Synthesis of Linoleic Acid with Chiral Isotopic Labelling at a Flanking and a Medial Allylic Methylene: the (8*R*,9*Z*,12*Z*)-[8-^{2}H] and (11*R*,9*Z*,12*Z*)-[11-^{2}H]-Stereoisomers, and (*Z*)-[2,2-$^2H_2$]Non-3-enal

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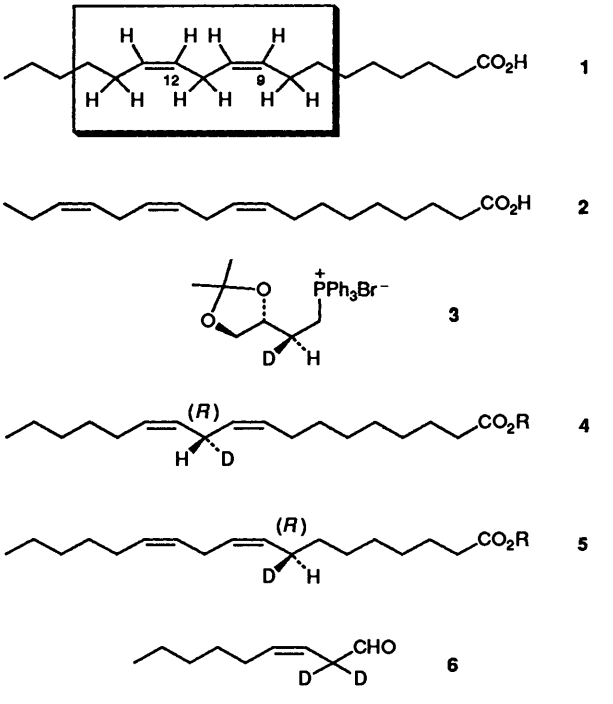
[1-^{2}H]Oct-2-ynal is converted by fermenting bakers' yeast into (1*S*)-[1-^{2}H]oct-2-yn-1-ol with an enantiomeric purity of >96% as measured by Mosher's MTPA method. The alcohol, as its tosyl ester, was then converted by copper-catalysed coupling with the di-Grignard of dec-9-ynoic acid and catalytic semi-hydrogenation, into (11*R*)-[11-^{2}H]linoleic acid having less than 2% *E*-material and >95 atom %D. Provision for (11*S*)-[11-^{2}H]linoleic acid was made by configurational inversion of (1*R*)-[1-^{2}H]oct-2-yn-1-ol using Mitsunobu chemistry.

(8*R*)-[8-^{2}H]Linoleic acid is made by a similar approach, the labelled chiral centre being formed on (8*S*)-8-hydroxy-[8-^{2}H]octanoic acid (>96% ee). Reaction of the corresponding tosate with lithium acetylide–ethylenediamine complex gave, with configurational inversion, (8*R*)-[8-^{2}H]dec-9-ynoic acid, built into (8*R*)-[8-^{2}H]linoleic acid. A degradative circuit is applied to estimate the extent of configurational inversion in the displacement. Mitsunobu inversion of the (8*S*)-8-hydroxy compound provides access to (8*S*)-[8-^{2}H]linoleic acid.

(*Z*)-Non-3-enal, a labile aldehyde from the enzymic degradation of linoleic acid, is made in [2,2-$^2H_2$]-labelled form by a synthesis which uses, as a key step, the reaction between triheptynylborane and deuteriodiazoacetic ester.

Fatty acids labelled with deuterium or tritium are important instruments for study of their chemistry and biochemistry, and we have made much use of them in our earlier work in this area.[1] In the plant world, the two key acids having methylene-interrupted (or 'skipped') conjugation are the diene linoleic acid **1** and the triene linolenic acid **2**. In the former case, most of the important chemistry and biochemistry concerns the ten boxed hydrogen atoms: in the latter, there are fourteen such atoms. For the more simple linoleic acid there are two pairs of olefinic hydrogens and three pairs of allylic methylenes, the latter forming three prochiral centres. These may be designated forward flanking (C-8), rearward flanking (C-14) and medial (C-11) methylene centres. Work on the synthesis of such isotopically labelled centres in chiral form has been comparatively limited. Corey[2] has reported an elegant total synthesis of (7*R*)-[7-^{2}H]arachidonic acid involving >20 steps and using the valuable Wittig synthon **3**, but the approach used most frequently has been to form a stereospecifically placed label by tosylation of a resolved secondary alcohol, followed by tosate displacement, with inversion, using lithium aluminium deuteride (or the less accessible tritide). The labelled fragment is then assembled into a C_{18} stearic acid unit, followed by treatment with a desaturase enzyme to introduce the necessary double bonds regiospecifically and stereospecifically. The green alga *Chlorella vulgaris* has been used to convert stearate into linoleate,[3] while the fungus *Saprolegnia parasitica* has been used for the conversion of stearate into arachidonate—the latter process involving a two-carbon elongation.[4] Other organisms have also been used for desaturation, but the inherent disadvantage of the approach is substantial dilution of the label by endogenous fatty acid.

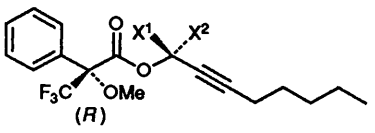
The major objective of the present work was the synthesis of (11*R*)-[11-^{2}H]linoleic acid **4**, R = H (Scheme 1) and (8*R*)-[8-^{2}H]linoleic acid **5**, R = H (Scheme 3) using well established yeast reduction at the critical stage. Both syntheses are adaptable for the (*S*)-enantiomers through configurational inversion. In addition, the synthesis of (*Z*)-[2,2-$^2H_2$]non-3-enal **6** (Scheme 6), a labelled form of the labile product obtained from the decomposition of linoleic acid (9*S*)-hydroperoxide by hydroperoxide lyase and required for associated studies, is reported.



