# Epimerization in acid degradation products of artemisinin

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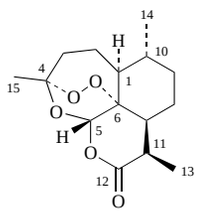
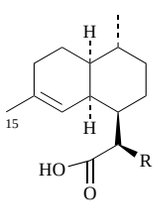
**Treatment of artemisinin 1 with acid leads to either a cyclohexane dione degradation product 10, which is a useful intermediate for biosynthetic studies of artemisinin, or to a decalin system which has undergone epimerization 8. It is shown by NMR spectroscopy, chemical reactions and molecular modelling that the bulky 7-substituent in the epimerized decalin series (8, 17, 14) adopts an axial solution conformation and that this is thermodynamically favoured over the natural configuration for which this substituent is equatorial (11, 15, 13). Conversely, for the cyclohexane dione series, the natural configuration in which the 7-substituent is equatorial is more favoured. Reasons for the differing conformational preferences in the two series, which are ultimately responsible for promoting epimerization, are discussed and a simple spectroscopic procedure for identification of epimerized products is presented.**

## Introduction

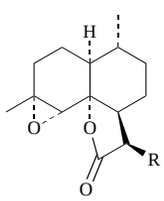
Artemisinin (qinghaosu) **1**, a natural product from the Chinese plant *Artemisia annua*,[1] is one of the most promising new antimalarial drugs, which is currently exciting great interest both in respect of its biological activity and its unusual chemical structure.[2] We are particularly interested in delineating the biosynthetic route to **1**, and have adopted a strategy which first involves synthesis of a variety of postulated biosynthetic intermediates such as **2a**–**4b**[3,4] incorporating C/H isotopic labels at



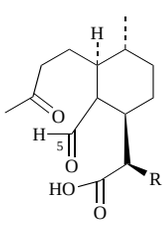
**1**



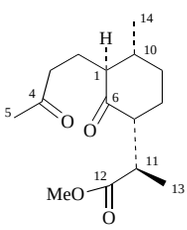
**2a** R = Me
**2b** R = =$CH_2$



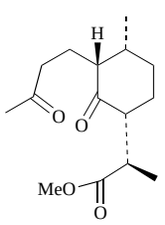
**3a** R = Me
**3b** R = =$CH_2$



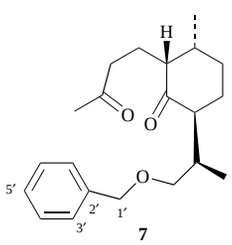
**4a** R = Me
**4b** R = =$CH_2$



**5**



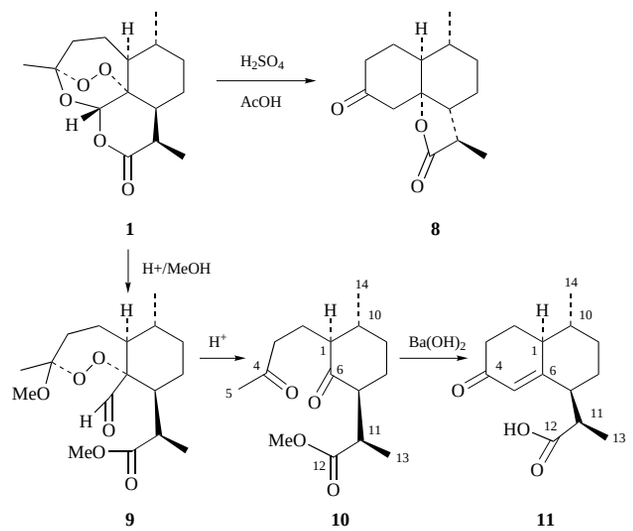
**6**



**7**

the 15- and/or 5-positions, and then feeding of these intermediates to the plant. Some of these target compounds such as artemisinic acid **2b**, arteannuin B **3b** and the seco-cadinane **4b** have also been reported as natural products from *Artemisia annua*.[3,5,6]

Compound **1** has been reported to undergo acid degradation accompanied by loss of formaldehyde to give a variety of products including the nor-sesquiterpenes **10** and **8**[7–12] (Scheme 1).



**Scheme 1** Reported acid-degradation reactions of artemisinin

The cyclohexane dione **10** would seem to have good potential for conversion into the biosynthetic intermediates required for our project; Robinson annulation of **10** under mild conditions has been reported to give the decalin acid **11** in the natural configuration.[12] We were interested to note that the decalin lactone **8**, produced directly by treatment of **1** with sulfuric acid–acetic acid, was reported to have undergone epimerization at the 7-position.[7,9–11] Because control of absolute chemistry in the preparation of isotopically labelled **2a**–**4b** is obviously critical if these products are to be useful in establishing a biosynthetic pathway, we have set out to determine the reasons for such epimerization and establish a simple means for recognizing its occurrence.

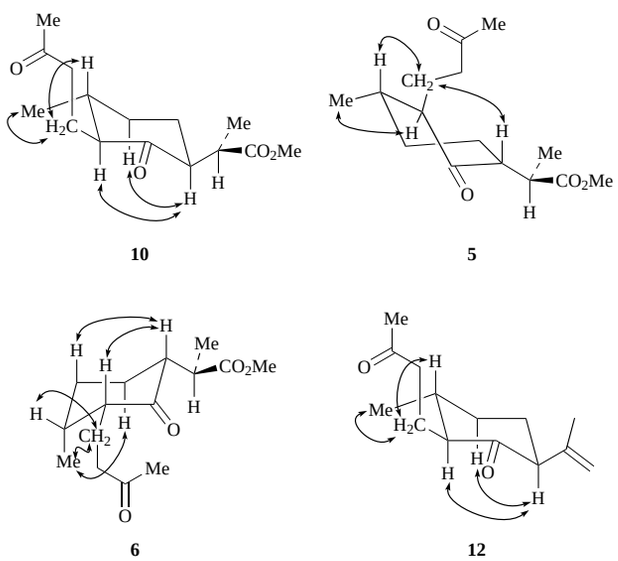
## Results and discussion

Treatment of the endoperoxide **9** (obtained from **1** by a literature procedure[8]) with acid led exclusively to the expected cyclohexane dione **10**. An alternative direct procedure, involving treatment of **1** with 30% MeOH–$H_2SO_4$ also yielded predominantly **10**, contaminated by small amounts of products **5** and **6**, which had undergone epimerization at the 1- and/or 7-positions. $^{13}C$ and $^{1}H$ NMR assignments (Table 1) for these three isomers (and for all compounds cited herein) were rigorously determined by 2D-NMR (HSQC, HMBC and

**Table 1** NMR data for diketone cyclohexane compounds used in Robinson annulations

| | $\delta_C$ | | | | | | $\delta_H$ | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Atom | **5** | **6** | **7** | **10** | **12** | **23** | **5** | **6** | **7** | **10** | **12** | **23** |
| 1 | 56.5 | 53.8 | 56.8 | 56.8 | 56.6 | 57.0 | 2.12 | 2.56 | 1.96 | 2.08 | 2.08 | 2.02 |
| 2 | 25.3 | 21.3 | 20.2 | 20.2 | 20.2 | 20.2 | 2.00 | 1.97 | 1.80 | 1.85 | 1.84 | 1.80 |
| | | | | | | | 1.99 | 1.39 | 1.75 | 1.80 | 1.77 | 1.74 |
| 3 | 40.7 | 41.7 | 41.4 | 41.2 | 41.3 | 41.3 | 2.41 | 2.49 | 2.49 | 2.54 | 2.58 | 2.50 |
| | | | | | | | 2.41 | 2.29 | 2.28 | 2.35 | 2.39 | 2.32 |
| 4 | 208.4 | 209.0 | 209.2 | 209.0 | 209.1 | 209.0 | | | | | | |
| 5 | 30.2 | 29.9 | 29.9 | 29.8 | 29.9 | 29.9 | 2.13 | 2.12 | 2.10 | 2.12 | 2.12 | 2.10 |
| 6 | 214.9 | 212.1 | 213.2 | 211.5 | 211.2 | 213.1 | | | | | | |
| 7 | 49.2 | 53.2 | 51.1 | 53.5 | 58.3 | 54.0 | 2.78 | 2.68 | 2.45 | 2.61 | 3.00 | 2.38 |
| 8α | 25.7 | 26.2 | 28.1 | 31.0 | 31.6 | 30.6 | 1.96 | 2.03 | 1.94 | 2.00 | 2.04 | 2.05 |
| 8β | | | | | | | 1.58 | 1.56 | 1.32 | 1.55 | 1.77 | 1.45 |
| 9α | 27.0 | 32.2 | 34.6 | 34.5 | 34.5 | 34.8 | 1.49 | 1.72 | 1.46 | 1.52 | 1.58 | 1.50 |
| 9β | | | | | | | 2.06 | 2.02 | 1.82 | 1.90 | 1.92 | 1.85 |
| 10 | 36.9 | 37.4 | 40.1 | 40.2 | 40.0 | 40.5 | 2.06 | 2.38 | 1.50 | 1.60 | 1.61 | 1.55 |
| 11 | 38.8 | 38.8 | 31.3 | 39.3 | 143.5 | 32.7 | 2.67 | 2.68 | 2.34 | 2.77 | | 2.15 |
| 12 | 177.3 | 177.0 | 72.9 | 176.1 | 112.5 | 72.9 | | | 3.30 | | 4.92 | 3.45 |
| | | | | | | | | | 3.30 | | 4.70 | 3.37 |
| 13 | 14.4 | 14.5 | 13.4 | 15.1 | 21.5 | 16.0 | 1.12 | 1.12 | 0.87 | 1.18 | 1.74 | 1.00 |
| 14 | 19.5 | 13.3 | 20.7 | 20.5 | 20.7 | 20.6 | 0.95 | 0.73 | 1.06 | 1.08 | 1.10 | 1.05 |
| OMe | 51.7 | 51.7 | | 51.6 | | | 3.67 | 3.69 | | 3.67 | | |
| 1′ | | | 73.4 | | | 73.0 | | | 4.48 | | | 4.49 |
| | | | | | | | | | 4.48 | | | 4.45 |
| 2′ | | | 138.6 | | | 138.7 | | | — | | | — |
| 3′/7′ | | | 128.4 | | | 128.3 | | | 7.32 | | | 7.31 |
| 4′/6′ | | | 128.3 | | | 127.5 | | | 7.32 | | | 7.31 |
| 5′ | | | 127.6 | | | 127.4 | | | 7.32 | | | 7.31 |

