.5
2	121.9	121.8	144.7	144.9	143.6	144.9	144.5	144.9
2-OMe			56.9	56.9	56.0	56.9	56.9	56.9
3	128.3	129.3	150.8	151.1	149.9	151.1	150.9	151.2
3-OMe			56.0	56.0	55.2	56.0	56.0	56.0
4	117.0	117.4	101.7	102.3	100.8	102.3	101.7	102.3
4a	154.5	154.1	149.5	149.0	148.6	149.0	149.5	149.1
6	66.5	66.0	67.4	67.0	66.5	67.0	67.4	67.0
6a	73.9	72.0	75.5	73.2	74.7	73.5	76.0	73.7
7a	160.3	160.9	158.2	158.9	156.2	157.8	155.2	156.1
8	117.5	118.0	114.0	113.8	109.2	109.9	117.6	114.5
9	135.5	136.2	167.4	167.9	159.0	160.5	160.2	160.5
10	121.4	121.6	105.0	105.1	110.9	111.7	106.6	106.6
11	127.8	127.8	130.3	130.3	128.1	129.0	123.7	123.6
11a	122.7	119.6	117.3	114.4	115.8	119.0	119.0	119.0
12	189.0	189.8	189.2	189.4	188.5	189.7	190.2	190.5
12a	46.7	45.5	46.8	45.1	45.8	44.9	46.9	45.3
12b	116.3	114.5	108.1	106.2	107.1	106.0	108.0	106.0
4′			31.9	31.8	115.2	116.2	98.3	98.5
5′			88.4	88.4	129.5	130.0	166.5	166.3
6′			144.7	144.6	77.5	78.4	28.9	28.1
7′			112.5	112.4	27.7	28.2	21.1	21.1
8′			17.2	17.2	27.3	28.6	21.1	21.1

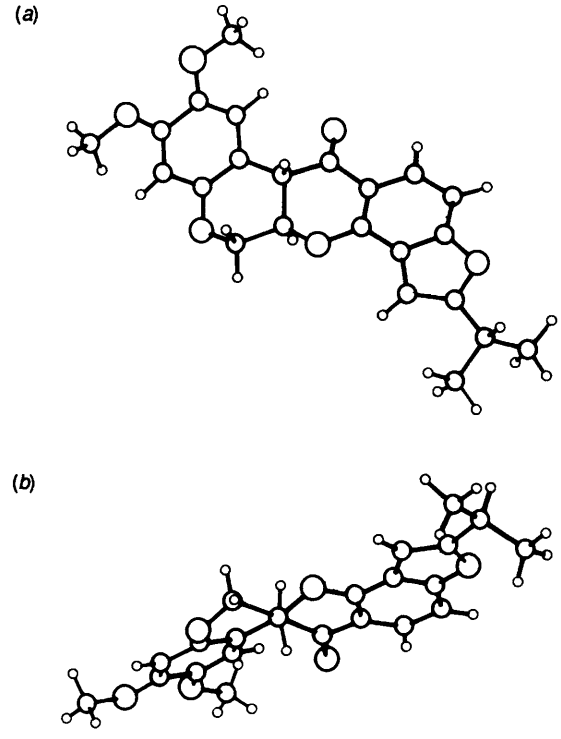


Fig. 3 X-Ray molecular structure of (±)-*trans*-B/C-isorotenone **5**. (*a*) Front view. (*b*) Side view.

model adopts the envelope conformation with five carbons in plane and only C-6a out of plane. All four X-ray structures discussed here show the B ring in half-chair conformation. In naturally derived (−)-bromodihydrorotenone ring C does adopt the idealised envelope conformation with only C-6a out of the plane formed by the other five carbon atoms by 0.53 Å. The atoms C-7a, C-11a, C-12 and C-12a display a torsion angle of only 1°. This brings the carbonyl into almost complete conjugation with the aromatic ring. The two independent (±)-*cis*-isorotenone structures also show C-6a strongly out of plane

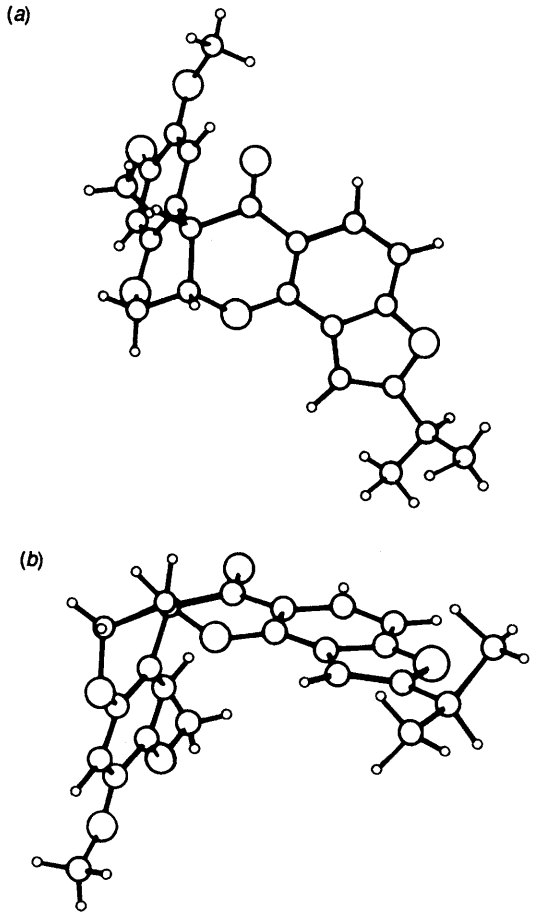


Fig. 4 X-Ray molecular structure of (±)-*cis*-B/C-isorotenone **6**. (*a*) Front view. (*b*) Side view.

