# Carotenoids and Related Compounds. Part 42.[1] Structure of Isomytiloxanthin

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Isomytiloxanthin, a pigment from the edible mussel *Mytilus edulis*, gave a monoacetate on acetylation, and a tetrahydro derivative on reduction with sodium borohydride. Spectroscopic studies on isomytiloxanthin and its derivatives showed that the natural pigment has the acetylenic structure 6,3'-dihydroxy-7',8'-didehydro-5,6,7,8,-tetrahydro-β,β-carotene-3,8-dione (**1**).

Reaction of but-1-yne with the ketone (**10**) gave the acetylenic alcohol (**12**) which, on hydroboration, yielded the ketone (**14**). Removal of the protecting group furnished (**16**), a model for the novel β-hydroxy ketone end group in isomytiloxanthin. Comparison of the n.m.r. bands of (**16**), and those of isomytiloxanthin, indicated that the latter has the relative configuration at 5,6 shown in (**1**).

A new carotenoid, isomytiloxanthin,‡ was encountered during the isolation of mytiloxanthin from the edible mussel, *Mytilus edulis*.[1] In this paper we detail the evidence for its formulation as (**1**).[2,3]

The molecular formula, $C_{40}H_{54}O_4$, like those of the key derivatives mentioned below, was established unambiguously by mass spectrometry. The fragmentation pattern, and n.m.r. studies, before and after deuteriation, revealed the presence of two hydroxy groups, but the test for an allylic hydroxy with hydrogen chloride in chloroform proved negative.

Treatment of isomytiloxanthin with acetic anhydride in pyridine gave a monoacetate (**2**) as the sole product. The retention of one hydroxy group was confirmed by the fragmentation, the observation of the appropriate metastable ion for loss of the elements of water, and by n.m.r. studies. Since this hydroxy group also resisted trimethylsilylation it was regarded as sterically hindered and probably tertiary.

The i.r. spectrum of isomytiloxanthin, in addition to bands associated with the hydroxy groups, revealed strong absorptions attributable to both a conjugated and an unconjugated carbonyl group, thus accounting for all four oxygen atoms. The i.r. spectrum, like those of its derivatives, also included a weak band similar to that exhibited by alloxanthin (**4**) and assigned to the carbon–carbon triple band stretching frequency in a conjugated acetylenic linkage.[4]

Reduction of isomytiloxanthin with sodium borohydride was accompanied by a shift in the light absorption maximum by *ca.* 20 nm to shorter wavelengths, indicating the loss of a conjugated carbonyl group from the main chromophore. The resulting tetrahydro derivative was shown by n.m.r. studies, before and after deuteriation, to contain four hydroxy groups, as expected. This 'isomytiloxanthinol' (**3**) exhibited a visible light absorption curve and maximum very similar to those of the borohydride reduction product of mytiloxanthin,[1] and underwent allylic dehydration on treatment with hydrogen chloride in chloroform.

The $^1$H n.m.r. spectrum of isomytiloxanthinol, like that of isomytiloxanthin (Table 1), contained a set of bands very similar to those in the spectrum of alloxanthin (**4**), indicating that one half of all three carotenoids were identical. This conclusion was supported by the $^{13}$C n.m.r. spectrum of isomytiloxanthin which included 18 signals at positions close to ones observed with alloxanthin[5] (Table 2). These bands were assigned by comparisons with those in the spectrum[5,6] of zeaxanthin (**5**), and with the unambiguous assignments of the bands in the broad band decoupled spectra of (**8**) and (**9**)[4] made after examining the $^1$H–$^{13}$C spin-spin couplings under single frequency off resonance conditions.

The results described above established the structure of isomytiloxanthin, apart from novel end which will now be considered.

The mass spectrum of isomytiloxanthin contained a strong line (68% of base peak) due to a fragment with the composition $C_9H_{15}O_2$. This was rationalised as the result of cleavage of the 6,7-bond by a retro-aldol type fission of a β-hydroxy ketone. Charge retention on the other moiety, with transfer of a hydrogen atom, gave an ion $C_{31}H_{40}O_2$. α-Cleavage on both sides of the conjugated carbonyl group was also observed. Isomytiloxanthin acetate (**2**), and the model compounds (**15**) and (**16**) (see below), showed analogous fragmentation patterns.

Support for the β-hydroxy ketone structure was obtained by a series of reactions carried out on a spectroscopic scale. Treatment of isomytiloxanthin with alkali produced an irreversible bathochromic shift of 13 nm, attributed to dehydration of the β-hydroxy ketone (*cf.* reversible shift with mytiloxanthin[1]). Subsequent reduction with sodium borohydride gave a product with light absorption properties similar to those of crocoxanthin (**6**),[4] presumably as the result of anionotropic rearrangement of the bis-allylic alcohol formed initially.

The $^1$H n.m.r. spectrum of isomytiloxanthin included a sharp singlet, integrating for two protons, at δ 2.84. This was consistent with a deshielded methylene group at C-7 and the absence of protons at either C-6 or C-8. A band was observed at the same position in the spectrum of the model (**16**), but not in that of isomytiloxanthinol (**3**). The spectra of isomytiloxanthin and its derivatives also included two three-proton singlets attributable to the geminal methyls at C-1, and a three-proton doublet assigned to the remaining methyl at C-5, the spin-spin coupling indicating that a hydrogen atom was also attached to C-5. The positions of these three methyl bands with isomytiloxanthin were very similar to those in the spectrum of isomytiloxanthinol. This indicated that the (unconjugated) carbonyl group was not adjacent to either methyl bearing carbon atom. It was therefore assigned to position 3.

The two models were prepared as follows. Reaction of the ketone (**10**)[7] with acetylene gave the acetylenic alcohol (**11**) which, on hydroboration, yielded the aldehyde (**13**). Removal of the protecting group then furnished the required keto aldehyde

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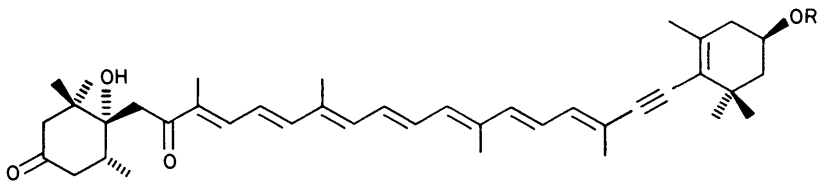
‡ See abstract and the experimental section for the naming of isomytiloxanthin and its derivatives according to the recommendations of the IUPAC-IUB Commission on Biochemical Nomenclature (*Pure Appl. Chem.*, 1975, **41**, 407). Positions in carotenoids are numbered according to the same recommendations, as illustrated in formula (**5**).