Results and Discussion

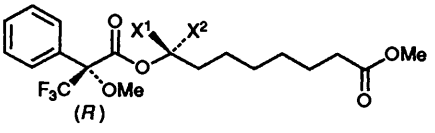
For the synthesis of (11*R*)-[11-^{2}H]linoleic acid **4**, our primary target was [1-^{2}H]-oct-2-ynal **9**, obtained by oxidation of [1,1-$^2H_2$]oct-2-yn-1-ol **8**, the latter being readily available in multigram quantities and high isotopic purity (Scheme 1). Hept-1-yne was converted into ethyl oct-2-ynoate **7** in 84% yield by treatment of its Grignard reagent in tetrahydrofuran (THF)

Table 1 ^{1}H NMR data (400 MHz; $CDCl_3$) for esters of optically active deuterio alcohols with (*R*)-(+)-Mosher's acid



15A

	(*S*)-(X^1 = D,X^2 = H)	(*R*)-(X^1 = H,X^2 = D)
(1*R*/*S*)-**13**[a]	δ 4.81 (t, $J_{1,4}$ 2.2 Hz)[b]	δ 4.93 (t, $J_{1,4}$ 2.2 Hz)[b]
(1*S*)-**10**	4.81 (t, $J_{1,4}$ 2.2 Hz)[b]	
(1*R*)-**17**		4.93 (t, $J_{1,4}$ 2.2 Hz)[b]

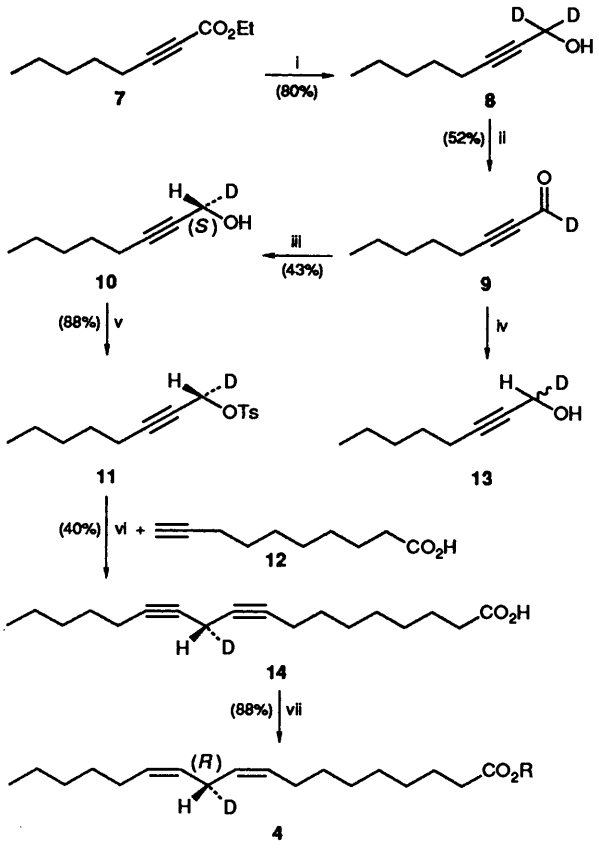


15B

(1*R*/*S*)	δ 4.31 (t, $J_{1,2}$ 6.6 Hz)	δ 4.27 (t, $J_{1,2}$ 6.6 Hz)
(1*S*)-**21**	4.31 (t, $J_{1,2}$ 6.6 Hz)	
(1*R*)-**27**		4.27 (t, $J_{1,2}$ 6.6 Hz)

[a] For **15A** (X^1 = X^2 = H) esterified with Mosher's acid [α-methoxy-α-(trifluoromethyl)phenylacetic acid] each 1-hydrogen (δ 4.82 and 4.92) resonated as a double triplet ($J_{1,2}$ 15 and $J_{1,4}$ 2.2 Hz). [b] Decoupling at the propargylic C-4 methylene (δ 2.20) collapsed the multiplet to a singlet.

with ethyl chloroformate. The latter was reduced with lithium aluminium deuteride (98 atom %D)[5] in THF at 0 °C to give [1,1-$^2H_2$]-[oct-2-yn-1-ol **8** (80%). With pyridinium chlorochromate (PCC) or pyridinium dichromate (PDC) as oxidant, yields of [1-^{2}H]oct-2-ynal **9** were poor, but activated manganese(IV)oxide[6] gave the aldehyde in 53% yield. Mass spectrometry revealed that the isotopic purity of the aldehyde (measured as the semicarbazone) was >98 atom %D.



Scheme 1 Synthesis of (11*R*)-[11-^{2}H]linoleic acid. *Reagents and conditions:* i, $LiAlD_4$, Et_2O, 5 °C; ii, $Mn^{IV}O_2$, CH_2Cl_2; iii, bakers' yeast, glucose; iv, $NaBH_4$, EtOH; v, TsCl, KOH, Et_2O, −50 to 0 °C; vi, EtMgBr, THF, CuBr·DMS; vii, H_2/Lindlar catalyst, quinoline, hexane.