(0.46 and 0.45 Å) but in these cases there is some deviation from ideality with C-12a out of plane to the extent of 0.17 and 0.18 Å. In the less stable *trans*-isorotenone structure the conformation approaches more closely the half-chair conformation: C-6a is

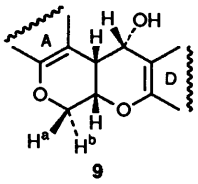
Table 4 Coupling constants (*J*/Hz) for 12-hydroxy isomers of the isorotenone series (*cf.* **5/6**)

	6-H^a,6-H^b	6-H^a,6a-H	6-H^b,6a-H	6-H^a,12a-H	6a-H,12a-H	12a-H,12-H
cis-B/C-α-12-OH- **9**	9.8	5.2	11.3	1.3	4.8	4.8
trans-B/C-α-12-OH- **10**	9.8	4.2	10.5	*a*	10.4	10.2
trans-B/C-β-12-OH- **11**	11.5	*a*	11.5	*a*	10.0	2.4

[a] Not observed.

0.46 Å above the mean plane when C-12a is 0.33 Å below the mean plane of the remaining four atoms of the ring. This brings the carbonyl group out of full conjugation with ring D. Thus the torsion angle C-7a–C-11a–C-12–C-12a is 10°.

From the point of view of biological testing, the enolisable carbonyl system can introduce ambiguities in a comparison of *cis*- and *trans*-B/C forms. Since it is known that there is substantial activity shown by the non-enolisable 12-hydroxy compounds in the mitochondrial particle test we have also made the (±)-12α-hydroxy-*cis*- and -*trans*-rotenoids, **9** and **10** respectively, for comparison.



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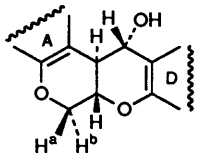

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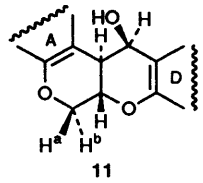
Treatment of (±)-*cis*-isorotenone with sodium borohydride in methanol gave the required (±)-12α-hydroxy-*cis*-isorotenoid **9** as a single isomer. The bent ridge-tile-like structure of the *cis*-compound results in the hydride ion being delivered from above, leading solely to the α-alcohol. This stereochemistry was confirmed by NMR analysis using specific decoupling experiments, and the coupling constants arrived at are shown in Table 4. The 4.8 Hz coupling between 6a-H and 12a-H confirms the *cis*-B/C fusion whilst the 12a-H, 12-H coupling, also of 4.8 Hz, indicates that 12-H is quasi-equatorial with the 12-hydroxy group quasi-axial.

In a similar fashion, (±)-*trans*-isorotenone was reduced with sodium borohydride. It was expected that the much flatter structure would allow delivery of hydride ion from both faces of the molecule, leading to a mixture of two epimers. TLC analysis indicated that two products had been formed, and these were separated by column chromatography: both products crystallised in needles from methanol–chloroform. The more rapidly eluted compound (m.p. 170–171 °C) was the (±)-12α-hydroxy-*trans*-rotenoid **10**, as demonstrated by the NMR data shown in Table 4. Thus the 10.4 Hz coupling between 6a-H and 12a-H confirms the *trans*-B/C fusion and the 12a-H, 12-H coupling of 10.2 Hz indicates that they are both axial or quasi-axial, which means that 12-OH is α (quasi-equatorial). The 6-H^a, 6a-H coupling of 4.2 Hz relates to their equatorial, axial arrangement. The large coupling of 10.5 Hz for 6-H^b, 6a-H is accounted for by their diaxial relationship.

The second fraction to be eluted had m.p. 201–203 °C but the NMR spectrum indicated that it was not a single compound, but a 2:1 mixture of (±)-12α-hydroxy-*cis*-isorotenoid **9** and (±)-12β-hydroxy-*trans*-isorotenoid **11**. The mixture behaved as a single spot on TLC, which co-eluted with pure (±)-12α-hydroxy-*cis*-isorotenoid **9**, and could not be resolved by further crystallisation. A mixed-m.p. determination with pure (±)-12α-hydroxy-*cis*-isorotenoid **9** gave a depression to 184 °C. This suggested that the unseparated product was not a simple mixture but had an ordered solid-state structure, though X-ray single crystal analysis has not yet been undertaken. The unwanted formation of compound **9** appears to be due to epimerisation of the *trans*-compound under the reaction conditions before reduction. Fortunately there is little overlap in the NMR signals of the two isomers, and with those for the (±)-12α-hydroxy-*cis*-isorotenoid **9** known, full assignments for the (±)-12β-hydroxy-*trans*-isorotenoid **11** can be made. The significant data are in Table 4. The 10.0 Hz coupling between 6a-H and 12a-H is consistent with the *trans*-B/C-fusion, and a coupling constant of 2.4 Hz between 12a-H and 12-H indicates their axial, equatorial-type relationship, placing the 12-hydroxy group β.

The study of the formation of *trans*-rotenoids was now enlarged from the isorotenone series, using 6a,12a-didehydro derivatives of rotenone, deguelin, α-toxicarol, and the synthetic rotenoid 'core', as examples. 6a,12a-Didehydrorotenone was obtained from natural (−)-rotenone by reflux with manganese dioxide in acetone;[2] it still retained a chiral centre (*R*) at C-5'. Reduction with DIBAL gave a pair of diastereoisomeric *trans*-rotenoids, 6a*S*,12a*R*,5'*R*- **12** and 6a*R*,12a*S*,5'*R*- **13**. These were partially separated by column chromatography, with monitoring of fractions by HPLC, and it was possible to obtain pure the isomer having the longer retention time by recrystallisation of partially purified fractions from methanol. This proved to be the 6a*S*,12a*R*,5'*R*- since epimerisation at C-12a by hydrogen chloride in methanolic chloroform gave a rotenone identical with natural rotenone (6a*S*,12a*S*,5'*R*) (acid conditions epimerise only C-12a, base conditions epimerise both C-12a and -6a through β-elimination).[11] The 6a*R*,12a*R*,5'*R*-isomer of rotenone is separable from its 6a*S*,12a*S*,5'*R*-isomer by HPLC so it can be identified with assurance.