$^1$H-$^1$H COSY). Following complete assignments for all protons, relative stereochemistry and conformations were determined from NOESY correlations, supplemented by the results of $^1$H-$^1$H *J*-resolved spectra. Critical NOESY correlations used in determining the configurations and conformations of **10**, **5** and **6** are shown in Scheme 2. The axial nature of the 14-methyl

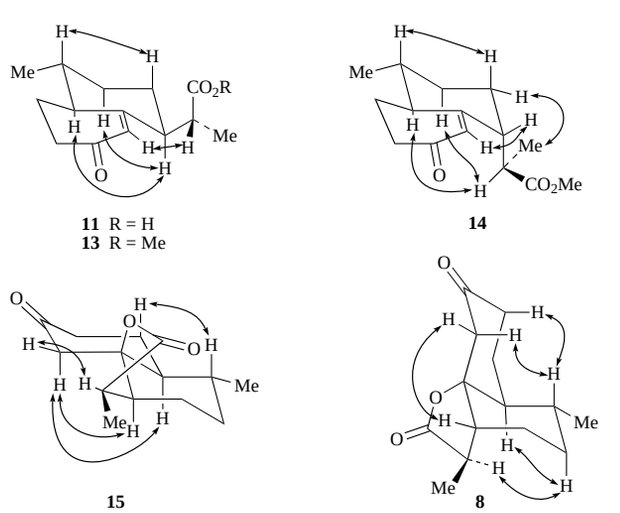



**Scheme 2** Critical NOESY correlations used in assigning the configuration and solution conformations of **10**, **5**, **6** and **12** indicated by double-headed arrows

group in the doubly epimerized compound **6** was confirmed by a large upfield shift in the $^{13}$C NMR spectrum (*ca*. 7 ppm) for this resonance when compared with **10** or **5**, which is the result of gauche interactions; in the $^1$H NMR spectrum of **6** the equatorial 10-proton had undergone a 0.5 ppm downfield shift relative to its axial counterpart in compound **10**. The bulky 7-substituent remained equatorial or pseudo-equatorial for all three cyclohexane dione epimers.

As expected, Robinson annulation of compound **10** in the presence of barium hydroxide gave the decalin acid **11** with retention of configuration at both 1- and 7-positions. As for the cyclohexane diones, this could be clearly demonstrated by use of NMR techniques such as NOESY and $^1$H-$^1$H *J*-resolved spectroscopy which showed the 7-isopropanoic acid substituent to be equatorial (NOESY correlations for the corresponding methyl ester **13** were identical; Scheme 3). As a solution in $CDCl_3$ the free acid **11** was converted slowly into the *cis*-lactone **15**, in which the 7-substituent remained equatorial and the decalin system was *trans*-fused.

The epimeric *cis*-lactone **8** (obtained directly from degradation of **1** in acetic–sulfuric acid) was shown by NOESY to adopt a conformation with the 7-substituent axial and the decalin system *cis*-fused (Scheme 3). This was confirmed by the




**Scheme 3** Critical NOESY correlations used in assigning the configuration and solution conformations of **11**, **13**, **14**, **15** and **8** indicated by double-headed arrows

results of $^1$H-$^1$H *J*-resolved spectra which demonstrated a W-coupling (2 Hz) between H-5α (dd, *J* 16, 2 Hz) and the equatorial H-7 (ddd, *J* 13, 7, 2 Hz). [No such coupling was observed in *J*-resolved spectra of **15**, for which the H-5β proton shared a W-coupling (2 Hz) with H-3β (ddd, *J* 13, 7, 2 Hz) but

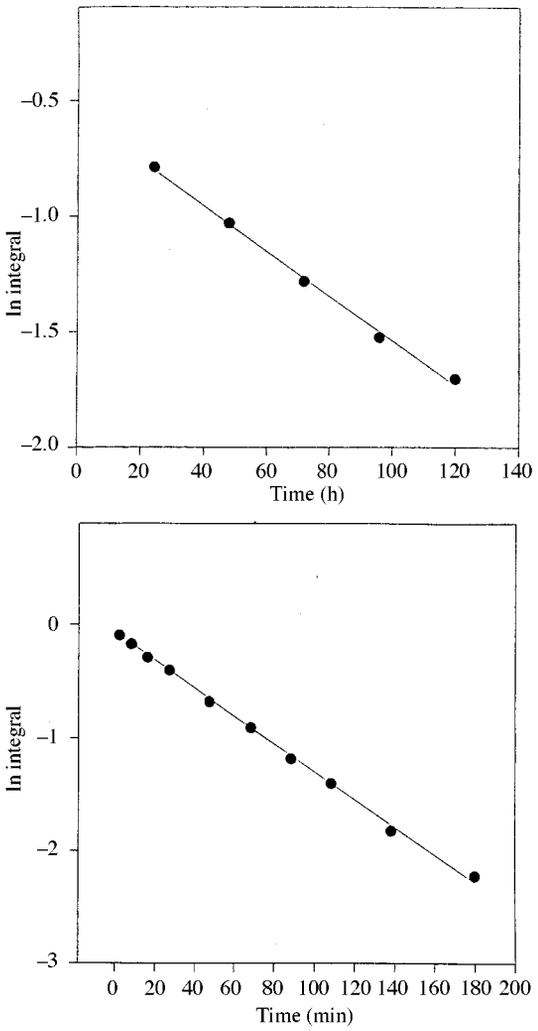
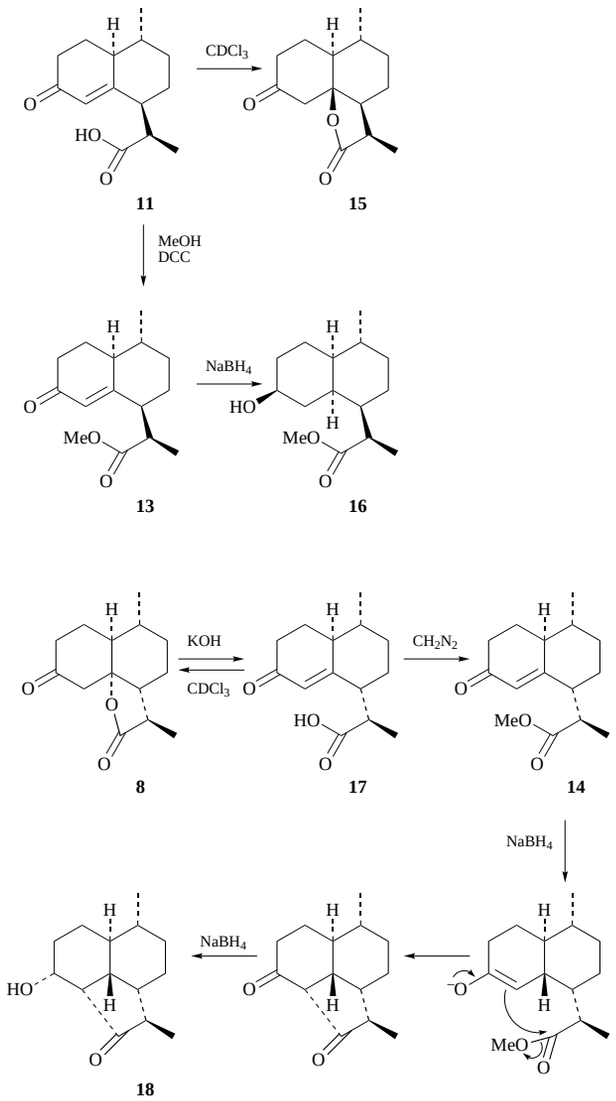



**Fig. 1** Rate of lactonization of **11** into **15** determined by ratio of integrals in $^1$H NMR at $\delta$ 1.27 (H-13, **11**) and $\delta$ 1.13 (H-13, **15**) as $9.7 \times 10^{-3}$ h$^{-1}$; rate of lactonization of **17** into **8** determined by ratio of integrals in $^1$H NMR at $\delta$ 5.93 (H-5, **17**) and $\delta$ 2.66 (H-11, **8**) as $1.2 \times 10^{-2}$ min$^{-1}$




**Scheme 4** Differing reactions of 7-epimeric degradation products **11** and **8**

not with the axial H-7.] Base-catalysed ring opening of **8** gave the free acid **17**, which quickly reverted to the parent lactone in $CDCl_3$ solution and was best derivatized as the methyl ester **14** for NMR analysis. The results of NOESY clearly indicated that the 7α-methyl isopropanoate ester substituent in **14** was also axial.