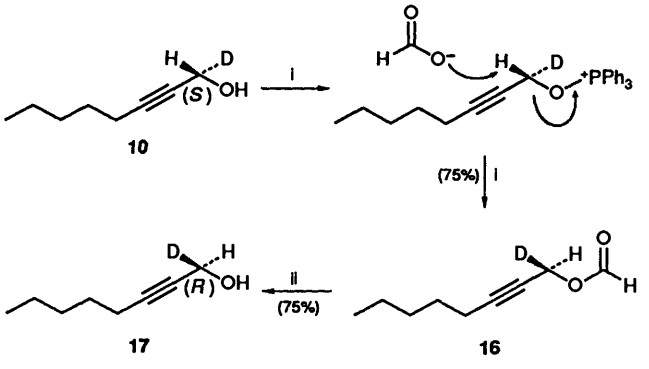
[1-^{2}H]Oct-2-ynal **9** was subjected to reduction by fermenting bakers' yeast and gave the required (1*S*)-[1-^{2}H]oct-2-yn-1-ol **10** in 43% yield. There is abundant precedent for the stereospecificity of the enzyme system to produce the (*S*)- configuration by hydrogen transfer to the carbonyl *si*-face in the reduction of deuterio aldehydes ranging from butanal to adamantanecarbaldehyde.[7] The enantiomeric purity of our alcohol was examined by the Mosher α-methoxy-α-(trifluoromethyl)phenylacetic ester (MTPA) technique.[8] The (*R*)-(+)-MTPA ester (see **15A**) of the chiral deuterio alcohol (1*S*)-**10** was compared with the MTPA esters of unlabelled oct-2-yn-1-ol and (1*R*/*S*)-[1^{2}H]oct-2-yn-1-ol, the latter alcohol **13** being obtained by reducing the deuterio aldehyde **9** with sodium borohydride. Table 1 gives the α-^{1}H NMR resonances (400 MHz) for the specimens. The spectrum of the Mosher's ester of the acetylenic alcohol having no deuterium substitution shows magnetic non-equivalence of the diastereotopic geminal protons by a twelve-line multiplet corresponding to J_{AB} 15 Hz with long-range coupling ^5J 2.2 Hz. Entry (1-*R*/*S*)-**13** shows the signals of the two diastereoisomers derived from the (±)-deuterio alcohol **13** as two cleanly separated singlets when decoupled at C-4. The two further entries for **15A** in Table 1 show diastereoisomers corresponding to (1*S*)-[1-^{2}H]oct-2-yn-1-ol **10** and (1*R*)-[1-^{2}H]oct-2-yn-1-ol **17** (see later) respectively: the signals in these spectra are also decoupled at C-4 and show singlets broadened by coupling to geminal deuterium. Since the absolute configuration of the α-deuteriated ester from (1*S*)-**10** is known from the stereospecificity of the enzyme, the prochirality of the geminal methylene protons in (1*R*/*S*)-**13** can be established with the *pro*-(*R*)-hydrogen resonating at higher field than the *pro*-(*S*). From the spectrum of compound (1*S*)-**10** we can assign an enantiomeric excess of >96%.

Incorporation of the -CHD-chiral centre into a C_{18} linoleic acid framework was carried out by conversion of the alcohol **10** into its tosyl derivative **11**[9] and coupling of the latter with the di-Grignard reagent from dec-9-ynoic acid **12**[10] in the presence of a copper(I) bromide–dimethyl sulfide complex as catalyst.[9,11] (11*R*)-[11-^{2}H]Octadeca-9,12-diynoic acid **14** was formed in 40% yield and the crystalline, though unstable, compound was immediately semi-hydrogenated over Lindlar catalyst in hexane to give (11*R*,9*Z*,12*Z*)-[11-^{2}H]linoleic acid **4**, R = H. For purification and characterisation purposes the acid was converted into its methyl ester **4**, R = Me using diazomethane. Analysis by ^{13}C NMR spectroscopy showed that <2% of *E*-isomer was present, though when ethanol was used as the hydrogenation solvent 5–10% of this unwanted isomer was formed.

Mass spectrometry shows that the methyl (11*R*,9*Z*,12*Z*)-[11-^{2}H]linoleate contains >95 atom %D, which is consistent with the isotopic purity of the intermediates and their spectroscopic data. On comparing the broad-band decoupled ^{13}C NMR spectrum of ester **4**, R = Me with that of the unlabelled material, it was noted that the C-11 singlet signal had collapsed to a low-intensity 1:1:1 triplet which analysed as a methine carbon in the DEPT experiments. A clean signal was

seen in the ^2H NMR spectrum (δ_D 2.78) and in the ^1H spectrum the C-11 proton signal integrated for one proton. In the mass spectrum, accurate mass measurement showed that one deuterium was present in the molecular ion of structure **4**, R = Me.

In order to provide for the synthesis of (11*S*,9*Z*,12*Z*)-[11-^2H]linoleic acid, the enantiomer of ester **4**, R = H, we have applied the Mitsunobu inversion[12] to (1*S*)-[1-^2H]oct-2-yn-1-ol. This reaction proceeds with inversion of configuration and we employed the easily hydrolysed[13] formate ester. Treatment of the deuteriated alcohol **10** (Scheme 2) with diethyl



10 **17** **16**

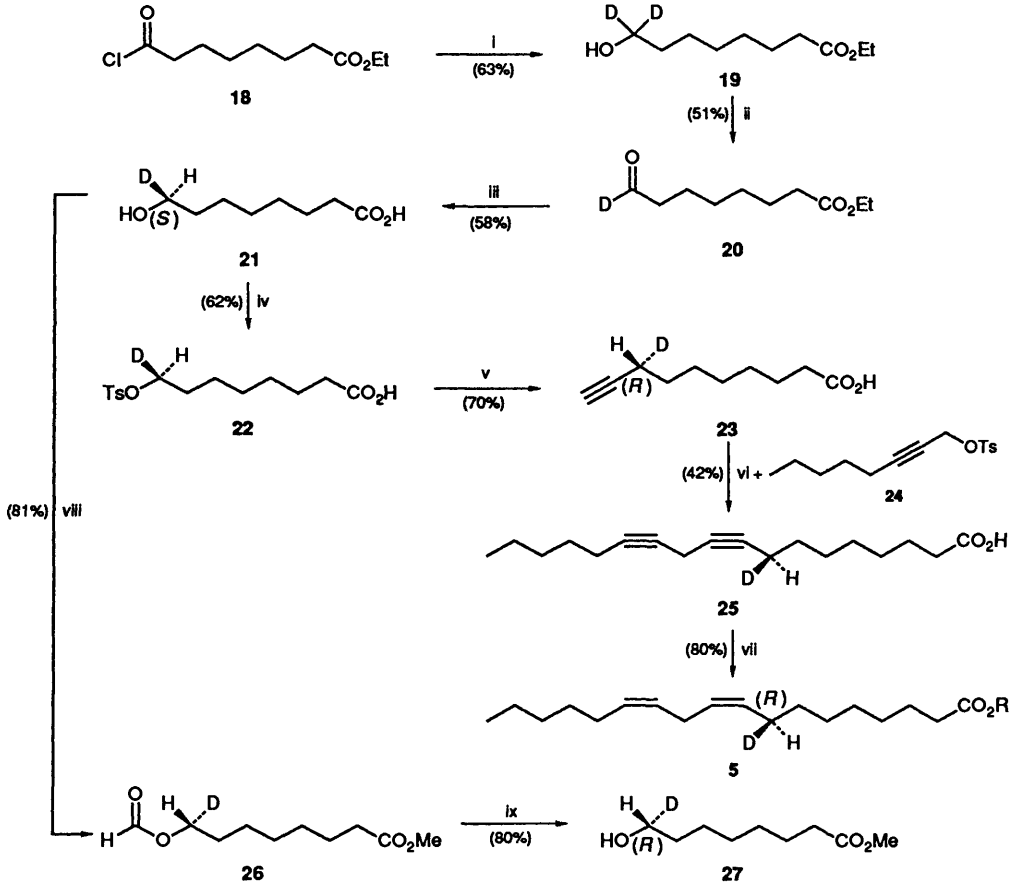
Scheme 2 (1*R*)-[1-^2H]Oct-2-yn-1-ol by Mitsunobu inversion. *Reagents:* i, DEAD, PPh_3, HCO_2H, THF; ii, MeOH, NH_4OH