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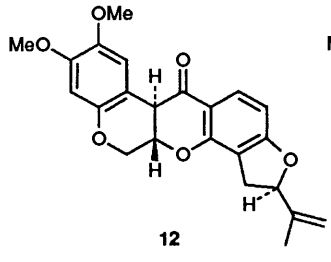

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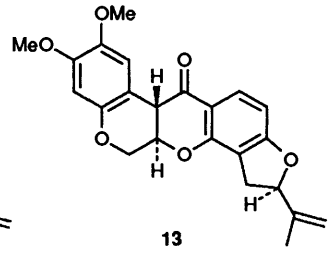

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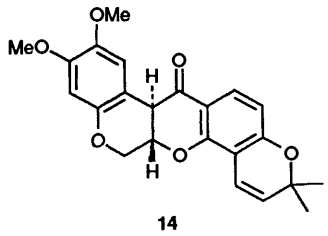
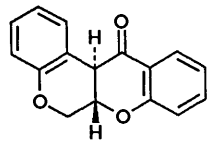
Naturally derived (*cis*)-deguelin[14] was similarly didehydrogenated at C-6a/12a with manganese dioxide in acetone, and on reduction with DIBAL in toluene–THF afforded (±)-*trans*-deguelin **14**. The compound was stable in acetone and was moderately stable to chromatography on silica with hexane–ethyl acetate elution. It soon epimerised to (±)-*cis*-deguelin in chloroform solution. The *trans*-rotenoid 'core' **15** (a chromanochromanone) was made from the synthetic 6a,12a-didehydrocompound[15] by reduction with DIBAL, but proved difficult to purify and isolate. Chromatography on 'flash' silica, and elution with hexane–ethyl acetate, resulted in extensive

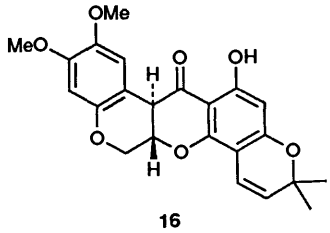
epimerisation and the pure *trans*-isomer could not be obtained in this way. It was, however, eventually isolated pure by direct crystallisation from ethyl acetate–methanol, immediately on work-up. The compound proved to be unstable in both chloroform and hexadeuterioacetone solution, complete epimerisation occurring within an hour. Fortunately it was stable in hexadeuteriobenzene, and full spectroscopic data are recorded in this solvent in the Tables. The greater stability of the *trans*-compounds having electron-donating methoxy groups in ring A may result from a retarding effect on enolisation at C-12a.



14



15



16

Finally, natural *cis*-α-toxicarol[16] was dehydrogenated by manganese dioxide, and the resulting 6a,12a-didehydro-α-toxicarol was reduced with DIBAL to form (±)-*trans*-α-toxicarol **16**. This too was more easily epimerised than its deguelin, isorotenone, and rotenone counterparts, and the ease of epimerisation may be associated with facilitation of C-12a enolisation by hydrogen bonding of the 11-hydroxy group to the 12-carbonyl. (±)-*trans*-α-Toxicarol **16** was just stable enough to be purified by chromatography on silica, with hexane–ethyl acetate as eluent, but it was unstable in acetone. The NMR spectra were recorded in [$^2H_6$]benzene in which it was quite stable.

Chemical-shift data for proton spectra are recorded in Table 1 for the five *trans*- and *cis*-pairs of rotenoids described above. For the reasons mentioned earlier, they are not all measured in the same solvent, though within any pair the solvent is the same. Coupling-constant data are given in Table 2 and ^{13}C data and assignments in Table 3. All the spectra have been recorded and analysed as for the *cis*- and *trans*-isorotenones above. The diagnostic shift (mentioned earlier) of the C-1 proton in going from the *cis*- to *trans*-fusion is clearly demonstrated in all five pairs of stereoisomers. There is also a smaller downfield shift of both the C-2 and C-3 methyl protons of the methoxy groups. Another general trend is that in going from a *cis*-fusion to a *trans*-fusion the shifts of the 6-H^a and 6-H^b protons are transposed. The 6-H^a proton (equatorial) is upfield in the *cis*-compounds, downfield in the *trans*-, with the 6-H^b proton (axial) downfield in the *cis*- and upfield in the *trans*-. This assignment is based on their coupling constants.

There are important changes in the coupling constants of the B- and C-rings. The 6a-H, 12a-H coupling constant for the ring fusion protons is ~4 Hz in the *cis*-isomers, whilst in the *trans*- it is increased to ~12 Hz. There is also a marked change in the coupling constants of the 6-H^a and 6-H^b protons. The *trans*-isomers have a value near 10 Hz, this being increased to ~12 Hz in the *cis*-isomers. In the *cis*-isomers no coupling is observed between 6-H^a (equatorial) and 6a-H, whilst in the *trans*-isomers it is ~4 Hz. For the *cis*-isomers the 6-H^b,6a-H coupling is ~3 Hz, whilst in the *trans*- it is ~10 Hz.

^{13}C Spectra show a downfield shift of 2–3 ppm for C-1 in going from *cis*- to *trans*-isomers. This is also observed for C-6a. C-12a shows a similar trend but its magnitude is nearer 1 ppm. The mass spectra of the *cis*- and *trans*-isomers are, unsurprisingly, essentially identical.

Experimental

For general experimental conditions see ref. 15. Optical rotations ($[\alpha]_D$) are given in units of 10^{-1} deg cm^2 g^{-1}.