The differing conformations predicted by NMR for the two decalin 7-epimeric series were confirmed by their chemical reactions (Scheme 4). Thus, free acid **11** underwent lactonization to **15** when stored in $CDCl_3$ some 75 times more slowly than the 7α-epimer **17** (Fig. 1). The rapid lactonization of **17** is consistent with the axial disposition of the carboxy group in this epimer. In the axial conformation the carboxylic acid nucleophile can easily participate in an intramolecular Michael reaction since it is well positioned to interact axially with the π-system of the enone: good interaction is much more difficult for the equatorial carboxy group in **11** (*cf.* Scheme 3). (Incidentally, this difference in reactivity also accounts for the observation that the decalin 7α-epimer is normally isolated as a lactone **8** from acid degradation of artemisinin, whilst the product with natural configuration **11** is obtained as the free acid.) Sodium borohydride reduction of the α-epimeric methyl ester **14** yielded a tricyclic product incorporating a *trans*-decalin ring **18**, whereas reduction of the corresponding β-epimer **13** gave the expected reduction product **16** (the equatorial 7-substituent in **13** blocks initial hydride attack at the conjugated double bond from the β-face resulting in a *cis*-decalin system; reduction at the ketone group is then only possible from the α-face). The relative stereochemistry of **18** was determined from $^1$H-$^1$H couplings: H-1 (dddd, *J* 11.4, 11.4, 11.4, 2.5 Hz) shows three large couplings to its neighbours and therefore requires a *trans*-fused decalin system, whilst H-5 (dd, *J* 6.3, 6.0 Hz) is equatorial and H-4 (ddd, *J* 11.7, 5.8, 6.0 Hz) is axial (these predictions were also confirmed by NOESY). The *cis*-decalin system of **16** was demonstrated by NOESY spectra (H-6 correlates both with H-4 and H-1) and the axial nature of H-4 (dddd, *J* 11.7, 11.7, 4.9, 4.9 Hz) was confirmed by observation of two large coupling constants in $^1$H NMR. The unexpected reaction pathway followed by **14** and the stereochemistry of the resulting decalin system is simply explained as a trapping of the enolate intermediate, generated during conjugate reduction of **14**, by the axially disposed methyl ester group. Such trapping is much less likely for the equatorial methyl ester of **13**.

One useful consequence of the preferred solution conformations for epimeric acid degradation products of artemisinin is that it is possible to distinguish products in which epimerization has occurred (such as **8**, **17** and **14**) from those in which it has not (such as **11**, **15** and **13**) by simple inspection of the $^1$H NMR spectrum. A diagnostic coupling pattern (dq, $J \approx 12$, 7 Hz) for the well-resolved H-11 resonance ($\delta$ 2.6–3.1 ppm) was noted for all epimerized products that was obviously different from the pattern for products of natural configuration (dq, $J \approx 7$, 7 Hz). This difference is a consequence of the axial or equatorial

**Table 2a** $^{13}$C NMR data for decalin products from Robinson annulations

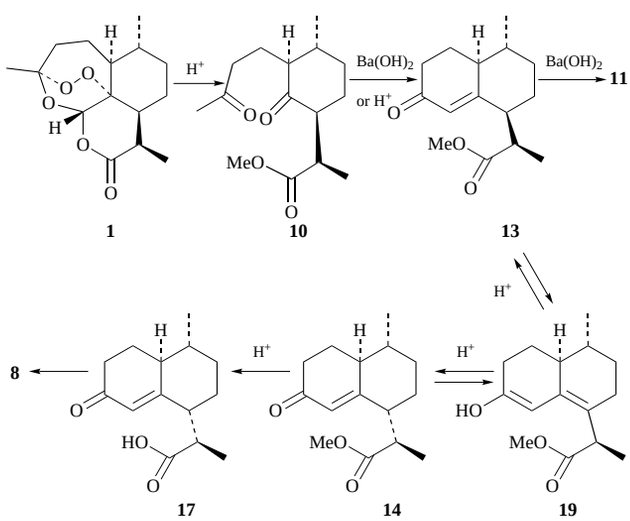
| Atom | **8** | **11** | **13** | **14** | **15** | **17** | **20** | **22** | **24** | **25** |
|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 44.2 | 45.9 | 46.0 | 40.8 | 48.1 | 40.7 | 45.0 | 41.6 | 46.1 | 41.0 |
| 2 | 21.6 | 24.6 | 24.5 | 26.2 | 24.0 | 26.1 | 26.1 | 26.2 | 25.0 | 25.6 |
| 3 | 35.9 | 34.2 | 34.0 | 36.1 | 40.3 | 35.9 | 35.8 | 36.1 | 34.5 | 35.5 |
| 4 | 207.2 | 199.9 | 199.6 | 199.8 | 207.0 | 200.5 | 201.0 | * | 199.9 | 200.0 |
| 5 | 47.2 | 121.4 | 121.3 | 125.6 | 50.7 | 125.5 | 123.2 | 126.8 | 121.6 | 125.1 |
| 6 | 87.6 | 168.0 | 168.1 | 167.1 | 85.9 | 167.3 | 167.7 | * | 169.8 | 169.8 |
| 7 | 48.9 | 48.3 | 48.7 | 47.9 | 43.3 | 47.6 | 52.4 | 49.9 | 48.7 | 48.1 |
| 8 | 22.5 | 33.0 | 33.4 | 26.8 | 24.4 | 26.7 | 31.7 | 28.5 | 30.9 | 28.6 |
| 9 | 29.6 | 35.0 | 35.0 | 29.1 | 32.5 | 29.1 | 34.6 | 30.2 | 35.3 | 29.7 |
| 10 | 30.7 | 38.6 | 38.5 | 39.4 | 31.2 | 39.5 | 39.0 | * | 38.9 | 39.4 |
| 11 | 36.4 | 40.5 | 40.6 | 41.6 | 39.9 | 41.5 | 144.7 | * | 32.9 | 33.2 |
| 12 | 177.7 | 180.2 | 176.1 | 175.4 | 178.0 | 179.4 | 113.8 | 112.6 | 72.6 | 74.2 |
| 13 | 13.3 | 15.8 | 16.0 | 15.0 | 9.2 | 14.9 | 21.4 | 22.3 | 17.1 | 15.8 |
| 14 | 18.9 | 20.2 | 20.2 | 20.0 | 19.9 | 20.0 | 20.3 | 20.2 | 20.3 | 20.2 |
| OMe | | | 51.7 | 51.4 | | | | | | |
| 1′ | | | | | | | | | 73.2 | 73.2 |
| 2′ | | | | | | | | | 138.6 | 138.3 |
| 3′/7′ | | | | | | | | | 128.4 | 128.4 |
| 4′/6′ | | | | | | | | | 127.5 | 127.7 |
| 5′ | | | | | | | | | 127.5 | 127.6 |

* Resonances not clearly resolved from those of **20**.

nature of the 7-substituent in the two epimeric series. For the α-epimers which adopt an axial conformation for the 7-substituent, H-11 must reside below the plane of the decalin ring in order to allow the more bulky methyl and carboxy groups to project away from the ring system. This leads to a dihedral angle of 180° between H-7 and H-11 and a relatively large 3-bond $^1$H-$^1$H coupling constant (≈12 Hz) as predicted by the Karplus equation. For the β-epimeric series, the 7-substituent is equatorial and a smaller dihedral angle arises between H-7 and H-11 (Scheme 3), the H-7/H-11 coupling constant is therefore correspondingly smaller (≈7 Hz). It is also evident from inspection of Table 2a, that $^{13}$C chemical shifts for the B-ring are generally upfield (by up to 7 ppm) for compounds belonging to the epimerized series as compared with those of natural configuration. This is the result of gauche effects arising from the axial nature of the 7-substituent in the α-epimer. Consequently, inspection of either the 1D- $^1$H or $^{13}$C spectrum is generally considered sufficient to recognize the occurrence of epimerization for the decalin series of acid degradation products.