azodicarboxylate (DEAD), triphenyl phosphine, and formic acid in THF gave the configurationally inverted ester **16** *via* displacement. The formate ester was hydrolysed using methanolic ammonia, giving (1*R*)-[1-^2H]oct-2-yn-1-ol **17** (see Table 1) in 75% yield. Examination of the enantiomeric purity by the MPTA technique showed that the (1*R*)-isomer had been formed in >96% ee.

The synthetic approach to (8*R*)-[8-^2H]linoleic acid **5**, R = H was similar to that used for the 11-isomer and is summarised in Scheme 3. Ethyl hydrogen suberate is readily prepared in quantity by co-pyrolysis of diethyl suberate with suberic acid, and treatment with thionyl dichloride converted it into the acid chloride **18**.[14] Incorporation of deuterium was achieved by reduction of the half-acid-ester chloride with sodium borodeuteride (98 atom %D) to give ethyl 8-hydroxy-[8,8-$^2H_2$]octanoate **19**. Oxidation to ethyl 7-([^2H]formyl)-heptanoate **20** could be effected with the Swern reagent[15] but best results were obtained with PDC.[16] Nonetheless the product required repeated distillation to attain sufficient purity for the reduction by yeast: the isolated yield (51%) is thus lower than the chemical yield.

On reduction with fermenting bakers' yeast, the deuterio aldehyde gave (8*S*)-8-hydroxy-[8-^2H]octanoic acid **21** as a crystalline solid in 58% yield, lipases in the fermenting mixture having hydrolysed the ester group. The enantiomeric purity of the acid, examined as its methyl ester, was compared with the racemic (*R*/*S*)-alcohol using Mosher's MTPA technique (see ester **15B**, Table 1) and was found to be >96%. The (*S*)-configuration was assigned on the basis of previous work (above).

The approach to the formation of (8*R*)-[8-^2H]dec-9-ynoic acid **23**, the next intermediate in the sequence, required formation of the tosate **22** by a standard procedure, and displacement with configurational inversion by acetylide ion.[10]

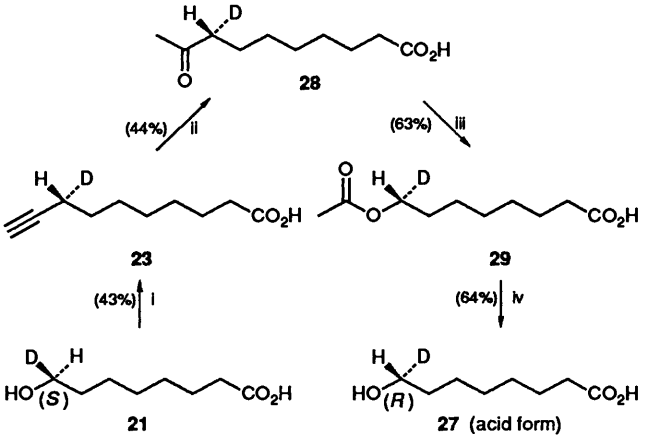


18 **19** **20** **21** **22** **23** **25** **5** **26** **27**

Scheme 3 Synthesis of (8*R*)-[8-^2H]linoleic acid and Mitsunobu inversion for the enantiomeric intermediate. *Reagents and conditions:* i $NaBD_4$, DMF–THF, 0 °C; ii, PDC, CH_2Cl_2; iii, bakers' yeast, glucose; iv, TsCl, pyridine; v, lithium acetylide–EDA, DMSO; vi, EtMgBr, THF, CuBr·DMS; vii, H_2/Lindlar catalyst, quinoline, hexane; viii, (a) CH_2N_2; (b) DEAD, PPh_3, HCO_2H, THF; ix, MeOH, NH_4OH.

There is good precedent for expecting complete inversion in this process and treatment of the deuterio tosate **22** with lithium acetylide–ethylenediamine complex in dimethyl sulfoxide (DMSO) gave (8*R*)-[8-^{2}H]dec-9-ynoic acid **23** in 70% yield. Using the same copper-catalysed Grignard procedure as that described above, the unstable (8*R*)-[8-^{2}H]octadeca-9,12-diynoic acid **25** was prepared by using the tosate **24**, and the diynoic acid was converted into (8R,9*Z*,12*Z*)-[8-^{2}H]linoleic acid **5**, R = H by semi-hydrogenation over Lindlar catalyst in hexane. Examination by ^{13}C NMR and mass spectrometry showed <2% of *E*-isomer and >95 atom %D isotopic purity. Evidence for substitution at C-8 by a single deuterium atom was similar to that given above.