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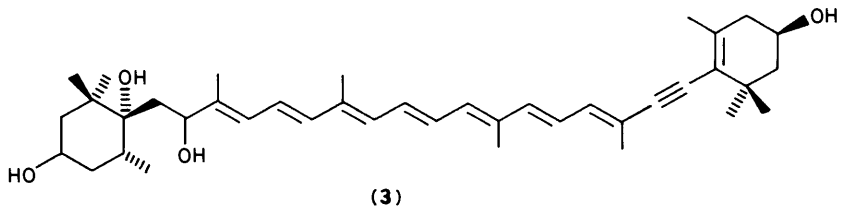

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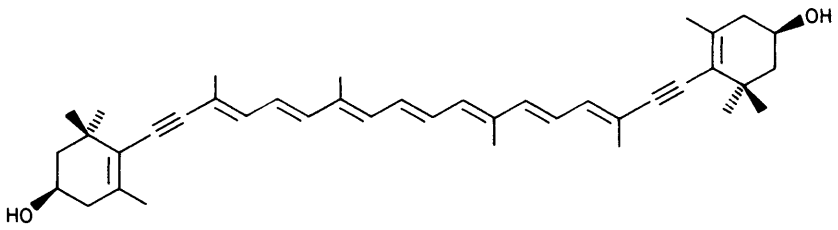

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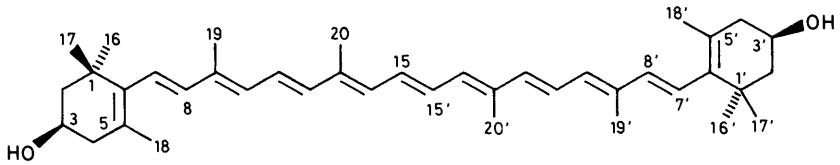

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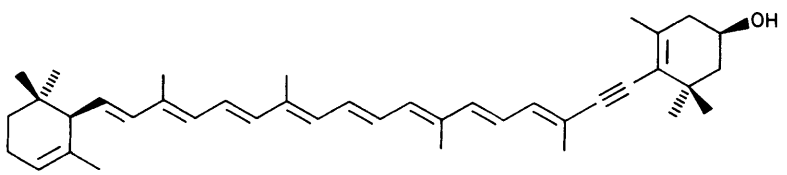

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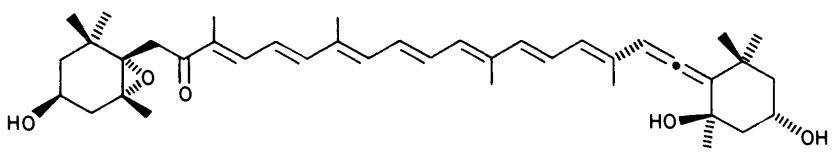

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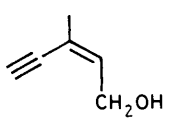

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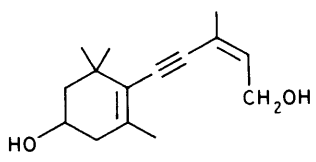

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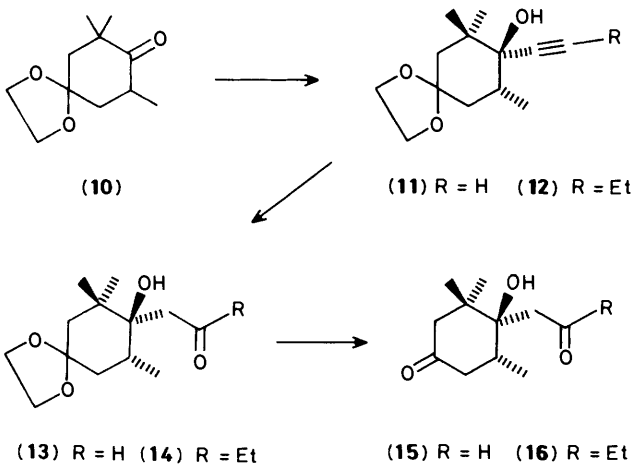
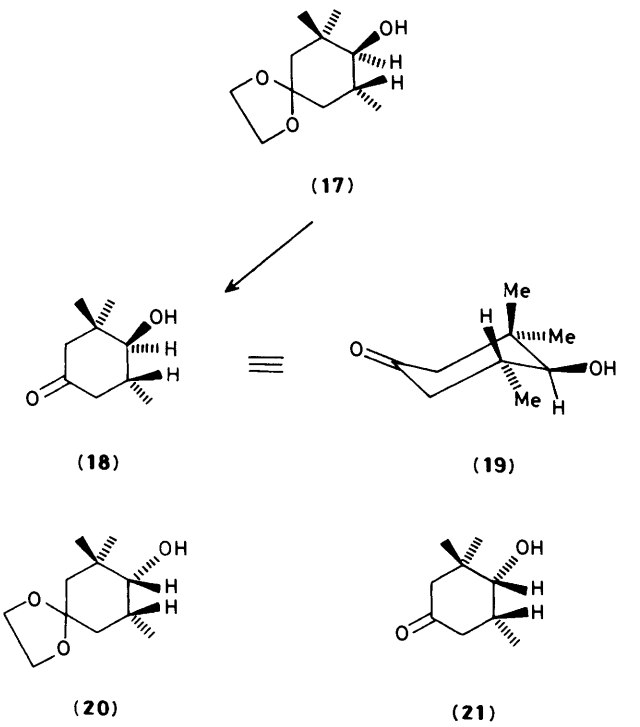
**Table 1.** Principal $^1$H n.m.r. bands (δ values) of carotenoids and the diketone (**16**)[a]

| Compound | 1-$Me_2$ | 5-Me | 9-Me | 13-Me | 13′-Me | 9′-Me | 5′-Me | 1′-$Me_2$ | Other bands |
|---|---|---|---|---|---|---|---|---|---|
| Alloxanthin (**4**) | 1.14, 1.19 | 1.90 | 1.99 | 1.94 | 1.94 | 1.99 | 1.90 | 1.19, 1.14 | 1.50 (s[b], 2 H), 2.29 (d *J* 7, 4 H) |
| Isomytiloxanthin (**1**) | 1.09, 1.26 | 0.98 (d, *J* 6.5[c]) | 1.94 | 1.98 | 1.98 | 1.98 | 1.91 | 1.20, 1.14 | 1.64 (s[b], 2 H), 2.14 (s, 2 H), 2.84 (s, 2 H) |
| Isomytiloxanthin Acetate (**2**) | 1.09, 1.26 | 0.98 (d, *J* 6.5) | 1.94 | 1.98 | 1.98 | 1.98 | 1.92 | 1.20, 1.18 | 1.53 (s[b], 1 H), 2.03 (s, 3 H, OAc), 2.14 (s, 2 H), 2.88 (s, 2 H) |
| Isomytiloxanthinol (**3**) | 1.08, 1.26 | 0.97 (d, *J* 6.5) | 1.94 | 1.98 | 1.98 | 1.98 | 1.91 | 1.20, 1.15 | 1.55 (br s[b], 4 H) |
| Diketone [d](**16**) | 0.98, 1.05 | 0.99 (d, *J* 6.5) | 1.12 (t, *J* 7.5) | | | | | | 1.95 (br s[b], 1 H) 2.00—2.50 (m, 5 H), 2.63 (q, *J* 7.5, 2H, $CH_2CH_3$), 2.83 (s, 2 H, $CH_2CO$) |

[a] All bands had the expected relative intensities. Methyl bands were observed as singlets, unless indicated to the contrary. The assignment of the bands due to methyl groups on $sp^2$ hybridised carbon atoms, and which differ by less than 5 p.p.m., is arbitrary. [b] Band due to hydroxy proton(s); removed after shaking the solution of the sample with deuterium oxide. [c] The coupling constant (*J*) was measured at both 60 and 100 MHz. [d] In this Table the positions in (**16**) are designated according to conventional carotenoid numbering, for ease of comparison.