(−)-cis-*Isorotenone* **6**.—This was prepared from natural rotenone by the sulfuric acid–acetic acid isomerisation procedure[3] in 61% yield, and had m.p. 175–176 °C (lit.,[3] 178–179 °C) as needles from chloroform–acetone (Found: M$^+$, 394.138. Calc. for $C_{23}H_{22}O_6$: M, 394.142); λ_{max}(EtOH)/nm 202, 243, 279 and 328 (ε 11 500, 11 000, 2800 and 900); ν_{max}(KBr)/cm^{-1} 1688s (CO), 1620s and 1602s (aromatics and C=C); *m/z* (EI, +ve) 394 (25%, M$^+$), 392 (4, M$^+$ − 2), 192 (100, $C_{11}H_{12}O_3^+$), 191 (20, $C_{11}H_{11}O_3^+$) and 177 (13, $C_{10}H_9O_3^+$). For other spectral data see Tables. The compound could be racemised by being refluxed in saturated ethanolic sodium acetate.

6a,12a-*Didehydroisorotenone* **4**.—Isorotenone (10 g, 25.4 mmol), ethanol (250 cm^3), benzene (25 cm^3), and anhydrous sodium acetate (15 g, 181 mmol) were heated together under reflux and a solution of iodine (5 g, 39.4 mmol) in ethanol (75 cm^3) was added dropwise, reflux being continued for 50 min after the addition. The product was cooled in ice and the yellow precipitate was filtered off. The latter was dissolved in hot chloroform, filtered to remove solid particles, and reprecipitated by addition of hot methanol. After cooling, bright yellow crystals of didehydroisorotenone **4** (4.93 g, 50%), m.p. 188–189 °C (lit.,[4] 189–191 °C) were collected; ν_{max}(KBr)/cm^{-1} 1640s (CO), 1615s, 1585s and 1500s (aromatics and C=C); δ_H(90 MHz; $CDCl_3$) 1.41 (6 H, d, *J* 8, 2 × Me), 3.15 (1 H, septet, *J* 8, 6′-H), 3.90 (3 H, s, OMe), 4.01 (3 H, s, OMe), 5.19 (2 H, s, 6-H$_2$), 6.57 (1 H, s, 4-H), 6.70 (1 H, s, 4′-H), 7.49 (1 H, d, *J* 9, 10-H), 8.15 (1 H, d, *J* 9, 11-H) and 8.55 (1 H, s, 1-H).

(±)-trans-*Isorotenone* **5**.—DIBAL in toluene (1 mol dm^{-3} solution; 0.673 cm^3, 0.673 mmol) was added dropwise to a solution of didehydroisorotenone **4** (100 mg, 0.255 mmol) in dry THF (5 cm^3) at −78 °C under nitrogen. After being stirred for 1 h at −78 °C the mixture was allowed to warm to room temperature and methanol (2 cm^3) was added dropwise. The mixture was stirred for 30 min at room temperature and was then poured into 1 mol dm^{-3} hydrochloric acid (5 cm^3) and extracted with dichloromethane. The extracts were washed successively with an aqueous suspension of calcium carbonate, water and brine, and were then dried ($MgSO_4$) and evaporated. The solid remaining was chromatographed on flash silica, and eluted with hexane–ethyl acetate (4:1), to give (±)-trans-*isorotenone* **5** (32 mg, 32%), m.p. 160–163 °C (from $CHCl_3$–MeOH) (Found: C, 70.05; H, 5.75%; M$^+$, 394.141. $C_{23}H_{22}O_6$ requires C, 70.05; H, 5.6%; M, 394.142); λ_{max}(EtOH)/nm 202, 241 and 286 (ε 13 150, 13 600 and 2500); ν_{max}(KBr)/cm^{-1} 1691s (CO), 1620s and 1595s (aromatic); *m/z* (EI, +ve) 394 (20%, M$^+$), 392 (11, M$^+$ − 2), 192 (100, $C_{11}H_{12}O_3^+$) and 177 (11, $C_{10}H_9O_3^+$). For NMR spectral data see Tables.

A small quantity of *trans*-isorotenone was dissolved in chloroform and monitored by TLC on silica gel plates, and development with hexane–ethyl acetate (7:3). The spot corresponding to *trans*-isorotenone **5** (R_f 0.37) began to be converted

into a spot corresponding to *cis*-isorotenone (R_f 0.20) after a few minutes, complete conversion taking ~ 12 h.

6a,12a-*Didehydrorotenone* (cf. **1**).—Natural (6a*S*,12a*S*,5′*R*)-(−)-*cis*-rotenone was extracted from Timbo resin[17] and had m.p. 165–166 °C (lit.,[17] 167–168 °C) (Found: M^+, 394.141. Calc. for $C_{23}H_{22}O_6$: M, 394.142); λ_{max}(EtOH)/nm 210, 236 and 293 (ε 35 700, 13 200 and 14 800); ν_{max}(KBr)/cm^{-1} 1725s (CO), 1610s and 1515s (aromatics).

Rotenone (10 g, 25.4 mmol) was refluxed with active manganese dioxide (50 g) in stirred acetone for 3 h. After filtration through a pad of Kieselguhr, the solvent was removed and the residue was crystallised from chloroform–methanol to give (5′*R*)-6a,12a-didehydrorotenone (2.4 g, 24%) as pale yellow needles, m.p. 223 °C (lit.,[2] 224 °C); ν_{max}(mull)/cm^{-1} 1634s (CO), 1605s (C=C) and 1505m (aromatic); δ_H(90 MHz; $CDCl_3$) 1.80 (3 H, s, 8′-H_3), 3.15 (1 H, dd, *J* 16 and 8, 4′-H^a), 3.52 (1 H, dd, *J* 16 and 10, 4′-H^b), 3.84 (3 H, s, OMe), 3.95 (3 H, s, OMe), 4.97 (3 H, br s, 6-H_2 and 7′-H), 5.13 (1 H, br s, 7′-H), 5.40 (1 H, dd, *J* 8 and 10, 5′-H), 6.52 (1 H, s, 4-H), 6.89 (1 H, d *J* 8, 10-H), 8.11 (1 H, d, *J* 8, 11-H) and 8.44 (1 H, s, 1-H).