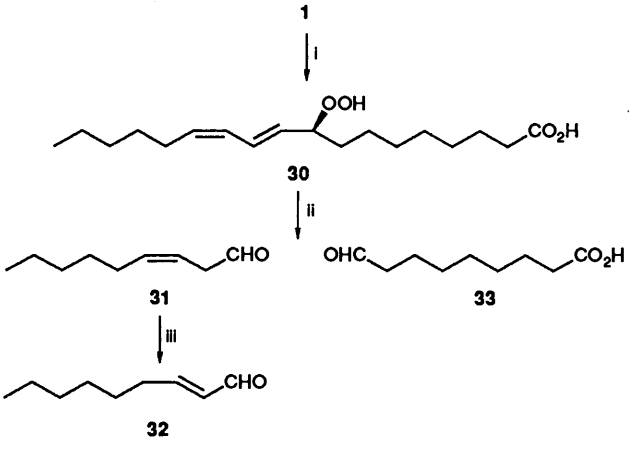
In order to establish the synthesis of (8*S*,9*Z*,12*Z*)-[8-^{2}H]linoleic acid, methyl (8*R*)-8-hydroxy-[8-^{2}H]octanoate **27** was prepared by Mitsunobu reaction [12] as shown in Scheme 3. As indicated, yields were good, and the enantiomeric purity was >96% as assessed by the MTPA method.



Scheme 4 Test circuit for configurational inversion by displacement using acetylide ion. *Reagents and conditions:* i, (a) TsCl, pyridine; (b) lithium acetylide–EDA, DMSO; ii, (a) mercury(II) acetate, aq. acetic acid; (b) $NaBH_4$; (c) PDC; iii, CF_3CO_3H, CH_2Cl_2, Na_2HPO_4, 0 °C; iv, EtOH, KOH.

During this work we have attempted to devise a reaction sequence which might permit us to estimate the extent of configurational inversion in the displacement of tosyloxy ion by acetylide ion (Scheme 4). Starting with (8*S*)-8-hydroxy-[8-^{2}H]octanoic acid **21**, the alcohol was tosylated and the acetylide inversion was effected. Attempts to hydrate the acetylene **23** by using the standard procedure [mercury(II) oxide and strongly acid conditions] caused epimerisation of the centre adjacent to the carbonyl. Under a variety of conditions, this epimerisation was confirmed by conducting experiments in deuteriated media. The most suitable technique, which involved little or no epimerisation, was to use stoichiometric addition of mercury(II) acetate under weakly acid conditions,[17] followed by reduction of the carbon–mercury bond with sodium borohydride and oxidation of the secondary alcohol to the methyl ketone **28** by using PDC in 44% yield. Baeyer–Villiger reaction [18] was achieved by using peroxytrifluoroacetic acid in buffered dichloromethane,[19] giving the acetate **29** in good yield. Hydrolysis with ethanolic potassium hydroxide then furnished (8*R*)-8-hydroxy-[8-^{2}H]octanoic acid. This was converted into the methyl ester (CH_2N_2) and the enantiomeric purity was assessed by the MTPA method. The latter showed that ester **27** was obtained in 85% enantiomeric excess, *i.e.* that it contained ~8% of the unwanted enantiomer. We are thus able to demonstrate that our sample of (8*R*)-[8-^{2}H]linoleic acid has at least this enantiomeric purity though in our opinion the actual purity is much closer to 100%. The epimerisation encountered probably lies in the deficiencies of the test system which uses an epimerisable ketone, rather than in lack of stereoselectivity in the acetylide displacement.

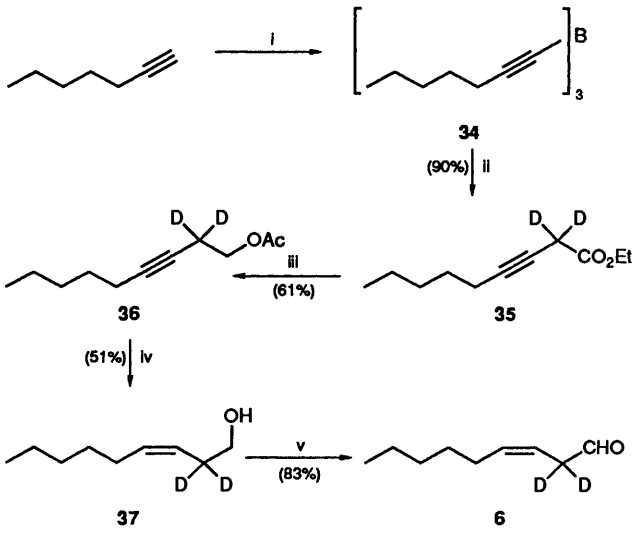
One of our interests [1] in the fatty acid field relates to the way in which smaller odoriferous molecules arise by oxidative degradation of linoleic or linolenic acid.[20] This can involve the formation by a (9*S*)-lipoxygenase (*e.g.*, from potato or tomato) of a hydroperoxide, *e.g.* **30**, and its decomposition by hydroperoxide lyase to an unsaturated aldehyde, *e.g.* **31**, and the aldehyde acid **33** (Scheme 5). The *Z*-3-enal system of compound **31** is labile and it is readily isomerised *in vitro* to the *E*-2-enal **32**, an isomerisation which in the plant is catalysed by an isomerase enzyme.[21] Further oxidation and reductions of the two aldehydes lead to other odoriferous compounds which also affect the organoleptic quality of fruit and vegetables. Study of these systems require not only isotopically labelled linoleic and linolenic acids, but also isotopically labelled aldehydes and for study of the enzymically induced *Z*-3-enal to *E*-2-enal change we initially required the labile (3*Z*)-[2,2-$^2H_2$]nonenal **6**.



Scheme 5 Formation of short-chain aldehydes from linoleic acid *via* the 9-hydroperoxide. *Reagents:* i, lipoxygenase, O_2; ii, hydroperoxide lyase; iii, cucumber isomerase.