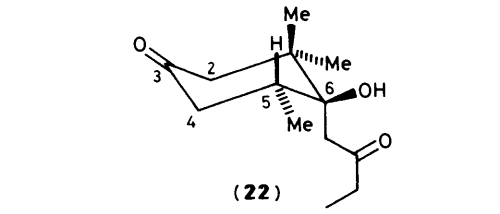




(**15**). The corresponding di-ketone (**16**) was similarly prepared from (**10**) and but-1-yne.

Sodium borohydride reduction of (**10**) gave the alcohol (**17**) which on hydrolysis yielded the keto alcohol (**18**). In the $^1$H n.m.r. spectrum of both (**17**) and (**18**) the coupling constant between the protons at C-5 and C-6 (carotenoid numbering) was found to be 9.5 Hz, indicating a *trans* diaxial stereochemistry between them [*e.g.* (**19**)], as would be expected from hydride attack on the carbonyl group in the preferred conformation of (**10**) with the methyl at C-5 in the equatorial position. The two products were, therefore, assigned the relative configurations shown. The isomeric products (**20**) and (**21**) were obtained, as expected, by Meerwein-Pondorff reduction of (**10**) and subsequent removal of the protecting group.

From earlier studies it was considered that attack on the ketone (**10**) by the lithium derivative of either acetylene or but-1-yne would follow the same stereochemical course as attack by hydride ion. The models (**15**) and (**16**) were, therefore, assigned the relative configurations shown. With both models the $^1$H n.m.r. band associated with one of the geminal methyls differed markedly from that of the corresponding methyl in the novel end group of isomytiloxanthin, which was, therefore, assigned the opposite relative configuration at 5,6, as shown in (**1**). Consideration of the preferred conformation (**22**) of the model (**16**), and of the isomytiloxanthin end group (**23**), together with the corresponding Newman projections for the 1,6- bonds, (**24**) and (**27**), and the 5,6- bonds, (**25**) and (**26**), indicated that the chemical environment of the C-5 methyl in the two series should be similar, likewise that for one of the geminal methyls. The observed chemical shifts were in good agreement with this analysis. The conclusion that the natural end group has the structure and stereochemistry shown in (**23**) was consistent with the associated $^{13}$C n.m.r. bands (Table 3). The presence of both carbonyl groups was confirmed by the resonances at 201.0 and 208.6 p.p.m. The similarity in the positions of the line due to the carbon atom in the unconjugated carbonyl of (**16**) and isomytiloxanthin confirmed the location of this group at C-3 in the natural carotenoid.

Isomytiloxanthin may be biogenetically related to fucoxanthin (**7**),[8,9] a common pigment in marine algae. The possible formation of the (3*R*)- alloxanthin type end group from the allenic end group in fucoxanthin has been discussed previously.[10] Hydride reduction of the epoxide in the other end group of fucoxanthin, and oxidation at C-3, would give the new end group found in isomytiloxanthin. It is tentatively suggested that (**1**) also represents the absolute configuration of isomytiloxanthin.

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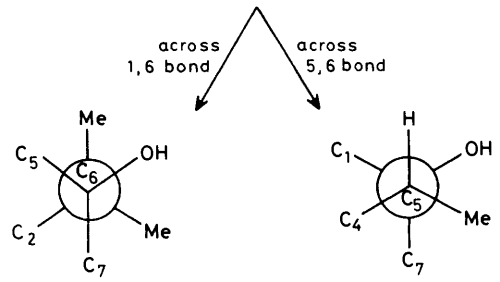


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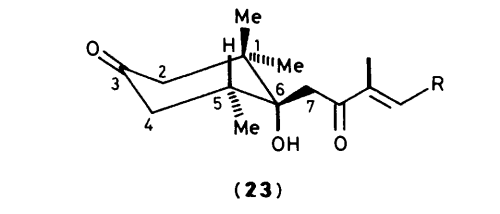


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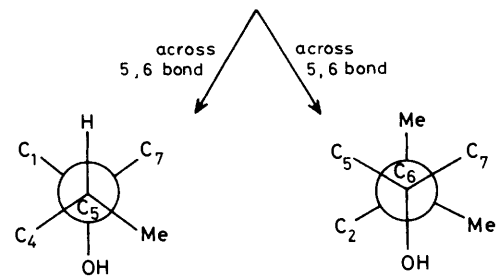


(26) (27)

## Experimental

The general comments in the Experimental section of Part 40* apply. $^{13}C$ N.m.r. spectra were recorded on a Brüker HFX-90 spectrometer at 22.63 MHz as deuteriochloroform solutions.

*Isolation of Isomytiloxanthin* (**1**).—The less polar pigment from *Mytilus edulis*, reported in the preceding paper, was further purified by chromatography first on a column of icing sugar (light petroleum–benzene), and then as a column of silica gel (8% acetone in benzene), giving *isomytiloxanthin* (30 mg) as a red gum which resisted all attempts at crystallisation; $\lambda_{max.}$(ethanol) 474infl and 451 nm; $\lambda_{max.}$(hexane) 474 and 447 nm; $\lambda_{max.}$(benzene) 486 (ε 111 000) and 459 nm; $\lambda_{max.}(CS_2)$ 504 and 476 nm; $\nu_{max.}(CHCl_3)$ 3 610, 2 975, 2 930, 2 170, 1 710, 1 640, 1 610, 1 175, 1 050, 1 028, and 970 $cm^{-1}$; δ, see Table 1; $^{13}C$ n.m.r., see Table 2; *m/z* 598.401 ($M^{+\bullet}$; Calc. for $C_{40}H_{54}O_4$: *m/z* 598.402, 6), 580 (*M* − 18, 1.9; *m** 562.0, $580^2/598$ = 562.5), 565 (*M* − 18 − 15, 0.5; *m** 550.0, $565^2/580$ = 550.4), 562 (*M* − 18 − 18, 0.5), 540 (*M* − 58, 0.5), 522 (*M* − 18 − 58, 0.5), 506 (*M* − 92, 3), 444.302 (*M* − 154; Calc. for $C_{31}H_{40}O_2$: *m/z* 444.303, 10), 429.278 (*M* − 169: Calc. for $C_{30}H_{37}O_2$: *m/z* 429.279, 1.2), 411 (429 − 18, 0.7), 401.283 (*M* − 197; Calc. for $C_{29}H_{37}O$: *m/z* 401.284, 2.4) 383 (0.8), 197.117 (Calc. for $C_{11}H_{17}O_3$: *m/z* 197.118, 4), 169.122 (Calc. for $C_{10}H_{17}O_2$: *m/z* 169.123, 3.7), 156 (23), 155.107 (Calc. for $C_9H_{15}O_2$: *m/z* 155.107, 68), 119 (10), 113 (155 − 42, 51; *m** 82.4, $113^2/155$ = 82.4), 111 (39), 105 (9), 97 (155 − 58, 22; *m** 60.7, $97^2/155$ = 60.7), 92 (35), 91 (48), and 43 (100); c.d. (dioxane) 228 nm (Δε −1.14) and 214 nm (Δε −1.82).