DIBAL in toluene (1.0 mol dm^{-3}; 6.38 cm^3, 6.38 mmol) was added dropwise to a solution of (5′*R*)-6a,12a-didehydrorotenone (1.00 g, 2.55 mmol) in dry THF (50 cm^3) under nitrogen at −78 °C. The mixture was stirred for 1 h and then was allowed to warm to room temperature, when dry methanol (30 cm^3) was added and the mixture was stirred for a further hour before being poured into 1 mol dm^{-3} hydrochloric acid and extracted with dichloromethane. After being washed successively with aq. calcium carbonate suspension, water, and brine, the extracts were dried ($MgSO_4$) and evaporated. At this stage the product contained mixed diastereoisomers—(6a*S*,12a*R*,5′*R*)- (**12**) and (6a*R*,12a*S*,5′*R*)-(**13**)-rotenone—and these were separated on a flash silica column, with hexane–ethyl acetate (4:1) as eluent, with monitoring of fractions by HPLC on a μ-Porasil analytical column eluted with the same solvent mixture. Fractions 23–36 were combined and evaporated, and the product was crystallised from methanol to give (6a*S*,12a*R*,5′*R*)-trans-(+)-*rotenone* **12** (31 mg) as needles, m.p. 150–153 °C; $[\alpha]_D^{25}$ +261 (*c* 0.031, acetone) (Found: 70.15; H, 5.75%; M^+, 394.144. $C_{23}H_{22}O_6$ requires C, 70.05; H, 5.6%; M, 394.141); λ_{max}(EtOH)/nm 211, 235 and 290 (ε 49 000, 15 900 and 17 800); ν_{max}(KBr)/cm$^-$ 1696s (CO), 1620s and 1507s (aromatics); *m/z* (EI, +ve) 394 (35%, M^+), 392 (10, $M^+ - H_2$), 192 (100, $C_{11}H_{12}O_3^+$), 191 (22, $C_{11}H_{11}O_3^+$) and 177 (13, $C_{10}H_9O_3^+$).

A solution of (6a*S*,12a*R*,5′*R*)-*trans*-(+)-rotenone (6 mg) in methanol (3 cm^3)–chloroform (1 cm^3) was stirred with 3 drops of conc. hydrochloric acid under nitrogen for 2 h, then was poured into water and extracted with chloroform. The product was worked up, and examined by HPLC [μ-Porasil, elution with hexane–ethyl acetate (4:1), 3 cm^3 min^{-1}]. It was indistinguishable from (6a*S*,12a*S*,5′*R*)-*cis*-rotenone (retention time, co-injection). In the same system 'mutarotenone',[2] a mixture of (6a*S*,12a*S*,5′*R*)-*cis*- and (6a*R*,12a*R*,5′*R*)-*cis*-rotenone diastereoisomers, was separated as two distinct peaks.

(−)-cis-*Deguelin* (*the* cis-*Form of Compound* **14**).—4′-Bromorot-2′-enonic acid was prepared[18] in 36% yield from rotenone and boron tribromide: it had m.p. 150–152 °C from methanol (lit.,[18] 152–154 °C). By treatment of the bromo compound with sodium cyanoborohydride in dry hexamethylphosphoric triamide according to a literature procedure,[18] rot-2′-enonic acid was prepared in 35% yield, m.p. 201–202 °C (from MeOH) (lit.,[18] 204–206 °C).

Benzeneselenenyl chloride (939 mg, 4.91 mmol) was added to a solution of rot-2′-enonic acid (1.85 g, 4.67 mmol) in dry dichloromethane (60 cm^3) at −30 °C under nitrogen. The reaction was stirred (2 h) while warming to room temperature, and then the solvent was removed. The residue was dissolved in dry THF (50 cm^3), and the solution was cooled to 0 °C, treated with 30% hydrogen peroxide (1 cm^3), and stirred first at 0 °C for 1 h, then overnight at room temperature. After dilution with diethyl ether (50 cm^3), the solution was washed successively with 5% aq. sodium hydrogen carbonate and brine, and was then evaporated. The glass was chromatographed on dry column silica, and eluted with chloroform, to give (−)-*cis*-deguelin (1.08 g, 59%) as a glass[14] (Found: M^+, 394.142. Calc. for $C_{23}H_{22}O_6$: M, 394.142); λ_{max}(EtOH)/nm 203 and 271 (ε 70 600 and 60 300); ν_{max}(KBr)/cm^{-1} 1673s (CO), 1600s and 1578s (aromatics).

(±)-trans-*Deguelin* **14**.—(−)-*cis*-Deguelin (539 mg, 1.37 mmol) was refluxed with active manganese dioxide 2.5 g) in acetone for 3.5 h and, after cooling, the mixture was filtered through Kieselguhr, which was then washed with chloroform. The filtrates were evaporated, and the product was heated under reflux with 10% methanolic sulfuric acid (30 cm^3) for 40 min. On cooling, the precipitate was filtered off, and was washed with cold methanol to give 6a,12a-didehydrodeguelin (340 mg, 63%), m.p. 223–224 °C (lit.,[4] 230–231 °C); ν_{max}(KBr)/cm^{-1} 1640s (CO), 1600m and 1507s (aromatics); δ_H(90 MHz; $CDCl_3$) 1.50 (6 H, s, 7′- and 8′-H_3), 3.87 (3 H, s, OMe), 3.95 (3 H, s, OMe), 5.01 (2 H, s, 6-H_2), 5.72 (1 H, d, *J* 10.2, 5′-H), 6.55 (1 H, s, 4-H), 6.75 (1 H, d, *J* 10.2, 4′-H), 6.85 (1 H, d, *J* 8.8, 10-H), 8.03 (1 H, d, *J* 8.8, 11-H) and 8.45 (1 H, s, 1-H).