The 7α-epimeric lactone **8**, obtained from acid treatment of **1**, must have arisen from Robinson annulation of a cyclohexane dione intermediate such as **10**. Since we have shown that there is little tendency for epimerization in the formation of **10** (the configuration of **10** is such that all substituents around the cyclohexanone ring are able to adopt equatorial conformations: compound **10** was always the predominant product accompanied by only small amounts of **5** and **6**), epimerization at the 7-position must occur following formation of the new carbocyclic ring (Scheme 5). The most natural mechanism would involve the intermediacy of an extended enol, **19**, which is generated in acidic medium: reversible reprotonation/deprotonation of **19** from either the α or β face allows formation of epimers at C-7. (Presumably such a mechanism does not operate under the heterogeneous reaction conditions when $Ba(OH)_2$ is employed as the cyclization catalyst.)

Some evidence for the intermediacy of an extended enol intermediate such as **19** comes from study of Robinson annulation of the closely related compound **12** (prepared in two steps from isopulegol). Compound **12** was obtained exclusively as the 1β,7β-epimer as proven by NOESY correlations and chemical shift values which compared well with those of **10** (see Table 1 and Scheme 2). The major product of annulation of the cyclohexane dione **12** is the highly conjugated compound **21** which has undergone isomerization of the terminal double bond (Scheme 6). Only small quantities of the two 7-epimers **20** and



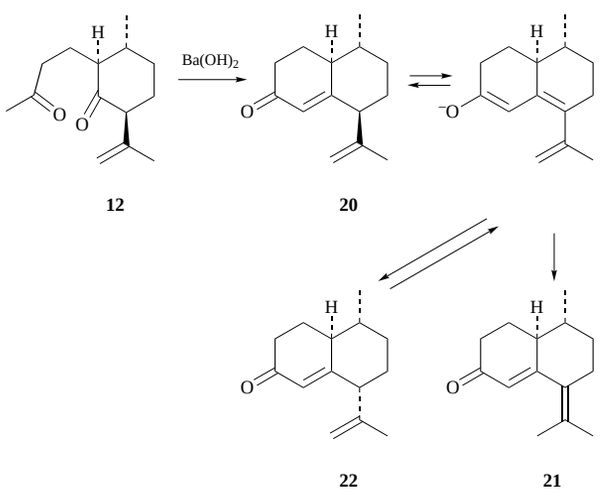

**Scheme 5** Postulated mechanism for epimerization in acid-catalysed degradation of artemisinin

**22** were isolated. (NOESY spectra used to assign conformations for **20** and **22** gave comparable results to those shown in Scheme 3 for **11** and **14**, *e.g.* correlations were observed from H-7 to H-9α and H-1α in compound **20**; but only from H-7 to H-5 in compound **22**. $^1$H and $^{13}$C chemical shift assignments for **20** and **22** also compared favourably with those for **11/13** and **17/14** shown in Tables 2a and 2b.) Such double bond isomerization is naturally explained as a consequence of formation of an extended enolic intermediate analogous to **19**.

Exclusive formation of the 7α-epimer **8** in the acid degradation of artemisinin, *via* this mechanism of extended enol formation, must then imply that the epimerized decalin product is thermodynamically favoured over that of the natural conformation since both epimers exist in equilibrium. Further evidence for this conclusion came from Robinson annulation of the related compound **23** [prepared as the 1β,7β-epimer together with a little 1α,7β-epimer **7** (see Table 1) in four steps from isopulegol]. The stereochemistry and conformation of the cyclohexane dione **23** was determined by $^1$H-$^1$H *J*-resolved spectroscopy as being all equatorial because both H-7 (ddd, *J* 12.4, 5.8, 5.8 Hz) and H-1 (ddd, *J* 10.9, 8.5, 3.2 Hz) were clearly axial and shown to be involved in large couplings with H-8α (12.4 Hz) and H-10 (10.9 Hz), respectively. That **7** is the 1-epimer was demonstrated by similar means [H-7 (ddd, *J* 12.4,

**Table 2b** $^1$H NMR data for decalin products from Robinson annulations

| Atom | **8** | **11** | **13** | **14** | **15** | **17** | **20** | **22** | **24** | **25** |
|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 1.48 | 1.89 | 1.89 | 2.21 | 1.49 | 2.23 | 1.94 | 2.10 | 1.85 | 2.03 |
| 2α | 2.12 | 2.15 | 2.14 | 2.25 | 2.19 | 2.25 | 2.25 | 2.24 | 2.15 | 2.09 |
| 2β | 2.08 | 1.93 | 1.96 | 1.59 | 1.85 | 1.56 | 1.72 | 1.69 | 1.85 | 1.63 |
| 3α | 2.30 | 2.32 | 2.30 | 2.37 | 2.33 | 2.26 | 2.28 | 2.20 | 2.25 | 2.12 |
| 3β | 2.30 | 2.37 | 2.36 | 2.20 | 2.54 | 2.37 | 2.39 | 2.40 | 2.35 | 2.32 |
| 5 | 2.85 (β)<br>2.35 (α) | 5.80 | 5.76 | 5.76 | 2.79 (β)<br>2.35 (α) | 5.93 | 5.77 | 5.91 | 5.88 | 5.82 |
| 7 | 1.85 | 2.35 | 2.35 | 2.47 | 2.15 | 2.52 | 2.79 | 3.02 | 2.12 | 2.16 |
| 8α | 1.84 | 2.02 | 1.89 | 1.97 | 1.76 | 1.98 | 1.89 | 2.14 | 2.05 | 1.98 |
| 8β | 1.80 | 1.38 | 1.31 | 1.65 | 1.12 | 1.62 | 1.60 | 1.69 | 1.12 | 1.55 |
| 9α | 1.30 | 1.32 | 1.31 | 1.35 | 1.10 | 1.34 | 1.33 | 1.40 | 1.25 | 1.40 |
| 9β | 1.73 | 1.86 | 1.85 | 1.62 | 1.72 | 1.59 | 1.87 | 1.58 | 1.82 | 1.58 |
| 10 | 1.68 | 1.64 | 1.64 | 1.36 | 1.37 | 1.35 | 1.45 | 1.42 | 1.56 | 1.40 |
| 11 | 2.66 | 2.75 | 2.73 | 2.82 | 3.02 | 2.75 | — | — | 2.08 | 2.05 |
| 12 | | | | | | | 4.98<br>4.79 | 4.96<br>4.74 | 3.48<br>3.35 | 3.24<br>3.11 |
| 13 | 1.21 | 1.27 | 1.22 | 1.20 | 1.13 | 1.22 | 1.71 | 1.71 | 1.07 | 1.05 |
| 14 | 1.04 | 1.05 | 1.04 | 1.05 | 1.00 | 1.02 | 1.05 | 1.01 | 1.02 | 1.01 |
| OMe | | | 3.69 | 3.54 | | | | | | |
| 1′ | | | | | | | | | 4.51<br>4.45 | 4.39<br>4.39 |
| 3′/7′ | | | | | | | | | 7.32 | 7.32 |
| 4′/6′ | | | | | | | | | 7.32 | 7.32 |
| 5′ | | | | | | | | | 7.32 | 7.32 |




**Scheme 6** Evidence for participation of an extended enolic intermediate in epimerization

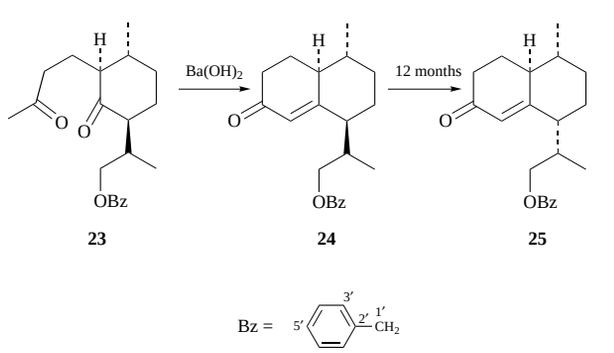
5.5, 5.2 Hz) shared a large coupling with H-8α (12.4 Hz) and therefore remained axial, whereas H-1 (ddd, *J* 7.6, 5.2, 1.7 Hz) is equatorial since it shares a small coupling (5.2 Hz) with H-10]. Robinson annulation of **23** produced the decalin product of natural configuration **24** as expected when $Ba(OH)_2$ was employed as catalyst at room temperature (significantly, a 1 : 1 mixture of 7-epimers was obtained at reflux, however). Following storage at room temperature for 1 year, it was discovered that **24** had been largely converted into the 7α-epimer **25** which must therefore be thermodynamically more stable (Scheme 7). Epimerization in **25** was determined by results of $^1$H-$^1$H *J*-resolved spectroscopy [H-7, $\delta$ 2.16 (ddd, *J* 11.3, 5.3, 3.0 Hz) now shared an 11.3 Hz coupling with H-11 which, as commented on previously, is diagnostic for epimerization at the 7-centre] and confirmed by NOESY correlations (as for compound **14** in Scheme 3). $^1$H and $^{13}$C chemical shifts for **25** also agreed well with those for epimerized products **17**, **14** and **22**.