No reaction was observed when isomytiloxanthin was treated with hydrogen chloride in chloroform. On treatment with ethanolic sodium hydroxide the light absorption maximum of the solution shifted slowly (90 min) from 452 to 465 nm; on subsequent addition of an excess of acetic acid there was no further change in $\lambda_{max.}$. Reduction of the alkali product in ethanol with sodium borohydride gave a solution with $\lambda_{max.}$ 472infl and 446 nm. Crocoxanthin has $\lambda_{max.}$(ethanol) 477 and 447 nm.[4]

*Isomytiloxanthin Acetate* (**2**).—Acetic anhydride (0.4 ml) was added to isomytiloxanthin (2 mg) in pyridine (0.5 ml) and the reaction was monitored by t.l.c. on Kieselgel H (acetone–light petroleum, 1:4); only one product was detected. When the reaction was complete ether was added and the solution was washed thoroughly with water, dried, and evaporated. Chromatography of the residue on a column of silica gel (benzene) gave *isomytiloxanthin acetate*; $\lambda_{max.}$(ethanol) 474infl and 452 nm; $\lambda_{max.}$(benzene) 484infl and 458 nm; $\nu_{max.}$(KBr) 2 170, 1 735, 1 710, 1 640, 1 610, 1 025, 965, 890, and 785 $cm^{-1}$; δ, see Table 1; *m/z* 640.412 ($M^{+\bullet}$; Calc. for $C_{42}H_{56}O_5$: *m/z* 640.413, 17), 622.401 (*M* − 18; Calc. for $C_{42}H_{54}O_4$: *m/z* 622.402, 1.5), 582 (*M* − 58, 1.7), 580 (*M* − 60, 2.1; *m** 525.6, $580^2/640$ = 525.62), 564 (*M* − 58 − 18, 1), 548 (*M* − 92, 1.5), 486.313 (*M* − 154, Calc. for $C_{33}H_{42}O_3$: *m/z* 486.313, 15), 471.290 (*M* − 169; Calc. for $C_{32}H_{39}O_3$: *m/z* 471.290, 0.5), 443.294 (*M* − 197; Calc. for $C_{31}H_{39}O_2$: *m/z* 443.295, 1.5), 426 (*M* − 154 − 60, 1.5; *m** 373.5, $426^2/486$ = 373.4), 411 (2), 383 (2.5), 197 (3.2), 169 (3.8), 156 (35), 155.107 (Calc. for $C_9H_{15}O_2$: *m/z* 155.107, 55), 119 (19), 113 (155 − 42, 100; *m** 82.4, $113^2/155$ = 82.4), 111 (23), 105 (14), 97 (155 − 58, *m** 60.7, $97^2/155$ = 60.7), 91 (23), and 43 (98).

Isomytiloxanthin acetate was resistant to trimethylsilylation at 20 °C. On reduction in ethanol with sodium borohydride the solution had $\lambda_{max.}$ 454 and 427 nm; identical maxima were observed after similar reduction of mytiloxanthin.

*Isomytiloxanthinol* (**3**).—Sodium borohydride (10 mg) was added slowly to isomytiloxanthin (2.5 mg) in ether (5 ml) and methanol (5 ml) at 0 °C. The solution was stirred for 2 h, evaporated under reduced pressure, and the excess of hydride was destroyed by cautiously adding water. The product was isolated with ether in the usual way. Chromatography of the residue on a column of silica gel (acetone–benzene) gave *isomytiloxanthinol* as a red gum; $\lambda_{max.}$(ethanol) 456 and 428 nm; $\lambda_{max.}(CHCl_3)$ 464 and 436 nm; $\nu_{max.}$(KBr) 3 400, 2 160vw, 1 050, 1 020, and 960 $cm^{-1}$; δ, see Table 1; *m/z* 602.435 ($M^+$; Calc. for $C_{40}H_{58}O_4$: *m/z* 602.434, 2.6), 584 (*M* − 18, 3; *m** 565.5, $584^2/602$ = 566.5), 566 (*M* − 18 − 18, 1; *m** 584.5, $566^2/584$ = 548.5), 510 (*M* − 92, 2.1), 492 (*M* − 92 − 18, 2.5; *m** 474.5, $492^2/510$ = 474.6), 478 (*M* − 18 − 106), 446.318 (*M* − 156; Calc. for $C_{31}H_{42}O_2$: *m/z* 446.318, 0.5), 428 (0.5), 201.148 (Calc. for $C_{11}H_{21}O_3$: *m/z* 201.149, 1.7), 183 (201 − 18, 3.5), 171 (3.2), 158 (17), 157.123 (Calc. for $C_9H_{17}O_2$: *m/z* 157.123, 100), 139 (157 − 18, 10; *m** 123.0, $139^2/157$ = 123.1), 121 (2.8), 119 (5), 113 (157 − 44, *m** 81.3, $113^2/157$ = 81.3), 105 (5.5), 99 (157 − 58, 10; *m** 62.5, $99^2/157$ = 62.4), 92 (7), 91 (13), and 43 (50). The same product was obtained by reduction of isomytiloxanthin in ether with lithium aluminium hydride.

After a few drops of hydrogen chloride in chloroform had been added to isomytiloxanthinol in chloroform the solution

* Part 40, A. K. Chopra, G. P. Moss, and B. C. L. Weedon, *J. Chem. Soc., Perkin Trans. 1*, 1988, 1371.