DIBAL in toluene (1.0 mol dm^{-3}; 0.730 cm^3, 0.730 mmol) was added dropwise to a solution of 6a,12a-didehydrodeguelin (114 mg, 0.291 mmol) in dry THF (8 cm^3) under nitrogen at −78 °C and the mixture was stirred at −78 °C (1 h) and was then allowed to warm to room temperature. Dry methanol (3 cm^3) was added and the solution was stirred for 1 h and was then poured into 1 mol dm^{-3} hydrochloric acid (20 cm^3) and extracted with chloroform. The extracts were washed with aq. calcium carbonate suspension, and evaporated, as described earlier. Chromatography on flash silica, and elution with hexane–ethyl acetate (3:2), gave (±)-trans-*deguelin* **14** (44 mg, 38%), m.p. 161–164 °C, as prisms from methanol (Found: C, 69.9; H, 5.75%; M^+, 394.142. $C_{23}H_{22}O_6$ requires C, 70.05; H, 5.6%; M, 394.142); λ_{max}(EtOH)/nm 203 and 267 (ε 30 400 and 27 000); ν_{max}(KBr)/cm^{-1} 1695s (CO), 1600s and 1518s (aromatics); *m/z* (EI, +ve) 394 (28%, M^+), 392 (22, $M^+ - 2$), 192 (100, $C_{11}H_{10}O_3^+$) and 177 (14, $C_{10}H_9O_3^+$).

trans-Deguelin was dissolved in chloroform at 20 °C and was monitored on silica gel TLC plates developed with hexane–ethyl acetate (2:3). The spot corresponding to *trans*-deguelin (R_f 0.43) began to be converted into a spot corresponding to *cis*-deguelin (R_f 0.26) after a few minutes, complete conversion taking *ca.* 12 h. No conversion was observed when the chloroform was replaced by acetone.

(±)-trans-6a,12a-*Dihydrorotoxen*-12(6H)-*one* **15**.—A solution of rotoxen-12(6*H*)-one[15] (152 mg, 0.608 mmol) in dry THF (6 cm^3) at −78 °C under nitrogen was treated with DIBAL in toluene (1.0 mol dm^{-3}; 1.52 cm^3, 1.52 mmol) and was then stirred at −78 °C for 1 h 15 min, then was allowed to warm to room temperature. Methanol (2 cm^3) was added and the reaction mixture was worked up as for the previous DIBAL reductions. (±)-trans-6a,12a-*Dihydrorotoxen*-12(6H)-*one* **15** (14 mg) was crystallised from ethyl acetate as needles, m.p. 116–118 °C (Found: C, 76.0; H, 4.75%; M^+, 252.078. $C_{16}H_{12}O_3$ requires C, 76.2; H, 4.8%; M, 252.079); λ_{max}(EtOH)/nm 201, 213, 249 and 314 (ε 36 540, 34 920, 9900 and 3490); ν_{max}(KBr)/cm^{-1} 1695s (CO), 1608s and 1530m (aromatics); *m/z* (EI +ve) 252 (17%, M^+), 250 (7, $M^+ - 2$), 132 (67, $C_9H_8O^+$) and 131 (100, $C_9H_7O^+$). For NMR data see Tables.

A small quantity of *trans*-**15** was dissolved in chloroform at

room temperature and monitored by TLC on silica gel plates developed with hexane–ethyl acetate (7:3). The spot corresponding to the *trans*-compound (R_f 0.54) was completely converted into a spot corresponding to the *cis*-isomer (R_f 0.38) within 5 min. When acetone replaced chloroform in the experiment, complete conversion was again observed in 5 min.

(±)-trans-α-*Toxicarol* **16**.—*cis*-α-Toxicarol (400 mg, 0.967 mmol) of natural origin was refluxed with manganese dioxide (2.0 g) in acetone (25 cm^3) for 3.5 h, then the mixture was cooled, filtered, and worked up as above. 6a,12a-Didehydro-α-toxicarol (221 mg, 55%) was crystallised from chloroform–methanol as yellow needles, m.p. 215–216 °C (lit.,[11] 230–231 °C); δ_H(90 MHz; $CDCl_3$) 1.47 (3 H, s, 7′-H_3), 1.54 (3 H, s, 8′-H_3), 3.88 (6 H, s, OMe), 5.00 (2 H, s, 6-H_2), 5.59 (1 H, d, *J* 10.1, 5′-H), 6.30 (1 H, s, 2 × 10-H), 6.57 (1 H, s, 4-H), 6.64 (1 H, d, *J* 10.1, 4′-H), 8.29 (1 H, s, 1-H) and 12.99 (1 H, s, 11-OH).

DIBAL in toluene (1.0 mol dm^{-3}; 1.10 cm^3, 1.10 mmol) was added dropwise to a solution of the didehydro compound (180 mg, 0.441 mmol) in dry THF (8 cm^3) under nitrogen at −78 °C. The reaction mixture was stirred (1 h) and worked up as before with chromatography on flash column silica, with hexane–ethyl acetate (4:1) as eluent, to give (±)-trans-α-*toxicarol* **16** (45 mg, 25%), m.p. 169–171 °C (Found: C, 67.0; H, 5.6%; M^+, 410.136. $C_{23}H_{22}O_7$ requires C, 67.3; H, 5.4%; M, 410.137); λ_{max}(EtOH)/nm 204, 229, 236, 270, 295 and 357 (ε 31 400, 12 700, 12 900, 28 000, 13 000, 2900); *m/z* (EI, +ve) 410 (63%, M^+), 192 (100, $C_{11}H_{12}O_3^+$) and 177 (17, $C_{10}H_9O_3^+$). For NMR data see Tables.

trans-α-Toxicarol **16** was dissolved in chloroform at room temperature and monitored by TLC on silica plates developed with hexane–ethyl acetate (4:1). The spot corresponding to *trans*-α-toxicarol (R_f 0.29) was completely converted into a spot corresponding to *cis*-α-toxicarol (R_f 0.16) within 5 min. The use of acetone gave similar results.