Molecular modelling studies have confirmed all the preceeding experimental results. The 7α-epimers of the decalin series of compounds are indeed expected to be significantly more stable than their 7β-counterparts by more than 10 kJ mol$^{-1}$ (Table 3). The predicted lowest energy conformations demonstrate an axial conformation preference for the α-epimers and an equatorial preference for the β-epimers. Molecular modelling studies

**Table 3** Calculated energies (kJ mol$^{-1}$) for most stable conformers of 7-epimeric artemisinin degradation products *

| Class of compound | 7β-series | Energy (kJ mol$^{-1}$) | 7α-series | Energy (kJ mol$^{-1}$) |
|---|---|---|---|---|
| Cyclohexane diones | **10** | 123.0 | **5** | 127.4 |
| Decalins (free acid) | **11** | 78.2 | **17** | 66.9 |
| Decalins (methyl ester) | **13** | 141.4 | **14** | 128.1 |

* Calculated using MM2 force field model.




**Scheme 7** Evidence for 7α-epimer being more thermodynamically stable

for the 7-epimeric cyclohexane diones **10** and **5** have also confirmed that, conversely, the natural configuration is of lower energy than its 7-epimer and that both isomers prefer the equatorial or pseudo-equatorial conformations predicted by NMR.

The relative stability and conformations adopted by the cyclohexane diones used in Robinson annulation conforms with a simplistic analysis in which bulky ring-substituents are assumed always to prefer an equatorial conformation, which is normally of lower energy. The conclusion from both modelling and experiment that the 7α-epimers of the cyclized decalin series are more stable and that the bulky 7-substituent of the epimerized series preferentially adopts an axial conformation does not accord with such a simplistic analysis. Furthermore, following epimerization, it is possible for either the 7- or the 10-substituent to adopt the energetically disfavoured axial con-

formation by flipping in the B-ring and, given a choice, the simplistic model would predict that the less bulky 10-methyl group should become axial (as observed for compound **6** for example) rather than the 7-substituent.

From the point of view of the 7-substituent, the major difference between the two series of compounds is the presence of either a vicinal double bond to O in the cyclohexane diones or a vicinal double bond to CH(R) in the decalins. It seems that the overriding factor in destabilizing the 7β-equatorial substituent in the decalin series is an interaction with the proton of the double bond, which is absent from the cyclohexane dione series. Such destabilizing influences, designated $A^{1,3}$ strain,[13] have been proposed previously for 2-substituted methylenecyclohexane systems, for which bulky substituents have been observed to be axial rather than equatorial.

We therefore propose that the tendency to epimerize in the acid degradation of artemisinin is attributable to the appearance of $A^{1,3}$ strain following formation of a decalin ring by Robinson annulation. Whilst products of natural configuration are unable to overcome such strain (flipping of the B-ring would require both 10- and 7-substituents to become axial, which is evidently even more energetically disfavoured than allowing $A^{1,3}$ strain), epimerization at the 7-centre allows only the 7-substituent to become axial and the ensuing relief of $A^{1,3}$ strain is thereby energetically favoured. Given that the natural tendency in preparation of postulated biosynthetic intermediates **2a**–**4b** is expected to be towards epimerization, such a possibility is best guarded against by employing mild conditions or displacing the 5,6 double bond at an early stage. Fortunately, as a consequence of our studies, simple 1D-NMR procedures now exist to detect such unwanted epimerization when it does occur.

## Experimental

### General details

All NMR experiments were run on Bruker DPX 300 or DRX 500 instruments with $CDCl_3$ as solvent. The chemical shifts were recorded relative to TMS as an internal standard and all coupling constants, *J*, are reported in Hz. Two-dimensional spectra such as HSQC, HMBC, $^1$H-$^1$H COSY, NOESY and $^1$H-$^1$H *J*-resolve were normally recorded with 1024 data points in $F_2$ and 256 data points in $F_1$ (occasional high resolution experiments had 4096 data points in $F_2$ and 1024 data points in $F_1$). High resolution MS were recorded in EI mode (70 eV) on a Finnigan-MAT 95 MS spectrometer. FTIR spectra were recorded in $CH_2Cl_2$ on a Shimadzu FTIR-8201 PC instrument. TLC plates were developed using *p*-anisaldehyde. Column chromatography was performed using silica gel 60–200 μm (Merck). HPLC separations were performed using a PREP-SIL 20 mm × 25 cm column, flow rate 8 ml min$^{-1}$. Optical rotations, $[\alpha]_D$, are recorded as $10^{-1}$ deg cm$^2$ g$^{-1}$.

### Acid conversion of 1 into the endoperoxide 9

Compound **1** (7.0 g) was added to ice-cooled $H_2SO_4$ (12 M)–MeOH (3:10, v/v; 130 ml). After being stirred for 3 h with ice-cooling, the reaction mixture was poured into cold water (200 ml), and extracted with $CHCl_3$ (3 × 100 ml). The combined extracts were then washed with water (3 × 300 ml), dried ($MgSO_4$), and evaporated to yield **9** (3.5 g) as a liquid, $[\alpha]_D$ +156 (*c* 1.22 in $CHCl_3$); $\nu_{max}$/cm$^{-1}$ ($CHCl_3$) 2955, 2941, 2871 and 1732; $\delta_H$($CDCl_3$) 9.92 (1H, d, *J* 2.7, H-5), 3.65 (3H, s, 12-OMe), 3.35 (3H, s, 4-OMe), 3.18 (1H, dq, *J* 3.5, 6.5, H-11), 2.32 (1H, m, H-10), 1.23 (3H, d, *J* 7.2, H-13), 1.19 (3H, s, H-4) and 0.89 (3H, d, *J* 6.5, H-14); $\delta_C$($CDCl_3$) 200.7 (CH, C-5), 175.3 (C, C-12), 108.5 (C, C-4), 94.1 (C, C-6), 59.5 (CH, C-1), 51.4 ($CH_3$, 12-OMe), 49.1 ($CH_3$, 4-OMe), 48.7 (CH, C-7), 40.6 ($CH_2$, C-3), 37.9 (CH, C-11), 35.5 ($CH_2$, C-9), 32.0 (CH, C-10), 25.3 ($CH_2$, C-8), 22.1 ($CH_2$, C-2), 20.5 ($CH_3$, C-14), 19.6 ($CH_3$, C-15) and 17.8 ($CH_3$, C-13); HREIMS *m*/*z* (rel. int %): 296.1988 [$M^+ - O_2$, $\Delta$ = 0.0 mmu for $C_{17}H_{28}O_4$] (3), 264 (3), 251 (4), 225 (11), 193 (15), 179 (40), 165 (24), 151 (29), 133 (18), 101 (47) and 86 (100).

### Direct conversion of 1 into the cyclohexane dione 10

Compound **1** (5.0 g) was added to $H_2SO_4$ (12 M)–MeOH (4:10, v/v; 140 ml). After being rapidly stirred, the reaction mixture was poured into cold water (100 ml). Unchanged **1** was filtered off and the filtrate was then extracted with $CHCl_3$ (3 × 100 ml). The combined extracts were washed with water (3 × 300 ml), dried ($MgSO_4$) and concentrated to give a pale yellow liquid which was subjected to column chromatography [hexane–EtOAc (3:1, v/v)] to yield **10** (3.6 g) together with small quantities of the epimers **5** and **6**.

**Compound 10.** Oil, $\delta_H$($CDCl_3$) 3.67 (3H, s), 2.77 (1H, dq, *J* 7.0, 7.0), 2.61 (1H, ddd, *J* 11.9, 7.0, 5.4), 2.54 (1H, ddd, *J* 17.3, 9.4, 5.3), 2.38 (1H, ddd, *J* 17.3, 9.2, 6.1), 2.12 (3H, s), 1.18 (3H, d, *J* 7.0) and 1.08 (3H, d, *J* 6.1); HREIMS *m*/*z* (rel. int %): 268.1672 [$M^+$, $\Delta$ = 0.2 mmu for $C_{15}H_{24}O_4$] (2), 250 (100), 235 (10), 209 (30), 180 (50), 163 (28) and 151 (28).

**Compound 5.** Oil, $[\alpha]_D$ −11.2 (*c* 0.46 in $CHCl_3$); $\nu_{max}$/cm$^{-1}$ ($CHCl_3$) 2957, 2871, 1732 and 1711; $\delta_H$($CDCl_3$) 3.67 (3H, s), 2.78 (1H, ddd, *J* 11.5, 9.2, 6.2), 2.67 (1H, dq, *J* 9.1, 7.1), 2.13 (3H, s), 1.12 (3H, d, *J* 7.1) and 0.95 (3H, d, *J* 7.0); HREIMS *m*/*z* (rel. int %) 250.1567 [$M - H_2O$, $\Delta$ = 0.2 mmu for $C_{15}H_{22}O_3$] (19), 209 (50), 208 (45), 180 (100), 163 (80) and 119 (95).