**Table 2.** $^{13}C$ N.m.r. chemical shifts for C-1′ to C-20′ in isomytiloxanthin and related compounds (in $CDCl_3$; $\delta_{TMS} = 0$)

| | Methylpentenynol (**8**)[a] | $C_{15}$-glycol (**9**)[a] | Alloxanthin (**4**)[b] | Isomytiloxanthin (**1**)[b] | Zeaxanthin (**5**)[b] |
|---|---|---|---|---|---|
| Ring carbons | | | | | |
| 1′ | | 36.6 | 37.0 | 37.0 | 37.1 |
| 2′ | | 46.8 | 46.8 | 46.9 | 48.2 |
| 3′ | | 64.8 | 65.0 | 65.0 | 65.1 |
| 4′ | | 41.5 | 41.6 | 41.7 | 42.4 |
| 5′ | | 138.2 | 138.2 | 138.2 | 125.5 |
| 6′ | | 124.1 | 124.5 | 124.6 | 142.3 |
| Chain carbons | | | | | |
| 7′ | 83.1 | 93.2[c] | 89.3 | *d* | 126.1 |
| 8′ | 82.2 | 92.2[c] | 99.0 | *d* | 138.5 |
| 9′ | 118.9 | 121.6 | 119.1 | 119.9 | 135.7 |
| 10′ | 138.1 | 134.2 | 133.3 | 133.4 | 131.3 |
| 11′ | 60.7 | 61.7 | 136.3 | 136.4 | 124.9 |
| 12′ | | | 130.3 | 130.2 | 137.6 |
| 13′ | | | 135.2 | 135.4 | 136.5 |
| 14′ | | | 130.3 | 130.2 | 132.6 |
| 15′ | | | 137.2 | 136.7 | 130.0 |
| Methyl carbons[e] | | | | | |
| 16′ | | 28.8 | 28.9 | 28.9 | 28.7 |
| 17′ | | 30.6 | 30.6 | 30.7 | 30.2 |
| 18′ | | 23.5 | 22.5 | 22.8 | 21.7 |
| 19′ | 22.8 | 22.6 | 18.1 | 18.2 | 12.8 |
| 20′ | | | 12.8 | 12.9 | 12.8 |

[a] For ease of comparison, the carbon atoms are numbered as the corresponding positions in alloxanthin. [b] The assignment of the bands for C-10′ to C-15′ is tentative. [c] Assignments of the two signals marked [c] is arbitrary, and the two assignments may need to be reversed. [d] Not detected. [e] Assignments of the C-16′ and C-17′ signals is arbitrary, and the two assignments in any one of the four compounds concerned may need to be reversed.

**Table 3.** $^{13}C$ N.m.r chemical shifts for C-1 to C-8 and for C-16 to C-20 in isomytiloxanthin and synthetic models (in $CDCl_3$; $\delta_{TMS} = O$)

| | Hydroxyketone (**21**)[a] | Hydroxyketone (**18**)[a] | Model (**15**)[a] | Model (**16**)[a] | Isomytiloxanthin (**1**) |
|---|---|---|---|---|---|
| Ring carbons | | | | | |
| 1 | 40.0 | 39.2 | 42.9 | 43.0 | 41.7 |
| 2 | 48.7 | 53.8 | 52.4 | 52.2 | 50.8 |
| 3 | 213.5 | 209.9 | 209.9 | 209.5 | 208.5 |
| 4 | 43.3 | 48.0 | 46.2 | 45.9 | 48.3 |
| 5 | 33.3 | 35.3 | 38.9 | 38.8 | 40.7 |
| 6 | 76.9 | 81.3 | 66.1 | 77.2 | 73.5 |
| Chain carbons | | | | | |
| 7 | | | 42.7 | 43.0 | *b* |
| 8 | | | 217.6 | 215.4 | 201.0 |
| 9 | | | | 38.1 | *b* |
| Methyl carbons[c] | | | | | |
| 16 | 25.8 | 35.3 | 25.8 | 25.8 | 31.2 |
| 17 | 27.3 | 19.2 | 23.2 | 23.7 | 29.3 |
| 18 | 18.4 | 28.7 | 17.0 | 17.3 | 23.7 |
| 19 | | | | 7.6 | 14.9 |
| 20 | | | | | 12.1 |

[a] Carotenoid numbering, for ease of comparison. [b] Not assigned. [c]Assignments to C-16 and C-17 may need to be reversed.

had $\lambda_{max.}$ 476, 448, and 345 nm (*cis*-peak) indicating that elimination of an allylic hydroxy group had occurred.

4,4-*Ethylenedioxy*-1-*ethynyl*-2,2,6-*trimethylcyclohexanol* (**11**).—Acetylene was bubbled through a solution of lithamide (0.3 g) in liquid ammonia (75 ml) for 2 h. Ether (50 ml) was added and the ammonia was allowed to evaporate. To the solution of lithium acetylide thus obtained 4,4-ethylenedioxy 2,2,6-trimethylcyclohexanol[7] (2.0 g) in ether (50 ml) was added over 20 min. The solution was stirred overnight at 20 °C, and ammonium chloride (1.0 g) was then added. Isolation of the product with ether and crystallisation from ether–light petroleum gave the ethynyl alcohol (2.0 g), m.p. 88 °C; $\nu_{max.}$(KBr) 3 285 and 2 110 $cm^{-1}$; δ 1.09 (d, *J* 6.5 Hz, 3 H, 6-Me), 1.15 (s, 6 H, 2-$Me_2$), 1.62—1.78 (m, 4 H, 3-$CH_2$ and 5-$CH_2$), 2.10 (br s, 1 H, disappears on deuteriation, OH), 2.51 (s, 1 H,

C≡CH), and 3.91 (m, 4 H; $OCH_2CH_2O$); *m/z* 224 ($M^{+\bullet}$; Calc. for $C_{13}H_{20}O_3$: *m/z* 224, 0.5), 209 (*M* − 15, 0.5), 191 (*M* − 18, 0.5), 127 (20), 113 (100), and 96 (30).

2-(4′,4′-*Ethylenedioxy*-1′-*hydroxy*-2′,2′,6′-*trimethylcyclohexyl*)*ethanal* (**13**).—Diborane, from sodium borohydride (320 mg) in diglyme (5 ml) and boron trifluoride–diethyl ether (2.1 ml) in diglyme (2 ml), was passed through a solution of 2-methylbut-2-ene (0.8 ml) in tetrahydrofuran (5 ml) at 0 °C. After the solution had been kept at 0 °C for 2 h the preceding ethynyl alcohol (336 mg) in tetrahydrofuran (5 ml) was added rapidly the temperature of the mixture being maintained below 10 °C. The mixture was stirred for 2 h and then for a further 2 h at 20 °C. The excess of diborane was decomposed by careful addition of water (1 ml). Hydrogen peroxide (15%; 2.25 ml) and aqueous sodium hydroxide (3M; 2.25 ml) was added whilst the mixture was stirred vigorously (pH 8—9). The mixture was stirred at 0 °C for 30 min and then at 20 °C for 30 min. It was then neutralised by the addition of acid resin (IR-120H) and filtered. The filtrate was saturated with sodium chloride and the product was isolated with ether. Preparative t.l.c. on Kieselgel Hf (acetone–light petroleum, 1:4), and crystallisation from ether–light petroleum, gave the aldehyde (225 mg) as colourless needles, m.p. 96 °C; $\nu_{max.}$($CHCl_3$) 1 725 $cm^{-1}$; δ 0.95 (s, 6 H; 2′-$Me_2$), 1.09 (d, *J* 6.5 Hz, 3 H, 6′-Me), 1.65 (s, 4 H, 3′-$CH_2$ and 5′-$CH_2$), 2.61 (d, *J* 2.5 Hz, 2 H, $CH_2C{=}O$), 3.95 (m, 4 H, $OCH_2CH_2O$), and 10.00 (t, *J* 2.5 Hz, 1 H; CHO); *m/z* 242 ($M^{+\bullet}$; Calc. for $C_{13}H_{22}O_4$: *m/z* 242, 0.5), 227 (*M* − 15, 0.5), 224 (*M* − 18, 0.5), 213 (*M* − 29, 0.5), 199 (*M* − 43, 4.5), 198 (*M* − 44, 2.5), 185 (7), 129 (90), 127 (40), and 113 (100).