The 12α-*Hydroxy*-cis-*isorotenoid* **9**: *Reduction of* (±)-cis-*Isorotenone with Sodium Borohydride*.—Sodium borohydride (50 mg, 1.316 mmol) was added in portions to a solution of (±)-*cis*-isorotenone (200 mg, 0.505 mmol) in methanol (5 cm^3) at room temperature over a period of 10 min and the mixture was then stirred (30 min). Water (5 cm^3) was added, most of the methanol was removed, and the precipitate was filtered off, and crystallised from chloroform–methanol to give the (±)-12α-hydroxy-*cis*-isorotenoid **9** (158 mg, 79%) as prisms, m.p. 191–192 °C (lit.,[11] 193 °C) (Found: C, 69.55; H, 6.15%; M^+, 396.155. Calc. for $C_{23}H_{24}O_6$: C, 69.7; H, 6.1%; M, 396.157); λ_{max}(EtOH)/nm 213, 252, 259 and 291 (ε 28 500, 15 000, 16 500 and 7400); ν_{max}(KBr)/cm^{-1} 3480s (OH), 1600, 1510 and 1480 (aromatics); δ_H(400 MHz; $CDCl_3$) 1.34 (6 H, d, *J* 6.9, 2 × Me), 1.95 (1 H, d, *J* 2.1, OH), 3.05 (1 H, septet, *J* 6.9, 6′-H), 3.43 (1 H, dd, *J* 4.8 and 4.8, 12a-H), 3.83 (3 H, s, OMe), 3.85 (3 H, s, OMe), 4.27 (1 H, ddd, *J* 9.8, 5.2 and 1.3, 6-H^a), 4.66 (1 H, dd, *J* 11.3 and 9.8, 6-H^b), 4.91 (1 H, ddd, *J* 11.3, 5.2 and 4.8, 6a-H), 5.01 (1 H, dd, *J* 4.8 and 2.1, 12-H), 6.46 (2 H, s, 4′- and 4-H), 6.72 (1 H, s, 1-H), 7.04 (1 H, d, *J* 7.8, 10-H) and 7.06 (1 H, d, *J* 7.8, 11-H); δ_C(100 MHz; $CDCl_3$) 21.0 (CH_3, C-7′, -8′), 28.2 (CH, C-6′), 38.3 (CH, C-12a), 55.9 (CH_3, OMe), 56.6 (CH_3, OMe), 64.9 (CH_2, C-6), 66.4 (CH, C-6a), 69.2 (CH, C-12), 97.1 (CH, C-4′), 100.8 (CH, C-4), 104.5 (CH, C-10), 108.9 (C, C-12b), 111.7 (CH, C-1), 113.8 (C, C-8), 118.2 (C, C-11a), 125.3 (CH, C-11), 143.9 (C, C-2), 145.5 (C, C-4a), 149.5 (C, C-3), 149.8 (C, C-7a), 156.2 (C, C-9) and 164.4 (C, C-5′); *m/z* (EI, +ve) 396 (4%, M^+), 378 (100, $M^+ - H_2O$), 363 (26, $C_{22}H_{19}O_5^+$) and 192 (40, $C_{11}H_{12}O_3^+$).

The 12α- *and* 12β-*Hydroxy*-trans-*isorotenoids*, **10** *and* **11**: *Reduction of* (±)-trans-*Isorotenone with Sodium Borohydride*.—Sodium borohydride (50 mg, 1.316 mmol) was added portionwise to a stirred suspension of (±)-*trans*-isorotenone **5** (200 mg, 0.505 mmol) in methanol (5 cm^3) at room temperature. After completion of the addition, the mixture was stirred for 30 min and then worked up as above, with chromatography of the product on flash column silica and elution with hexane–ethyl acetate (7:3). The first eluted compound was the (±)-12α-*hydroxy*-trans-*isorotenoid* **10** (49 mg, 25%) as needles from chloroform–methanol, m.p. 170–172 °C (Found: C, 69.55; H, 6.1%; M^+, 396.154. $C_{23}H_{24}O_6$ requires C, 69.7; H, 6.1%; M, 396.157); λ_{max}(EtOH)/nm 211, 250, 258 and 290 (ε 22 600, 12 800, 11 600 and 4800); ν_{max}(KBr)/cm^{-1} 3290s br (OH), 1620m and 1510s (aromatics); δ_H(400 MHz; $CDCl_3$) 1.33 (6 H, d, *J* 6.9, 2 × Me), 2.09 (1 H, d, *J* 10.2, OH), 3.05 (1 H, septet, *J* 6.9, 6′-H), 3.09 (1 H, dd, *J* 10.4 and 10.2, 12a-H), 3.83 (3 H, s, OMe), 3.88 (3 H, s, OMe), 4.09 (1, H, dd, *J* 10.5 and 9.8, 6-H^b), 4.29 (1 H, ddd, *J* 10.5, 10.4 and 4.2, 6a-H), 4.54 (1 H, dd, *J* 9.8 and 4.2, 6-H^a), 4.94 (1 H, dd, *J* 10.2 and 10.2, 12-H), 6.42 (2 H, s, 4′- and 4-H), 7.10 (1 H, d, *J* 8.6, 10-H), 7.37 (1 H, d, *J* 8.6, 11-H) and 7.71 (1 H, s, 1-H); δ_C(100 MHz; $CDCl_3$) 21.0 (CH_3, C-7′ and -8′), 28.3 (CH, C-6′), 43.8 (CH, C-12a), 55.9 (CH_3, OMe), 56.5 (CH_3, OMe), 67.2 (CH_2, C-6), 70.9 (CH, C-6a), 71.8 (CH, C-12), 96.9 (CH, C-4′), 100.4 (CH, C-4), 105.3 (CH, C-10), 111.4 (CH, C-1), 112.8 (C, C-8), 118.0 (C, C-11a), 119.8 (C, C-12b), 122.4 (CH, C-11), 144.0 (C, C-2), 146.8 (C, C-4a), 148.1 (C, C-3), 149.3 (C, C-7a), 155.6 (C, C-9) and 164.9 (C, C-5′); *m/z* (EI +ve) 396 (22%, M^+), 378 (100, $M^+ - H_2O$), 363 (28, $C_{22}H_{19}O_5^+$), 192 (51, $C_{11}H_{12}O_3^+$) and 179 (55, $C_{10}H_{11}O_3^+$).