**Compound 6.** Oil, $[\alpha]_D$ +61.5 (*c* 1.03 in $CHCl_3$); $\nu_{max}$/cm$^{-1}$ ($CHCl_3$) 2955, 2899, 1732 and 1715; $\delta_H$($CDCl_3$) 3.69 (3H, s), 2.68 (2H, m), 2.49 (1H, ddd, *J* 17.4, 8.8, 3.0), 2.29 (1H, ddd, *J* 17.4, 8.5, 6.7), 2.12 (3H, s), 1.56 (1H, dddd, *J* 12.9, 12.9, 12.9, 4.0), 1.12 (3H, d, *J* 6.8), 0.74 (3H, d, *J* 7.1); HREIMS *m*/*z* (rel. int %) 268.1681 ($M^+$, $\Delta$ = −0.6 mmu for $C_{15}H_{24}O_4$) (0.5), 250 (100), 218 (45), 208 (50), 190 (65), 180 (70), 178 (75), 153 (80) and 136 (60).

### Conversion of 9 into 10

Compound **9** (1 g) was dissolved in dilute aq. HCl (10%; 10 ml) and the solution stirred for 30 min, after which it was extracted with $CHCl_3$. The extract was washed, dried ($MgSO_4$) and evaporated under reduced pressure to yield compound **10** (0.3 g).

### Acid conversion of 1 into the decalin lactone 8

Compound **1** (30.0 g) was added to $H_2SO_4$ (12 M)–AcOH (glacial) (1:10, v/v; 330 ml). After being stirred overnight at room temperature, the reaction mixture was poured into cold water (500 ml), and extracted with $CHCl_3$ (3 × 300 ml). The combined extracts were then washed with water (5 × 600 ml), dried ($MgSO_4$) and evaporated. The residual dark-brown gum was subjected to recrystallization twice (EtOH) to yield **8** (10.2 g) as crystals, mp 151–152 °C; $[\alpha]_D$ −15.9 (*c* 1.1 in $CHCl_3$); $\nu_{max}$/cm$^{-1}$ ($CHCl_3$) 2965, 2934, 1771 and 1720; $\delta_H$($CDCl_3$) 2.85 (1H, d, *J* 15.9), 2.66 (1H, dq, *J* 13.8, 6.9), 2.35 (1H, dd, *J* 15.9, 2.0), 1.21 (3H, d, *J* 7.0) and 1.04 (3H, d, *J* 6.2); HREIMS *m*/*z* (rel. int %) 236.1419 ($M^+$, $\Delta$ = −0.7 mmu for $C_{14}H_{20}O_3$), 208 (65), 192 (70), 163 (70), 134 (50) and 119 (100).

### Robinson annulation of 10

$Ba(OH)_2 \cdot 8H_2O$ (4.5 g) was added to a solution of **10** (3.6 g) in EtOH (110 ml). After being stirred for 2 h at room temperature the reaction mixture was acidified with aq. HCl (3 M). The resulting precipitate was filtered off and dissolved in $CH_2Cl_2$, and the solution was washed with water, dried ($MgSO_4$) and evaporated to yield **11** (1.1 g) as a solid, mp 158–159 °C; $[\alpha]_D$ +13.9 (*c* 0.17 in $CHCl_3$); $\nu_{max}$/cm$^{-1}$ ($CHCl_3$) 3400–2400br, 2959, 2931, 2874, 1709 and 1669; $\delta_H$($CDCl_3$) 5.80 (1H, s), 2.75 (1H, dq, *J* 8.2, 7.0), 1.27 (3H, d, *J* 6.9) and 1.04 (3H, d, *J* 6.5); HREIMS *m*/*z* (rel. int %) 236.1412 [$M^+$, $C_{14}H_{20}O_3$ $\Delta$ = 0.0 mmu] (75), 191 (70) and 163 (100). When left in $CDCl_3$, compound **11** was slowly converted into compound **15**, a solid, mp 171–172 °C; $[\alpha]_D$ −60.9 (*c* 0.36, $CHCl_3$); $\nu_{max}$/cm$^{-1}$ ($CHCl_3$)

2938, 2862, 1772 and 1717; $\delta_H(CDCl_3)$ 3.02 (1H, dq, *J* 6.9, 6.9), 2.79 (1H, dd, *J* 14.9, 2.0), 2.54 (1H, ddd, *J* 15.4, 2.5, 2.5), 2.35 (1H, d, *J* 14.9), 1.13 (3H, d, *J* 7.2) and 1.00 (3H, d, *J* 6.5); HREIMS *m/z* (rel. int %) 236.1412 ($M^+$, $\Delta = 0.3$ mmu for $C_{14}H_{20}O_3$) (93), 208 (100), 180 (75), 179 (72), 163 (67) and 134 (35).

**Conversion of 8 into the free acid 17**

Compound **8** (9.0 g) was dissolved in aqueous KOH (10% w/v; 180 ml), after which sufficient aq. HCl (3 M) was added to the solution to bring it to pH *ca*. 3. The resulting solid was filtered off, washed with a small amount of ice-cold water and then subjected to suction to dry it. Compound **17** as a white solid (4.5 g) resulted; $\delta_H(CDCl_3)$ 5.93 (1H, s), 2.75 (1H, dq, *J* 11.2, 6.8), 2.52 (1H, dd, *J* 11.1, 3.3), 2.37 (1H, ddd, *J* 15.0, 4.0, 4.0), 1.22 (3H, d, *J* 6.7) and 1.02 (3H, d, *J* 5.9); HREIMS *m/z* (rel. int %) 236.1409 [$M^+$, $\Delta = 0.3$ mmu for $C_{14}H_{22}O_3$] (93), 208 (100), 180 (75), 179 (72), 163 (67) and 134 (35).

**Methylation of 11**

To a mixture of **11** (0.20 g), MeOH (0.14 g) and 4-dimethylaminopyridine (0.04 g) in $CH_2Cl_2$ (2 ml), was added a solution of dicyclohexylcarbodiimide (DCC) in $CH_2Cl_2$ (1.446 M; 0.4 ml). After the mixture had been stirred overnight, a precipitate formed which was filtered off. The filtrate was concentrated to give **13** as a colourless liquid (0.08 g). This was purified by preparative TLC (hexane–EtOAc, 7:3) to afford an oil, $[\alpha]_D$ +4.9 (*c* 0.12 in $CHCl_3$); $\nu_{max}/cm^{-1}$ ($CHCl_3$) 2955, 2855, 1732 and 1668; $\delta_H(CDCl_3)$ 5.76 (1H, s), 3.69 (3H, s), 2.73 (1H, dq, *J* 8.5, 6.8), 1.22 (3H, d, *J* 6.8) and 1.04 (3H, d, *J* 5.9); HREIMS *m/z* (rel. int %): 250.1570 [$M^+$, $C_{15}H_{22}O_3$ $\Delta = -0.1$ mmu] (55), 191 (100) and 163 (95).

**Methylation of 17**

To a mixture of **17** (6.6 g) and Diazald (30.0 g) in MeOH (153 ml), was added dropwise a methanolic solution of KOH (7.8 g/241 ml) with ice–salt cooling. The reaction mixture was stirred overnight at room temperature to afford a white precipitate which was filtered off. The filtrate was concentrated to give **14** as a pale yellow liquid (3.3 g). This was purified by column chromatography (hexane–EtOAc, 16:9); $\delta_H(CDCl_3)$ 5.76 (1H, s), 3.54 (3H, s), 2.82 (1H, dq, *J* 11.2, 6.8), 2.37 (1H, ddd, *J* 15.9, 5.1, 4.0), 1.20 (3H, d, *J* 6.8) and 1.05 (3H, d, *J* 5.9); HREIMS *m/z* (rel. int %) 250.1570 [$M^+$, $\Delta = -0.1$ mmu for $C_{15}H_{22}O_3$] (20), 191 (10), 163 (70) and 162 (100).

**$NaBH_4$ reduction of 14**

A solution of **14** (575 mg) in pyridine (9 ml) was added to a suspension of $NaBH_4$ (504 mg) in pyridine (7 ml). The reaction mixture was stirred at room temperature for 6 h, after which it was acidified with aq. HCl (3 M) and then extracted with $Et_2O$ (60 ml). The extract was washed with dil. aq. HCl and water, dried ($MgSO_4$), and concentrated to give **18** as an oily crude product (200 mg). This was purified by column chromatography (hexane–EtOAc, 7:3 v/v) to afford an oil, $[\alpha]_D$ −63.9 (*c* 1.1 in $CHCl_3$); $\nu_{max}/cm^{-1}$ ($CHCl_3$) 3503, 2924, 2854 and 1720; $\delta_H(CDCl_3)$ 4.19 (1H, br s, OH), 3.85 (1H, ddd, *J* 11.7, 6.0, 5.8, H-4), 2.68 (1H, dd, *J* 6.3, 6.0, H-5), 2.28 (1H, dq, *J* 11.5, 7.2, H-11), 2.08 (1H, m, H-3β), 1.61 (1H, ddd, *J* 13.4, 6.8, 3.6, H-9β), 1.25 (1H, dddd, *J* 12.7, 12.7, 11.7, 3.6, H-3α), 1.09 (3H, d, *J* 7.2, H-13), 0.87 (3H, d, *J* 6.3, H-14), 0.83 (1H, dddd, *J* 12.8, 12.8, 12.8, 3.0, H-2β) and 0.55 (1H, dddd, *J* 11.4, 11.4, 11.4, 2.5, H-1); $\delta_C(CDCl_3)$ 225.3 (C, C-12), 70.2 (CH, C-4), 54.8 (CH, C-5), 45.0 (CH, C-6), 44.5 (CH, C-11), 43.3 (CH, C-1), 42.8 (CH, C-7), 35.3 (CH, C-10), 34.5 ($CH_2$, C-3), 31.7 ($CH_2$, C-9), 26.0 ($CH_2$, C-2), 25.0 ($CH_2$, C-8), 19.3 ($CH_3$, C-14) and 15.1 ($CH_3$, C-13); HREIMS *m/z* (rel. int %) 222.1617 [$M^+$, $\Delta = 0.2$ mmu for $C_{14}H_{22}O_2$] (28), 204 (60), 173 (40), 163 (50), 149 (100) and 148 (98).