In one experiment, presumably owing to the presence of an excess of diborane, the product isolated was the corresponding primary alcohol, m.p. 81 °C; δ 0.96 (d, *J* 6.5 Hz, 3 H, 6′-Me), 0.97 (s, 3 H, 2′-Me), 1.10 (s, 3 H, 2′-Me), 1.57 (s, 4 H, 3′-$CH_2$ and 5′-$CH_2$), 1.81 (t, *J* 5.5 Hz, 2 H; 2-$CH_2$), 2.47 (br s, 2 H, disappears on deuteriation, both OH), 3.85 (t, *J* 5.5 Hz, 2 H, 1-$CH_2$), and 3.90 (m, 4 H, $OCH_2CH_2O$); *m/z* 244 ($M^{+\bullet}$; Calc. for $C_{13}H_{24}O_4$: *m/z* 244, 0.5), 229 (0.5), 199.134 (*M* − 45; Calc. for $C_{11}H_{19}O_3$: *m/z* 199.133, 7), 127 (11), 113 (21), 99 (24), 45 (25), and 31 (10).

2-(1′-*Hydroxy*-2′,2′,6′-*trimethyl*-4′-*oxocyclohexyl*)*ethanal* (**15**).—Sulphuric acid (0.5M; 1 ml) was added to the preceding aldehyde (10 mg) in acetone (1 ml) at 0 °C and the mixture was stirred at 0 °C for 3 h. An excess of saturated aqueous sodium hydrogen carbonate was added, the aqueous layer was saturated with sodium chloride, and the product was isolated with ether. Crystallisation from ether–light petroleum gave the *keto aldehyde* (5 mg), m.p. 78—79 °C; $\nu_{max.}$($CHCl_3$) 1 740 and 1 715 $cm^{-1}$; δ 1.04 (s, 6 H, 2′-$Me_2$), 1.04 (d, *J* 6.5 Hz, 3 H, 6′-Me), 2.04—2.60 (m, 5 H, 3′-$CH_2$, 5′-$CH_2$, and 6′-CH), 2.84 (d, *J* 2 Hz, 2 H, $CH_2CO$), and 10.03 (t, *J* 2 Hz, 1 H, CHO); $^{13}C$ n.m.r., see Table 2; *m/z* 198.125 ($M^{+\bullet}$; Calc. for $C_{11}H_{18}O_3$: *m/z* 198.126, 60), 183 (*M* − 15, 7; *m** 169.0, $183^2/198$ = 169.1), 180.114 (*M* − 18; Calc. for $C_{11}H_{16}O_2$; *m/z* 180.115, 11; *m** 163.7, $180^2/198$ = 163.6), 169 (0.5), 165 (*M* − 18 − 15, 5), 155.107 (*M* − 43; Calc. for $C_9H_{15}O_2$: *m/z* 155.107, 10), 154.099 (*M* − 44; Calc. for $C_9H_{14}O_2$: *m/z* 154.099, 15; *m** 119.9, $154^2/198$ = 119.8), 140 (*M* − 58, 8), 139 (154 − 15, 60; *m** 125.5, $139^2/154$ = 125.5), 114 (10), 113 (50), 100 (50), 97 (11), 83 (35), 71 (30), 70 (71), 69 (60), 57 (20), 56 (56), 55 (30), 44 (1.2), and 41 (100).

When the removal of the protecting group was carried out at 20 °C, dehydration of the tertiary hydroxy group also occurred to give (55%) 2-(2′,2′,6′-*trimethyl*-4′-*oxocyclohexylidene*)*ethanal* which crystallised from ether–light petroleum as colourless needles, m.p. 86 °C; $\nu_{max.}$($CHCl_3$) 1 720, 1 670, 1 620, and 1 600 $cm^{-1}$; δ 1.21 (s, 3 H, 2′-Me), 1.30 (s, 3 H, 2′-Me), 1.37 (d, *J* 7 Hz, 3 H, 6′-Me), 2.2—2.8 (m, 4 H, 3′-$CH_2$ and 5′-$CH_2$), 3.45 (q, *J* 7 Hz, 1 H, 6′-CH), 6.10 (d, *J* 8 Hz, 1 H, 2-H) and 10.10 (d, *J* 8 Hz, 1 H, CHO): *m/z* 180 ($M^{+\bullet}$; Calc for $C_{11}H_{16}O_2$: *m/z* 180, 60), 165 *M* − 15, 10), 151 (5), 138 (*M* − 42, 20), 137 (17), 124 (12), 123 (*M* − 42 − 15, 100; *m** 109.6, $123^2/138$ = 109.6), and 109 (15).

2-(1′-*Hydroxy*-2′,2′,6′-*trimethyl*-4′-*oxocyclohexyl*)*ethanol*. —Sulphuric acid (0.5M; 1 ml) was added to 2-(4′,4′-ethylenedioxy-1′-hydroxy-2′,2′,6′-trimethylcyclohexyl)ethanol (10 mg) in acetone (1 ml), and the mixture was stirred at 20 °C for 3 h. Isolation of the product in the usual way, and crystallisation from ether–light petroleum, gave the *keto glycol* (7 mg), m.p. 75 °C; $\nu_{max.}$($CHCl_3$) 3 610, 3 500, and 1 710 $cm^{-1}$; δ 1.05 (s, 6 H, 2′-$Me_2$), 1.05 (d, *J* 6.5 Hz, 3 H, 6′-Me), 2.00 (m, 3 H, 2-$CH_2$ and 6′-CH), 2.18 (s, 4 H, 3′-$CH_2$ and 5′-$CH_2$), 3.01 (br s, 1 H, disappears on deuteriation, OH), 3.53 (br s, 1H, disappears on deuteriation, 1 H, OH), and 3.98 (t, *J* 5.5 Hz, 2 H, 1-$CH_2$); *m/z* 200.141 ($M^{+\bullet}$; Calc. for $C_{11}H_{20}O_3$: *m/z* 200.141, 60), 183 (*M* − 15; 3), 182 (*M* − 18, 13; *m** 165.5, $182^2/200$ = 165.6), 167 (*M* − 18 − 15; 38), 155.106 (Calc. for $C_9H_{15}O_2$: *m/z* 155.107), 149 (167 − 18, 80; *m** 132.9, $149^2/167$ = 132.9), 116 (95), 115 (100), 102 (75), 98 (116 − 18, 65; *m** 82.8, $98^2/116$ = 82.8), 97 (115 − 18, 30; *m** 81.9, $97^2/115$ = 81.8), 85 (60), 84 (50), 83 (60), 69 (60), and 31 (40).