The second fraction to be eluted was crystallised from chloroform–methanol and was recognised spectroscopically to be a ~2:1 mixture (63 mg, 31%) of (±)-12α-hydroxy-*cis*-isorotenoid **9** and (±)-12β-*hydroxy*-trans-*isorotenoid* **11**, obtained as needles, m.p. 201–203 °C (Found: C, 69.8; H, 6.15%; M^+, 396.153. $C_{23}H_{24}O_6$ requires C, 69.7; H, 6.1%; M, 396.157); λ_{max}(EtOH)/nm 215, 252, 258 and 290 (ε 22 400, 12 400, 13 600 and 5900); ν_{max}(KBr)/cm^{-1} 3490 (OH), 1595 and 1510 (aromatics). The NMR data for the 12β-*trans*-compound which follow were obtained by subtraction of the spectrum of the 12α-*cis*-compound (determined above) from that of the mixture: δ_H(400 MHz; $CDCl_3$) 1.34 (6 H, d, *J* 6.9, 2 × Me), 1.92 (1 H, d, *J* 2.7, OH), 3.07 (1 H, septet, *J* 6.9, 6′-H), 3.25 (1 H, dd, *J* 10.0 and 2.4, 12a-H), 3.85 (1 H, s, OMe), 3.90 (3 H, s, OMe), 4.13 (1 H, dd, *J* 11.5 and 11.5, 6-H^b), 4.67 (2 H, m, 6a-H and 6-H^a), 5.26 (1 H, m, 12-H), 6.47 (1 H, s, 4′-H), 6.50 (1 H, s, 4-H), 6.82 (1 H, s, 1-H), 7.11 (1 H, d, *J* 8.3, 10-H) and 7.24 (1 H, d, *J* 8.3, 11-H); *m/z* (EI +ve) 396 (3%, M^+), 378 (100, $M^+ - H_2O$), 363 (25, $C_{22}H_{19}O_5^+$) and 192 (8, $C_{11}H_{12}O_3^+$). A sample of the mixture was ground with pure (±)-12α-hydroxy-*cis*-isorotenoid **9** and the m.p. was redetermined: it was depressed to 184 °C.

Crystallographic Analysis of cis-*Isorotenone* **6** *and* trans-*Isorotenone* **5**.—Crystal data for *cis*-**6**: $C_{23}H_{22}O_6$, M = 394.44. Triclinic, *a* = 11.449(1), *b* = 11.768(1), *c* = 15.953(1) Å, α = 108.31(1)°, β = 103.79(1)°, γ = 94.38(1)°, *V* = 1955.11 $Å^3$, *Z* = 4, D_c = 1.34 g cm^{-3}, *F*(000) = 832. Space group $P\bar{1}$, Cu-Kα radiation, λ = 1.541 78 $Å^3$, μ(Cu-Kα) = 8.09 cm^{-1}, crystal size 0.6 × 0.4 × 0.3 mm.

Crystal data for *trans*-**5**; $C_{23}H_{22}O_6$, M = 394.44. Triclinic, *a* = 4.651(1), *b* = 9.325(1), *c* = 22.721(2) Å, α = 100.72(1)°, β = 91.73(1)°, γ = 100.36(1)°, *V* = 950.43 $Å^3$, *Z* = 2, D_c = 1.38 g cm^{-1}, *F*(000) = 416. Space group $P\bar{1}$, Cu-Kα radiation, λ = 1.541 78 Å, μ(Cu-Kα) = 8.32 cm^{-1}, crystal size 0.45 × 0.15 × 0.07 mm.

Crystals of each compound were mounted on an Enraf–Nonius CAD4 diffractometer and 25 reflections were used to determine accurate lattice parameters. Intensity data were collected for 1° < θ < 76°. Totals of 8154 (**6**) and 3937 (**5**) independent reflections were measured of which 4788 and 1991

respectively had $I > 3\sigma(I)$ and were considered observed and used in the subsequent refinement. Periodic measurement of standard reflections throughout data collection demonstrated their stability. The data were corrected for Lorentz and polarisation factors but no absorption corrections were made. Crystallographic calculations were performed using the CRYSTALS[19] system of programs. The structures were solved by direct methods using the MULTAN[20] program. Least-squares refinement including anisotropic thermal parameters for non-hydrogen atoms and isotropic refinement of hydrogen atoms located in a difference Fourier synthesis terminated at R 0.0814 (R_w 0.0939) for **6** and at R 0.0384 (R_w 0.0348) for **5**. The resulting molecular structures are illustrated in Figs. 3 and 4.*

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* Refined fractional atomic coordinates, fractional atomic coordinates of hydrogen atoms, bond lengths and angles and thermal parameters have been deposited at the Cambridge Crystallographic Data Centre. For details of the deposition scheme, see 'Instructions for Authors,' *J. Chem. Soc., Perkin Trans. 1*, 1993, Issue 1.

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