Literature approaches to this type of aldehyde were unpromising, with little opportunity for isotopic incorporation, and with the unwanted 2*E*-material a frequent by-product. We have therefore adopted a new approach to the synthesis of (3*Z*)-[2,2-$^2H_2$]nonenal **6** (Scheme 6). The highly unstable triheptynylborane **34** was made by the addition of boron trifluoride–diethyl ether to three mole equivalents of lithio-1-heptyne [22] at −40 °C and this mixture was added to ethyl deuteriodiazoacetate (made by heterogeneous exchange of ethyl diazoacetate in D_2O–diethyl ether with potassium carbonate as catalyst),[23] followed by quenching with D_2O, which gave ethyl [2,2-$^2H_2$]non-3-ynoate **35** in excellent yield. By ^{1}H NMR analysis it contained >96 atom %D.

Reduction of the ester by careful treatment with lithium aluminium hydride gave the homopropargylic alcohol. The organoborane method [24] was used for reduction of the acetylene to a *Z*-olefin and this necessitated formation of the acetate **36** by using acetic anhydride in pyridine with 4-(dimethylamino)pyridine (DMAP) as catalyst. The acetylene was reduced with diisoamylborane (Sia_2BH) in diglyme, followed by acidic, then oxidative, work-up. Finally, after hydrolysis to give the alcohol **37**, selective oxidation to the *Z*-3-enal **6** was carried out by using the Dess–Martin periodinane reagent [25] (superior in this context to Collins-type reagent).[26] The yield was high and importantly there was no *E*-2-aldehyde by-product. Examination by GLC showed only a little unchanged starting material.



Scheme 6 Synthesis of (*Z*)-[2,2-$^2H_2$]Non-3-enal. *Reagents and conditions:* i, (a) BuLi, THF 0 °C; (b) BF_3•Et_2O, −40 °C; ii, N_2CDCO_2Et, −40 °C, D_2O; iii, (a) $LiAlH_4$, THF; (b) Ac_2O, pyridine, DMAP; iv, (a) Sia_2BH, diglyme; (b) AcOH; (c) H_2O_2, NaOAc; (d) $LiAlH_4$; v, Dess–Martin periodinane, CH_2Cl_2.

Experimental

^1H NMR spectra were recorded using a Bruker AM400, a Bruker WM250, a Bruker WP80SY, or a Perkin-Elmer R32 spectrometer for solutions in $CDCl_3$ unless stated otherwise. *J*-Values are in Hz. ^{13}C NMR spectra were measured ($CDCl_3$) on the Bruker AM400 spectrometer operating at 100 MHz or the WM250 operating at 63 MHz, with broad-band decoupling and assignment of secondary, tertiary and quaternary carbons by the DEPT pulse sequence. ^2H NMR spectra were recorded ($CHCl_3$) on the Bruker WM250 operating at 40 MHz. Analytical TLC plates were inspected under UV-light and, if necessary, spots were visualised by anisaldehyde or permanganate sprays. Column chromatography using silica gel refers to Merck 9385 flash silica. GLC was carried out on a Pye 104 machine using glass or stainless steel columns (5′ × ¼″) containing Carbowax 20M. Light petroleum refers to the fraction boiling in the range 60–80 °C.

Ethyl Oct-2-ynoate **7**.—Heptynylmagnesium bromide was prepared by addition of hept-1-yne (25 cm^3, 0.19 mol) to a cooled (−5 °C) Grignard reagent prepared from bromoethane (16.5 cm^3, 0.22 mol) and magnesium (4.86 g, 0.20 g-atom) in THF (20 cm^3) under nitrogen. The mixture was then heated under reflux for 1 h at 55 °C.

Heptynylmagnesium bromide was then added under nitrogen to a solution of ethyl chloroformate (19.1 cm^3, 0.20 mol) in THF (50 cm^3) at 0 °C during 2 h. After refluxing (1 h), the product was cooled to 0 °C and decomposed with saturated aq. ammonium chloride. The organic layer was separated and the aqueous layer was extracted with diethyl ether. The united organic extracts were washed, dried ($MgSO_4$), and evaporated. Distillation gave ethyl oct-2-ynoate **7** (26.8 g, 84%), b.p. 51–52 °C at 0.2 mmHg (Found: C, 71.3; H, 9.75. Calc. for $C_{10}H_{16}O_2$: C, 71.4; H, 9.6%); δ_H 0.90 (3 H, t, Me), 1.31 (3 H, t, *J* 7, OCH_2Me), 1.20–1.80 (6 H, m, 3 × CH_2), 2.37 (2 H, t, CH_2) and 4.26 (2 H, q, *J* 7, OCH_2Me); ν_{max}(neat)/cm^{-1} 2240 (C≡C) and 1650 (C=O).

[1,1-$^2H_2$]*Oct-2-yn-1-ol* **8**.—A solution of ethyl oct-2-ynoate **7** (7.62 g, 45 mmol) in dry diethyl ether (25 cm^3) was added slowly (1 h) to a stirred suspension of lithium aluminium deuteride (1 g, 24 mmol; 98 atom%) in diethyl ether (40 cm^3) at 5 °C under nitrogen, and the mixture was stirred for 2 h. Ethyl acetate (10 cm^3), followed by water (100 cm^3), was added and the pH was adjusted to pH 1 by 2 mol dm^{-3} hydrochloric acid. Work-up with diethyl ether, washing of the extracts with saturated brine and drying over $MgSO_4$, gave, after evaporation, an oil, which was chromatographed over silica gel [eluent: ethyl acetate–hexane (1:19, then 3:17)]. The product was then distilled to give the *alcohol* **8**, b.p. 40–42 °C at 0.1 mmHg (4.65 g, 80%) [Found: *m/z*, 110.106. $C_8H_{12}D_2O - H_2O$ requires *m/z*, 110.106]; δ_H(90 MHz) 0.90 (3 H, t, Me), 1.20–1.80 (6 H, m, 3 × CH_2), 2.23 (2 H, t, CH_2) and 2.48 (1 H, s, OH); ν_{max}(film)/cm^{-1} 3600–3100br (OH) and 2220 (C≡C).