1-(4′,4′-*Ethylenedioxy*-1′-*hydroxy*-2′,2′,6′-*trimethylcyclohexyl*)*but*-1-*yne* (**12**).—But-1-yne (2.25 g) in ether (50 ml) was added to a suspension of lithamide (600 mg) in liquid ammonia (100 ml), and the mixture was stirred for 2 h. 4,4-Ethylenedioxy-2,2,6-trimethylcyclohexanone[7] (3.96 g) in ether (50 ml) was added dropwise over 30 min. The mixture was stirred at 20 °C overnight and ammonium chloride (2 g) was then added. Isolation of the product with ether, and crystallisation from ether–light petroleum, gave the acetylenic alcohol (4.5 g), m.p. 79 °C; $\nu_{max.}$(Nujol) 3 500 and 2 280 $cm^{-1}$; δ 1.03 (d, *J* 6.5 Hz, 3 H, 6′-Me), 1.09 (s, 6 H, 2′-$Me_2$), 1.14 (t, *J* 7.5 Hz, 3 H, $CH_2CH_3$), 1.40—2.00 (m, 6 H, reduced on deuteriation), 2.23 (q, *J* 7.5 Hz, 2 H, $CH_2CH_3$) and 3.88 (m, 4 H, $OCH_2CH_2O$); *m/z* 252 ($M^{+\bullet}$; Calc. for $C_{15}H_{24}O_3$: *m/z* 252, 0.5), 237 (*M* − 15, 0.5), 234 (*M* − 18, 0.5), 198 (0.4), 129 (98), 127 (29), and 113 (100).

1-(4′,4′-*Ethylenedioxy*-1′-*hydroxy*-2′,2′,6′-*trimethylcyclohexyl*)*butan*-2-*one* (**14**).—A solution of the preceding acetylenic alcohol (1.134 g) in tetrahydrofuran (10 ml) was cooled to 0 °C. A solution (0.136M; 20 ml) of diborane in the same solvent was added slowly. After 12 h more of the reagent solution (20 ml) was added and the mixture kept for 12 h at 20 °C; t.l.c. then showed the absence of starting material. Ethylene glycol was added slowly to decompose the excess of hydride. The organoborane was then oxidised by adding hydrogen peroxide (15%; 3.4 ml) whilst the pH was maintained at 8. After the mixture had been stirred for 1 h, acid resin (IR-120H) was added, and the mixture was filtered. The filtrate was saturated with sodium chloride and the product was isolated with ether. Preparative t.l.c. on Kieselgel Hf (ethyl acetate–hexane, 1:2), elution with chloroform, and crystallisation from ether–hexane, gave the required ketone (300 mg), m.p. 71 °C; $\nu_{max.}$($CHCl_3$) 3 430 and 1 695 $cm^{-1}$; δ 0.85 (s, 3 H, 2′-Me), 0.91 (d, *J* 7 Hz, 3 H, 6′-Me), 1.07 (t, *J* 7 Hz, 3 H $CH_2CH_3$), 1.13 (s, 3 H, 2′-Me), 1.32 (m, 1 H, 6′-CH), 1.60 (s, 4 H, 3′-$CH_2$ and 5′-$CH_2$), 2.54 (q, *J* 7 Hz, 2 H, $CH_2CH_3$), 2.60 (s, 2 H, $CH_2CO$), 3.92 (m, 4 H, $OCH_2CH_2O$), and 5.46 (s, 1 H, disappears on deuteriation, OH); *m/z* 270 ($M^{+\bullet}$; Calc. for $C_{15}H_{26}O_4$: *m/z* 270, 0.5), 252 (*M* − 18, 0.5), 241 (*M* − Et, 0.5), 226 (0.5), 213 (0.5), 191 (8), 129 (22), 127 (33), 113 (100), and 86 (17).

Attempts to condense the diketone with polyene dialdehydes were unsuccessful. Under the alkaline conditions used the β-hydroxy ketone grouping underwent retro-aldol fission, and the

products from reaction with the polyene dialdehyde were those expected from butan-2-one.

1-(1′-*Hydroxy*-2′,2′,6′-*trimethyl*-4′-*oxocyclohexyl*)*butan*-2-*one* (**16**).—Sulphuric acid (0.5M 4 ml) was added to the preceding ketone (100 mg) in acetone (4 ml) at 0 °C. The solution was allowed to warm to 20 °C and kept for 3 h. The acid was neutralised with saturated aqueous sodium hydrogen carbonate, the mixture was saturated with sodium chloride, and the product was isolated with ether. Preparative t.l.c. on Kieselgel H (ethyl acetate–hexane, 1:2), and crystallisation from ether–light petroleum, gave the diketone (82 mg), m.p. 49 °C; $\nu_{max.}$($CHCl_3$) 3440 and 1700 $cm^{-1}$; δ 0.98 (s, 3 H, 2′-Me), 0.99 (d, *J* 6.5 Hz, 3 H, 6′-Me), 1.05 (s, 3 H, 2′-Me), 1.12 (t, *J* 7.5 Hz, 3 H, $CH_2CH_3$), 1.95 (br, 1 H, disappears on deuteriation, OH), 2.00–2.50 (m, 5 H), 2.63 (q, *J* 7.5 Hz, 2 H, $CH_2CH_3$), and 2.83 (s, 2 H, 1-$CH_2$); $^{13}C$ n.m.r. see Table 2; *m/z* 226 ($M^{+\bullet}$; Calc. for $C_{13}H_{22}O_3$: *m/z* 226, 32), 211 (*M* − 15, 1.5), 208 (*M* − 18, 8; *m** 191.5, $208^2/216$ = 191.4), 197 (*M* − 29, 2), 193 (*M* − 18 − 15, 5), 169 (*M* − 57, 1), 155.107 (Calc. for $C_9H_{15}O_2$ 155.107, 12), 154 (11.5), 142 (40), 141 (70), 139 (154 − 15, 45; *m** 125.5, $139^2/154$ = 125.46), 128 (40), 113 (14), 98 (27), 97 (15), 85 (24), 83 (22), 72 (14), 70 (30), 69 (26), 57 (90), 43 (85), and 41 (100).

1-(1′,4′-*Dihydroxy*-2′,2′,6′-*trimethylcyclohexyl*)*butan*-2-*ol*.—Sodium borohydride (11 mg) was added to a solution of the preceding diketone (68 mg) in methanol (5 ml), and the mixture was stirred at 20 °C for 3 h. Water (2 ml) was added to destroy the excess of hydride, and the solution was saturated with sodium chloride. Isolation with ether, and chromatography on a column of silica gel (ethyl acetate–hexane) gave two isomers of the required triol.