**$NaBH_4$ reduction of 13**

Compound **13** was reduced under similar conditions to those employed with compound **14** to yield **16** as an oil, $[\alpha]_D$ −14.6 (*c* 0.58 in $CHCl_3$); $\nu_{max}/cm^{-1}$ ($CHCl_3$) 3502br, 2924, 2854 and 1720; $\delta_H(CDCl_3)$ 3.66 (3H, s, OMe), 3.59 (1H, dddd, *J* 11.7, 11.7, 4.9, 4.9, H-4), 2.30 (1H, dq, *J* 11.1, 6.8, H-11), 1.10 (3H, d, *J* 6.9, H-13) and 0.83 (3H, d, *J* 6.2, H-14); $\delta_C(CDCl_3)$ 177.7 (C, C-12), 71.8 (CH, C-4), 51.4 ($CH_3$, OMe), 43.7 (CH, C-7), 42.9 (CH, C-1), 42.3 (CH, C-11), 36.0 (CH, C-6), 35.6 ($CH_2$, C-9), 30.4 ($CH_2$, C-3), 30.1 ($CH_2$, C-5), 27.4 (CH, C-10), 26.5 ($CH_2$, C-8), 26.3 ($CH_2$, C-2), 19.6 ($CH_3$, C-14) and 15.0 ($CH_3$, C-13); HREIMS *m/z* (rel. int %) 254.1888 [$M^+$, $\Delta = -0.6$ mmu for $C_{15}H_{26}O_3$] (1), 236 (2), 205 (1), 177 (2), 149 (100), 121 (2), 107 (3), 93 (3) and 88 (52).

**Preparation of the cyclohexane dione 12 from isopulegol**

A solution of (−)-isopulegol (8.0 g) in acetone (30 ml) was cooled to 0–5 °C and to it was added cooled Jones oxidation reagent at a rate sufficient to maintain the temperature of the reaction mixture at *ca*. 20 °C. The reaction mixture was stirred for a further 1.5 h, after which it was extracted with light petroleum (bp 40–60 °C; 3 × 30 ml). The combined extracts were washed successively with saturated brine (2 × 30 ml), saturated aq. $NaHCO_3$ (2 × 30 ml) and then again saturated brine (30 ml), after which they were dried ($MgSO_4$) and evaporated under reduced pressure to give 2-isopropenyl-5-methylcyclohexanone (5.5 g) as an oil, $[\alpha]_D$ −3.10 (*c* 3.8 $CHCl_3$); $\nu_{max}/cm^{-1}$ ($CHCl_3$) 3079, 3024, 3013, 2930, 2872, 1703 and 1647; $\delta_H(CDCl_3)$ 4.94 (1H, s), 4.72 (1H, s), 2.96 (1H, dd, *J* 12.9, 5.4), 2.41 (1H, ddd, *J* 13.0, 3.5, 2.2), 1.75 (3H, s) and 1.03 (3H, d, *J* 6.1); $\delta_C(CDCl_3)$ 209.8 (C), 143.6 (C), 112.8 ($CH_2$), 57.9 (CH), 50.7 ($CH_2$), 35.3 ($CH_2$), 34.0 ($CH_2$), 31.3 ($CH_2$), 22.3 ($CH_3$) and 21.3 ($CH_3$). A solution of 2-isopropenyl-5-methylcyclohexanone (100 mg) in THF (1 ml) was added dropwise to a solution of lithium diisopropylamine (LDA) in THF at −78 °C [prepared from BuLi (1.6 M; 0.5 ml) diisopropylamine (0.5 ml) and THF (3 ml)]. After being stirred for 30 min, the mixture was treated with 3-trimethylsilylbut-3-en-2-one[14] (140 mg) in THF (1 ml), added dropwise, after which stirring was continued at −78 °C for 1 h. The reaction mixture was then allowed to warm to 0 °C (2.5 h) after which it was acidified with 10% aq. HCl (pH *ca*. 3) and stirred for a further 15 min. It was then neutralized with 5% aq. $NaHCO_3$ and extracted with EtOAc. The extract was washed, dried and concentrated to give **12** as a crude product (55 mg). This was purified by column chromatography (15% EtOAc–hexane) to afford an oil, $[\alpha]_D$ +27.2 (*c* 1.6 in $CHCl_3$); $\nu_{max}/cm^{-1}$ ($CHCl_3$); 2959, 2934, 2860 and 1708; $\delta_H(CDCl_3)$ 4.92 (1H, s), 4.70 (1H, s), 3.00 (1H, d, *J* 13.0, 5.2), 2.58 (1H, ddd, *J* 17.2, 9.2, 5.4), 2.39 (1H, ddd, *J* 17.2, 9.1, 6.3), 2.12 (3H, s), 1.74 (3H, s) and 1.10 (3H, d, *J* 5.9); HREIMS *m/z* (rel. int %) 222.1617 ($M^+$, $\Delta = 0.3$ mmu for $C_{14}H_{22}O_2$) (85), 207 (20), 164 (70) and 109 (100).

**Robinson annulation of 12**

To a solution of the diketone **12** (50 mg) in EtOH (4 ml) was added $Ba(OH)_2 \cdot 8H_2O$. The mixture was stirred at room temperature for 1.5 h and then neutralized with 10% aq. HCl and concentrated under reduced pressure. The residue was extracted with $CH_2Cl_2$ and the extract was washed, dried and evaporated under reduced pressure. The crude product was purified by HPLC (23% EtOAc–hexane) to give **21** (20 mg) ($R_t$ 13.34 min) and **20/22** as a mixture (5 mg) ($R_t$ 13.07 min).

**Compound 21.** Oil, $[\alpha]_D$ −205.4 (*c* 0.28 in $CHCl_3$); $\nu_{max}/cm^{-1}$ ($CHCl_3$) 3011, 2961, 2930 and 2874; $\delta_H(CDCl_3)$ 5.76 (1H, d, *J* 2.0, H-5), 2.76 (1H, ddd, *J* 13.7, 3.7, 3.7, H-8), 2.42 (1H, m, H-3), 1.83 (1H, ddd, *J* 12.8, 7.5, 3.7, H-9), 1.77 (3H, s, H-12), 1.76 (3H, s, H-13), 1.25 (1H, dddd, *J* 13.0, 13.0, 13.0, 3.9, H-9) and 1.03 (3H, d, *J* 6.4, H-14); $\delta_C(CDCl_3)$ 200.3 (C, C-4), 164.0 (C, C-6), 133.1 (C, C-7), 129.6 (C, C-11), 126.5 (CH, C-5), 45.4 (CH, C-1), 39.6 (CH, C-10), 36.4 ($CH_2$, C-3), 34.7 ($CH_2$, C-9),

30.1 ($CH_2$, C-8), 27.0 ($CH_2$, C-2), 22.8 ($CH_3$, C-13), 20.9 ($CH_3$, C-12) and 20.1 ($CH_3$, C-14); HREIMS *m/z* (rel. int %) 204.1512 ($M^+$, $\Delta$ = 0.2 for $C_{14}H_{20}O$) (10), 176 (10), 161 (100), 134 (55) and 122 (60).

**Compound 20/22 (mixture of isomers).** Oil, HREIMS *m/z* (rel. int %) 204.1511 [$M^+$, $\Delta$ = 0.3 mmu for $C_{14}H_{20}O$] (100), 189 (44), 161 (47), 147 (55), 133 (74), 105 (63), 91 (72) and 77 (37). Compound **20** $\delta_H(CDCl_3)$ 5.77 (1H, s), 4.98 (1H, d, *J* 1.7), 4.79 (1H, s), 2.79 (1H, dd, *J* 12.5, 2.5), 1.71 (3H, s), 1.05 (3H, d, *J* 6.3). Compound **22** $\delta_H(CDCl_3)$ 5.91 (1H, s), 4.96 (1H, s), 4.74 (1H, s), 3.02 (1H, m), 1.71 (3H, s) and 1.01 (3H, d, *J* 6.3).

