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SYNTHESIS OF 5-[^{18}O]MEVALONOLACTONE

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Abstract—A preparation of *RS*-mevalonolactone containing over 90 at.% of ^{18}O attached to C-5 is described. The isotope was introduced by acid-catalysed hydrolysis of the dimethyl acetal of methyl mevaldate with $H_2{}^{18}O$, followed by reduction *in situ* with sodium borohydride.

5-[^{18}O]Mevalonate (from its lactone **4**) can be used experimentally to test the course of biosynthetic processes wherein the persistence of an O atom originally attached to C-5 of a mevalonate molecule can be questioned. Since dilution with endogenous material or partial loss of oxygen by exchange may arise as complications, it is useful to start with mevalonate having the highest economically attainable excess of isotope. A solution of this problem is presented.

Methyl 5,5-dimethoxy-3-hydroxy-3-methylpentanoate[1] (methyl mevaldate dimethyl acetal, **1**) in diglyme containing a little hydrogen chloride and a sixfold molar excess of water reached, after a few hours at room temperature, an equilibrium in which *ca.* 80% of the acetal had been hydrolysed to the aldehyde (**2**). Neutralization with dry ammonia followed by addition of sodium borohydride then converted the aldehyde into mevalonolactone (**4**) via methyl mevalonate (**3**), without affecting the residual acetal ester (**1**).

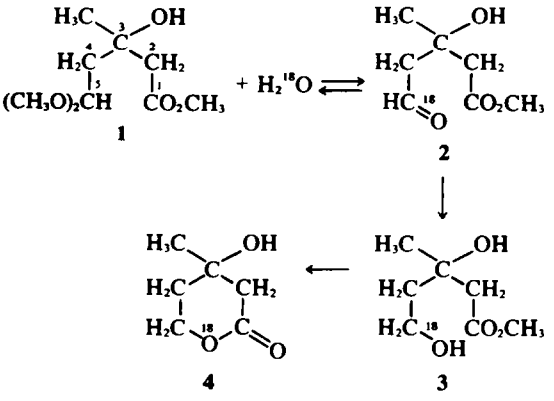
When this procedure was followed with $H_2{}^{18}O$, pure distilled mevalonolactone (**4**) was obtained in 70% yield, reckoned on unrecovered acetal ester, and 12% yield reckoned on $H_2{}^{18}O$. The molecular-ion region in mass spectra of this mevalonolactone and of the derived benzhydrylamide yielded the measurements reported in Table 1.

Table 1.

Mass spectra of mevalonolactone and its benzhydrylamide

Ion	Lactone *m/e* (%)	Benzhydrylamide *m/e* (%)
M^+	130 (3·8)	313 (3·7)
$M^+ + 2$	132 (74·6)	315 (74·0)
$M^+ + 4$	134 (21·6)	317 (22·3)

The numbers indicate labelling with ^{18}O at two positions, and analysis of the mass spectrum of mevalonolactone[2] locates these as the oxygens attached to C-1 and C-5. Assuming that the two oxygen positions were labelled by independent processes, calculation shows 95–96 at.% ^{18}O at one position and 23 at.% at the other. *Prima facie* the higher proportion of ^{18}O might be on the CO group; but the method of preparation excludes this since the C-5 oxygen is supplied from the labelled water alone and could not have a content of ^{18}O so much less than that of the water. The additional ^{18}O at the CO group is of no consequence for biosynthesis since the carboxyl group of mevalonate is lost at an early stage of terpenoid construction; if necessary this oxygen could be removed by alkali-catalysed equilibration of water and mevalonate anion, or acid-catalysed exchange between water and mevalonolactone. It is presumed to have been introduced by exchange between the C-1 CO group and $H_2{}^{18}O$ in the alkaline medium of the borohydride reduction.



EXPERIMENTAL

The $H_2{}^{18}O$ (97·3 at.% ^{18}O; 0·51 at.% ^{17}O), was supplied by Miles Laboratories Inc., Indiana, U.S.A. The mass spectra were run on an AEI MS-902 instrument at an ionizing potential of 70eV and an ion source temperature of 50–100°.

5-[^{18}O]*Mevalonolactone.* To a soln of methyl 5,5-dimethoxy-3-hydroxy-3-methylpentanoate (1·012 g) in dry diethylene glycol dimethyl ether (diglyme; 4 ml) under N_2 was added with stirring $H_2{}^{18}O$ (0·55 g), followed by a solution of dry HCl in dry diglyme (1 ml of 0·52 N). The mixture was stirred at ambient temp. for 6·5 hr, when a sample (50 μl) added to dry ether (0·25 ml) containing a little dry K_2CO_3 and analysed by GLC (3% SE-30; 120°) showed the equilibrium molar ratio (determined in earlier experiments with ordinary water) of 1 acetal (**1**): 4 aldehyde (**2**). Dry ammonia gas 20 ml; dried over barium oxide) was then passed through the mixture which was flushed with N_2, treated with more ammonia gas (10 ml) and purged with N_2 until a sample was neutral (pH around 7) to indicator paper. Sodium borohydride (209 mg in 5·5 ml dry diglyme) was added and the mixture was stirred for 25 min. Acetone (5 ml) was then added to destroy excess of borohydride and stirring was continued for 15 min. Water (10 ml) was added and the mixture was extracted thrice with ether (2 × 10 ml; 1 × 5 ml). The ethereal extracts on evaporation left a residue (235 mg) which by GLC appeared to be mainly the acetal (**1**) and smaller amounts of several impurities. To the aqueous layer was added enough 2 N NaOH to raise the pH to 13.

The mixture was stirred for 50 min, brought to pH 8 by addition of 4 N H_2SO_4, and extracted continuously overnight with chloroform (freshly distilled). This chloroform extract contained no mevalonolactone. The aqueous layer was then acidified to pH 2 with 4 N H_2SO_4 and extracted continuously with chloroform for 24 hr. The chloroform extract was dried ($MgSO_4$), evaporated and the residue evaporated three times with MeOH (10 ml portions) to remove traces of boric acid. The pale yellow oil (368 mg) was nearly pure mevalonolactone (TLC; IR; GLC; mass spectrum). A sample (10·8 mg) was reserved for conversion into the benzhydrylamide (m.p. 100–101·5° after two recrystallizations from benzene-light petroleum (b.p. 60–80°)) and the remainder was subjected to short-path distillation in high vacuum to yield colourless 5[^{18}O]mevalonolactone (331 mg), NMR spectrum identical with that of authentic mevalonolactone.

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REFERENCES

[1]G. Popják, DeW. S. Goodman, J. W. Cornforth, R. H. Cornforth and R. Ryhage, *J. Biol. Chem.* **236**, 1934 (1961).
[2]R. T. Gray, *Org. Mass Spectrometry* **9**, 346 (1974).