(i) Less strongly adsorbed, a viscous oil (24 mg); $\nu_{max.}$($CHCl_3$) 3 595 and 3 500 $cm^{-1}$; δ 0.95 (s, 3 H, 2′-Me), 0.95 (t, *J* 6.5 Hz, 3 H, $CH_2CH_3$), 0.98 (d, *J* 6.5 Hz, 3 H, 6′-Me), 1.02 (s, 3 H, 2′-Me), 1.2–1.8 (br m), 1.33 (s, 3 H, disappears on deuteriation, 3 OH), and 6.06 (br m, 2 H, 2 *CH*OH); *m/z* 230 ($M^{+\bullet}$; Calc. for $C_{13}H_{26}O_3$: *m/z* 230, 0.5), 212 (*M* − 18, 10), 201 (1), 197 (0.5), 194 (*M* − 18 − 18, 0.5), 157.123 (Calc. for $C_9H_{17}O_2$: *m/z* 157.123, 0.5), and 43 (100).

(ii) More strongly adsorbed (12 mg), crystallised from ether–ethyl acetate and had m.p. 168 °C; $\nu_{max.}$(KBr) 3 485 and 3 400; *m/z* 230 ($M^{+\bullet}$, 0.5), 212 (*M* − 18, 100), 201 (4), 197 (*M* − 18 − 15, 4), 194 (*M* − 18 − 18, 2), and 157.123 (11).

trans-4,4-*Ethylenedioxy*-2,2,6-*trimethylcyclohexanol* (**17**).—Sodium borohydride (95 mg) in water (1 ml) was added to 4,4-ethylenedioxycyclohexanone [7] (490 mg) in methanol (5 ml), and the mixture was stirred for 24 h. Water was then added and the product was isolated with ether. Chromatography on a column of silica gel (5% acetone in light petroleum), and crystallisation from ether–light petroleum, gave the alcohol (320 mg), m.p. 98 °C; $\nu_{max.}$(Nujol) 3 440, 2 900, 1 460, 760, and 720 $cm^{-1}$; δ 1.02 (d, *J* 6 Hz, 3 H, 6-Me), 1.03 (s, 6 H, 2-$Me_2$), 1.3—2.0 (m, 5 H, 3-$CH_2$, 5-$CH_2$, and 6-CH), 1.55 (br s, disappears on deuteriation, 1 H, OH), 2.95 (d, *J* 9.5 Hz, 1 H, 1-axial H), and 3.92 (m, 4 H, $OCH_2CH_2O$); *m/z* 200 ($M^{+\bullet}$; Calc. for $C_{11}H_{20}O_3$: *m/z* 200, <0.5), 185 (*M* − 15, <0.5), 182 (*M* − 18, <0.5), 170 (*M* − 15 − 15, <0.5), 167 (*M* − 15 − 18, <0.5), 127 (35), and 113 (100).

trans-2,2,6-*Trimethyl*-4-*oxocyclohexanol* (**18**).—0.5M Sulphuric acid (5 ml) was added to a solution of the preceding alcohol (300 mg) in acetone (5 ml), and the mixture was stirred for 3 h. An excess of saturated aqueous sodium hydrogen carbonate was added, and the product was isolated with ether. Crystallisation from ether–light petroleum gave the keto alcohol (200 mg), m.p. 75 °C; $\nu_{max.}$(Nujol) 3 420, 2 900, 1 730, 1 460, and 760 $cm^{-1}$; δ 0.88 (s, 3 H, 2-Me), 1.12 (s, 3 H, 2-Me), 1.13 (d, *J* 6 Hz, 3 H, 6-Me), 1.56 (br m, 1 H, 6-axial H), 2.22 (m, 4 H, 3-$CH_2$ and 5-$CH_2$), 2.31 (br, disappears on deuteriation, 1 H, OH) and 3.34 (d, *J* 9.5 Hz, 1 H, 1-axial H); *m/z* 156 ($M^{+\bullet}$; Calc. for $C_9H_{16}O_2$: *m/z* 156; 55), 141 (*M* − 15, 60), 128 (*M* − 28, 10), 123 (*M* − 15 − 18, 10), 113 (*M* − 15 − 28, 15), and 41 (100).

cis-*Trimethyl*-4-*oxocyclohexanol* (**21**).—A solution of 4,4-ethylenedioxy-2,2,6-trimethylcyclohexanone [7] (500 mg) and aluminium isopropoxide (800 mg) in isopropyl alcohol (15 ml) was heated gently under reflux until the distillate gave a negative test for acetone (Brady's reagent). The mixture, containing the alcohol (**20**), was cooled and 2M hydrochloric acid (25 ml) was added. The mixture was shaken vigorously and then poured into ice–water. Isolation of the product with ether, and chromatography on a column of silica gel (acetone in light petroleum), gave the keto alcohol (200 mg) as an oil; $\nu_{max.}$(liq. film) 3 450, 2 960, 2 940, 2 870, 1 700, 1 460, and 760 $cm^{-1}$; δ 0.90 (s, 3 H, 2-Me), 1.08 (d, *J* 6 Hz, 6-Me), 1.11 (s, 3 H, 2-Me), 1.86 (dd, $J_1$ 14 Hz, $J_2$ 1.5 Hz, 1 H, 3-equatorial H), 1.2—2.4 (m, 3 H, 5-$CH_2$ and 6-CH), 2.63 (d, *J* 14 Hz, 1 H, 3-axial H), 2.70 (s, disappears on deuteriation, 1 H, OH), and 3.33 (br s, 1 H, 1-equatorial H); *m/z* 156 ($M^{+\bullet}$, 30), 141 (*M* − 15, 31), 128 (*M* − 28, 4), 123 (*M* − 15 − 18, 4), 113 (*M* − 15 − 28, 7), 85 (35), 83 (40), 58 (80), and 41 (100).

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## References

1 Part 41, preceding paper.
2 B. C. L. Weedon, *Pure Appl. Chem.*, 1973, **35**, 113.
3 A Khare, G. P. Moss, and B. C. L. Weedon, *Tetrahedron Lett.*, 1973, 3921.
4 A. J. Davies, A. Khare, A. K. Mallams, R. A. Massy-Westropp, G. P. Moss, and B. C. L. Weedon, *J. Chem. Soc., Perkin Trans. 1*, 1984, 2147.
5 G. P. Moss and B. C. L. Weedon in 'Chemistry and Biochemistry of Plant Pigments,' ed. T. W. Goodwin, Academic Press, London, 1976 2nd edn.
6 G. P. Moss., J. Szabolcs, Gy. Tóth, and B. C. L. Weedon, *Acta Chim. Acad. Sci. Hung.*, 1975, **87**, 301.
7 O. Isler, H. Lindlar, M. Montavon, R. Rüegg, G. Saucy, and P. Zeller, *Helv. Chim. Acta*, 1956, **39**, 2041.
8 R. Bonnett, A. K. Mallams, A. McCormick, A. A. Spark, J. L. Tee, and B. C. L. Weedon, *J. Chem. Soc. C*, 1969, 429.
9 K. Bernhard, G. P. Moss, Gy Tóth, and B. C. L. Weedon, *Tetrahedron Lett.*, 1976, 115.
10 B. C. L. Weedon, *Pure Appl. Chem*, 1970, **20**, 51.

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