### Preparation of the diketone cyclohexanones 7 and 23 from isopulegol

To a solution of (−)-isopulegol (1.08 g) in THF (10 ml), was added dropwise a solution of $BH_3$·THF complex in THF soln. (1 M; 7 ml) with ice-cooling. After being stirred for 1 h at room temperature, the mixture was treated with THF–water (1:1; 4 ml), followed by a mixture of aq. KOH (10%; 5 ml) and aq. $H_2O_2$ (50%; 2 ml). After this, the reaction mixture was stirred for 5 min, and then filtered and evaporated. The residue was washed with water and taken up into $Et_2O$ and the ethereal solution was dried ($MgSO_4$) and evaporated to give a white solid. This was purified by sublimation (*ca*. 60 °C, 10 mmHg) to yield 9-hydroxyisopulegol (0.85 g) as crystals, mp 88–89 °C; $\delta_H(CDCl_3)$ 4.58 (2H, br s), 3.63 (1H, dd, *J* 11.8, 5.2), 3.57 (1H, dd, *J* 11.5, 3.4), 3.42 (1H, ddd, *J* 10.5, 10.5, 3.9), 1.95 (1H, br d, *J* 17), 1.21 (1H, ddd, *J* 12.7, 12.7, 3.5), 0.95 (3H, d, *J* 7.3) and 0.91 (3H, d, *J* 6.3); $\delta_C(CDCl_3)$ 69.8 (CH), 66.8 ($CH_2$), 48.6 (CH), 44.4 ($CH_2$), 38.6 (CH), 34.6 ($CH_2$), 31.4 (CH), 29.6 ($CH_2$), 22.1 ($CH_3$) and 11.9 ($CH_3$); HREIMS *m/z* (rel. int %) 172.1458 [$M^+$, $\Delta$ = 0.5 mmu for $C_{10}H_{20}O_2$] (5), 154 (10), 139 (17), 124 (42), 112 (49), 95 (47) and 81 (100). A solution of 9-hydroxyisopulegol (2.74 g) in THF–DMF (4:1; 35 ml) was added slowly to a suspension of NaH (60% suspension in paraffin) (2.45 g) in THF–DMF (70 ml). The reaction mixture was stirred for 1 h after which it was treated with a solution of benzyl chloride (3.02 g) in THF–DMF (10 ml); stirring was then continued for 3 h at *ca*. 10 °C. After this, the reaction mixture was filtered, added to ice–water (100 ml) and extracted with $Et_2O$ (2 × 100 ml). The combined extracts were evaporated to give 9-benzyloxylisopulegol (2.76 g) as a yellow liquid; $\delta_H(CDCl_3)$ 7.33 (5H, m), 4.55 (1H, d, *J* 12), 4.48 (1H, d, *J* 12), 3.64 (1H, br s), 3.50 (1H, dd, *J* 9.1, 6.2), 3.40 (1H, dd, *J* 9.1, 3.7), 1.13 (1H, dddd, *J* 12.7, 12.7, 12.7, 3.4), 0.96 (3H, d, *J* 7.1) and 0.91 (3H, d, *J* 6.6); $\delta_C(CDCl_3)$ 137.7 (C), 128.3 (CH × 2), 127.6 (CH × 3), 74.3 ($CH_2$), 73.3 ($CH_2$), 70.4 (CH), 48.9 (CH), 44.9 ($CH_2$), 35.4 (CH), 34.6 ($CH_2$), 31.4 (CH), 27.8 ($CH_2$), 22.1 ($CH_3$) and 13.5 ($CH_3$). A solution of 9-benzyloxylisopulegol (1.34 g) in $CH_2Cl_2$ (5 ml) was added to a suspension of pyridinium chlorochromate (4.83 g) in $CH_2Cl_2$ (20 ml) and the mixture was stirred for 24 h. It was filtered, washed with water (30 ml) and 10% aq. $Na_2SO_3$ (30 ml), dried ($MgSO_4$) and evaporated to yield 9-benzyloxylisopulegone (1.13 g) as a yellow liquid; $[\alpha]_D$ − 8.3° (*c* 1.4 in $CHCl_3$); $\nu_{max}/cm^{-1}$ ($CHCl_3$) 2959, 2930, 2872 and 1705; $\delta_H(CDCl_3)$ 7.31 (5H, m), 4.48 (1H, d, *J* 12.3), 4.46 (1H, d, *J* 12.3), 3.47 (1H, dd, *J* 9.0, 5.4), 3.38 (1H, dd, *J* 9.0, 6.1), 1.01 (3H, d, *J* 6.8) and 0.99 (3H, d, *J* 6.1); $\delta_C(CDCl_3)$ 212.0 (C), 138.7 (C), 128.2 (CH), 127.5 (CH), 73.0 ($CH_2$), 72.9 ($CH_2$), 52.2 (CH), 50.9 ($CH_2$), 35.3 (CH), 34.1 ($CH_2$), 32.6 (CH), 29.4 ($CH_2$), 22.3 ($CH_3$) and 15.3 ($CH_3$); HREIMS *m/z* (rel. int %) 260.1774 [$M^+$, $\Delta$ = 0.2 mmu for $C_{17}H_{24}O_2$] (6), 202 (35), 169 (31), 153 (21), 151 (17) and 91 (100). A solution of 9-benzyloxyisopulegone (2.16 g) in THF (3 ml) was added dropwise to a solution of LDA in THF at −78 °C [prepared from BuLi (1.6 M in hexane; 7.1 ml), diisopropylamine (1.6 ml) and THF (3 ml)]. After being stirred for 30 min, the mixture was treated with a solution of 3-trimethylsilylbut-3-en-2-one[14] (2.26 g) in THF (3 ml), added dropwise, and then stirred for 1 h at −78 °C and then for 2 h at 0 °C. It was then treated with 10% aq. HCl to bring it to pH *ca*. 3. After this the reaction mixture was stirred at 0 °C for 15 min, and then extracted with EtOAc. The extract was washed with water to neutrality, dried ($MgSO_4$) and evaporated to afford **23** as a pale yellow liquid (1.01 g). This was purified by means of column chromatography (hexane–EtOAc, 7:3, v/v). Compound **7** was also obtained as a minor product from the column. Compound **23** $\delta_H(CDCl_3)$ 7.31 (5H, m), 4.49 (1H, d, *J* 12.8), 4.45 (1H, d, *J* 12.8), 3.45 (1H, dd, *J* 9.1, 4.9), 3.37 (1H, dd, *J* 9.1, 5.7), 2.10 (3H, s), 1.05 (3H, d, *J* 5.9) and 1.00 (3H, d, *J* 6.8). Compound **7** $\delta_H(CDCl_3)$ 7.32 (5H, m), 4.48 (2H, s), 3.30 (2H, m), 2.10 (3H, s), 1.06 (3H, d, *J* 5.8) and 0.87 (3H, d, *J* 7.0).

### Robinson annulation of 23

Compound **23** (0.27 g) was added to a suspension of $Ba(OH)_2$·$8H_2O$ (0.25 g) in EtOH (8 ml). After being stirred at room temperature for 2.5 h, the reaction mixture was acidified (10% aq. HCl) and evaporated under reduced pressure. The residue was taken up in $CH_2Cl_2$ and the solution washed with water, dried ($MgSO_4$) and evaporated to afford a colourless liquid (0.07 g). A mixture of the colourless liquid (0.07 g) and oxalic acid (0.08 g) in EtOH (3.2 ml) was refluxed for 2.5 h and then neutralized (10% aq. $NaHCO_3$) and concentrated under reduced pressure. This residue was extracted with $CH_2Cl_2$. The extract was washed with water and evaporated to afford **24** as a pale yellow oil (0.04 g). This was purified by means of HPLC [hexane–EtOAc (3:1, v/v)]; $\delta_H(CDCl_3)$ 7.32 (5H, m), 5.88 (1H, s), 4.51 (1H, d, *J* 12.1), 4.45 (1H, d, *J* 12.1), 3.48 (1H, dd, *J* 9.1, 3.3), 3.35 (1H, dd, *J* 9.1, 6.0), 1.07 (3H, d, *J* 6.4) and 1.02 (3H, d, *J* 6.4); HREIMS *m/z* (rel. int %) 312.2088 [$M^+$, $\Delta$ = 0.1 mmu for $C_{21}H_{28}O_2$] (46), 254 (5), 221 (30), 191 (100), 164 (70), 119 (9) and 91 (33).

On storage for 1 year **24** was converted into **25**, which was purified by HPLC as above; $\delta_H(CDCl_3)$ 7.32 (5H, m), 5.82 (1H, d, *J* 1.3), 4.39 (2H, s), 3.24 (1H, dd, *J* 9.2, 4.3), 3.11 (1H, dd, *J* 9.2, 6.5), 1.05 (3H, d, *J* 6.6) and 1.01 (3H, d, *J* 6.1); HREIMS *m/z* (rel. int %) 312.2071 [$M^+$, $\Delta$ = 1.8 mmu for $C_{21}H_{28}O_2$] (7), 221 (26), 209 (17), 182 (42), 163 (29),124 (49) and 91 (100).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