[1-^2H]*Oct-2-ynal* **9**.—[1,1-$^2H_2$]Oct-2-yn-1-ol **8** (4.0 g, 31 mmol) and activated manganese(IV) oxide (30 g) were stirred together in dry dichloromethane (200 cm^3) for 48 h under nitrogen. The product was filtered through a short column of Florisil and the filtrate was evaporated and chromatographed on silica gel and eluted with ethyl acetate–hexane (1:19). It was then distilled to give the aldehyde **9** (2.01 g, 53%), b.p. 21–23 °C/0.1 mmHg; δ_H(90 MHz) 0.90 (3 H, t, Me), 1.20–1.80 (6 H, m, 3 × CH_2) and 2.44 (2 H, t, CH_2); ν_{max}(film)/cm^{-1} 2320, 2220 (C≡C), 2130 (C–D) and 1650 (C=O). The *semicarbazone* had m.p. 92–94 °C (Found: M^+, 182.127. $C_9H_{14}DN_3O$ requires *M*, 182.128).

(1S)-[1-^2H]*Oct-2-yn-1-ol* **10** *by Yeast Reduction of* [1-^2H]-*Oct-2-ynal* **9**.—A solution of deuterio aldehyde **9** (2.3 g, 18.4 mmol) in ethanol (1 cm^3) was added during 2 h to a rapidly fermenting mixture of bakers' yeast, *Saccharomyces cerevisiae* [35 g, Sigma YSC type(II)] and glucose (45 g) in tap water (800 cm^3) at 30 °C. The mixture was gently stirred for 16 h, at which time fermentation had ceased. The reaction mixture was filtered through a Florisil pad and the filtrate was continuously extracted with diethyl ether (300 cm^3). The extract was washed (saturated brine), dried ($MgSO_4$), chromatographed on silica gel with ethyl acetate–hexane (1:9) as eluent, and distilled from *p*-hydroquinone (20 mg) to give (1*S*)-[1-^2H]oct-2-yn-1-ol **10** (930 mg, 43%), b.p. 40–42 °C/0.1 mmHg; δ_H(80 MHz) 0.90 (3 H, t, Me), 1.10–1.70 (6 H, m, 3 × CH_2), 1.91 (1 H, s, OH), 2.22 (2 H, m, CH_2) and 4.24 (1 H, br s, C*H*DOH); δ_D(40 MHz) 4.24 (1 D, s, CHDOH); δ_C(100 MHz) 13.97 (C-8), 18.74 (C-4), 22.24 (C-7), 28.37 (C-5), 31.10 (C-6), 51.01 (C-1, t), 78.36 (C-3) and 86.49 (C-2); ν_{max}(film)/cm^{-1} 3600–3100br (OH), 2300, 2240 (C≡C), 2190 and 2140 (C–D). The Mosher ester derivative had δ_H(400 MHz) 0.89 (3 H, t, *J* 7.0, Me), 1.28–1.38 (4 H, m, 2 × CH_2), 1.49 (2 H, m, CH_2), 2.20 (2 H, m, C≡CCH_2), 3.58 (3 H, t, *J* 1.2, OMe), 4.81 (1 H, t, *J* 2.2, CHD), 7.40 3 H, m, 3 × ArH) and 7.54 (2 H, m, 2 × ArH). See also Table 1.

(1S)-1-(p-*Tolylsulfonyloxy*)-[1-^2H]*oct-2-yne* **11**.—Powdered potassium hydroxide (750 mg) was added to a solution of (1*S*)-[1-^2H]oct-2-yn-1-ol **10** (200 mg, 1.57 mmol) and toluene-*p*-sulfonyl chloride (360 mg, 1.90 mmol) in anhydrous diethyl ether (5 cm^3) at −50 °C under nitrogen and the stirred mixture was kept at 0 °C for 2 h. The product was poured into water and worked up by extraction with diethyl ether in the usual way to give the *title compound* **11** as an oil (390 mg, 88%) (Found: *m/z*, 126.104. $C_{15}H_{19}DO_3S - C_7H_7O_2$ requires *m/z*, 126.103); δ_H(80 MHz) 0.88 (3 H, t, Me), 1.10–1.60 (6 H, m, 3 × CH_2), 2.07 (2 H, m, CH_2), 2.44 (3 H, s, Ar*Me*), 4.69 (1 H, t, *J* 2.1, C*H*DOTs), 7.33 (2 H, d, *J* 8.5, 2 × ArH) and 7.81 (2 H, d, *J* 8.5, 2 × ArH); ν_{max}(film)/cm^{-1} 2310, 2240 (C≡C) and 1600 (Ar).

(11R)-[11-^2H]*Octadeca-9,12-diynoic Acid* **14** *by Copper-catalysed Grignard Coupling*.—A solution of dec-9-ynoic acid **12** (132 mg, 0.78 mmol) in anhydrous THF (1 cm^3) was added slowly to a solution of ethylmagnesium bromide (2.0 mol dm^{-3} in THF, 0.8 cm^3) at 0 °C and under nitrogen. Copper(I)

bromide–dimethyl sulfide complex (20 mg) was added and the mixture was stirred for 20 min. (1*S*)-1-(*p*-Tolylsulfonyloxy)-[1-^{2}H]oct-2-yne **11** was then added (during 20 min) and the mixture was stirred at 5 °C (30 min) and then at room temperature (4 h). Saturated aq. ammonium chloride (25 cm^3) was added and the product was worked up by extraction (Et_2O) to give a green oil, which was purified by chromatography on silica gel [eluent ethyl acetate–light petroleum (1:9) with 0.1% acetic acid] to give (11*R*)-[11-^{2}H]octadeca-9,12-diynoic acid **14** (81 mg, 40%) as a crystalline solid, used immediately in the next stage; δ_H(80 MHz) 0.90 (3 H, t, Me), 1.20–1.80 (16 H, m, 8 × CH_2), 2.16 (4 H, m, 2 × CH_2), 2.35 (2 H, t, CH_2CO_2H), 3.08 (1 H, t, CDH) and 9.59 (1 H, br s, CO_2H), ν_{max}($CHCl_3$)/cm^{-1} 3600–2500br (OH), 2150 (C–D) and 1705 (C=O).

Methyl (11R)-[11-^{2}H]*Octadeca*-9(Z),12(Z)-*dienoate* **4**, *R* = *Me*.—A stirred mixture of the diynoic acid **14** and lead-poisoned 5% palladium on calcium carbonate catalyst (Lindlar, 20 mg) in hexane (5 cm^3) containing 2% quinoline was treated with hydrogen, with monitoring by TLC. Reaction was judged complete in 6 h and the catalyst was filtered off. The filtrate was washed successively with 2 mol dm^{-3} hydrochloric acid and brine, dried ($MgSO_4$) and evaporated under reduced pressure to give (11*R*)-[11-^{2}H]octadeca-9(*Z*),12(*Z*)-dienoic acid **4**, R = H (65 mg, 86%).

The latter (65 mg) was treated with ethereal diazomethane and the methyl ester was purified by chromatography on silica gel [eluent diethyl ether–light petroleum (1:99)] to give *methyl* (11R)-[11-^{2}H]*octadeca*-9(Z),12(Z)-*dienoate* **4**, R = Me (55 mg, 81%) (Found: M^+, 295.262. $C_{19}H_{33}DO_2$ requires *M*, 295.263); δ_H(400 MHz) 0.89 (3 H, t, Me), 1.30 (14 H, m, 7 × CH_2), 1.61 (2 H, m, 3-H_2), 2.04 (4 H, m, 8; 14-H_2), 2.30 (2 H, t, *J* 7.4, CH_2CO_2Me), 2.75 (1 H, s, 11-H), 3.66 (3 H, s, OMe), 5.35 (4 H, m, 9-, 10-, 12- and 13-H); δ_D(40 MHz) 2.78 (1 D, s, 11-D); δ_C(100 MHz) 13.87 (C-18), 22.39 (C-17), 24.76 (C-3), 25.14 (C-11, t), 27.01 (C-8 and C-14), 28.93, 28.96, 29.07, 29.17, 29.26 and 29.40 (CH_2s), 31.34 (C-16), 33.92 (C-2), 51.24 (OMe), 127.68 (C-12), 127.81 (C-10), 129.87 (C-9) 130.04 (C-13) and 174.1 (C-1); ν_{max}(film)/cm^{-1} 3020 (olefinic C–H), 2350, 2150 (C–D), 1735 (ester C=O) and 1650 (C=C).

(1R)-[1-^{2}H]*Oct*-2-*yn*-1-*ol* **17**.—A mixture of (1*S*)-[1-^{2}H]-oct-2-yn-1-ol **10** (50 mg, 0.39 mmol), triphenylphosphine (153 mg, 0.58 mmol) and anhydrous formic acid (3 drops) in anhydrous THF (3 cm^3) was treated at room temperature under nitrogen with a solution of DEAD (101 mg, 0.58 mmol) in THF (0.5 cm^3). The reaction mixture was stirred (4 h), evaporated, and extracted with hexane. The combined extracts were evaporated to give an oil, which was purified by chromatography on silica gel, with ethyl acetate–light petroleum (1:19) as eluent, to give (1*R*)-[1-^{2}H]oct-2-ynyl formate **16** (45 mg, 75%), δ_H(80 MHz) 0.90 (3 H, t, Me), 1.20–1.70 (6 H, m, 3 × CH_2), 2.20 (2 H, m, CH_2), 4.74 (1 H, m, CHD) and 8.05 (1 H, d, *J* 0.8, OCOH); ν_{max}(film)/cm^{-1} 2320 (C–D), 2220 (C≡C) and 1725 (ester C=O).

Aq. ammonia (36%; 2 drops) was added to a solution of the formate **16** (20 mg) in methanol (2 cm^3) and the mixture was stirred overnight. The product was concentrated under reduced pressure and dissolved in diethyl ether–hexane (10 cm^3; 1:1). The organic layer was washed successively with water and saturated brine, dried ($MgSO_4$) and evaporated to give (1*R*)-[1-^{2}H]oct-2-yn-1-ol **17** (12 mg, 75%); δ_H(80 MHz) 0.90 (3 H, t, Me), 1.10–1.70 (6 H, m, 3 × CH_2), 2.24 (2 H, m, CH_2) and 4.23 (1 H, br s, C*H*DOH); ν_{max}(film)/cm^{-1} 3600–3100br (OH), 2300, 2240 (C≡C) and 2140 (C–D). See also Table 1.

7-(*Chloroformyl*)*heptanoate* **18**.—Suberic acid (37.6 g, 0.216 mol) and diethyl suberate (49.3 g, 0.214 mol) were heated together at 230 °C for 8 h. After cooling, the solidified product was extracted thoroughly with diethyl ether and the combined extracts were evaporated, and extracted with aq. sodium carbonate. After being washed with diethyl ether the aqueous extracts were acidified to pH 1 and extracted with diethyl ether. Evaporation of the ethereal extracts after they had been washed (water) and dried ($MgSO_4$) gave, on distillation, ethyl hydrogen suberate (18.3 g, 21%), b.p. 110–114 °C/0.15 mmHg (lit.,[14] 90–95 °C/0.1 mmHg) as an oil which solidified at 0 °C; ν_{max}(film)/cm^{-1} 1740 (C=O) and 1710 (C=O).

Ethyl hydrogen suberate (10 g, 50 mmol) was refluxed with thionyl dichloride (6.1 g, 84 mmol) overnight and the product was distilled to give the title compound (10.3 g, 95%), b.p. 78–80 °C/0.1 mmHg (lit.,[14] 75–80 °C/0.1 mmHg); δ_H(90 MHz) 1.23 (3 H, t, OCH_2Me), 1.10–1.80 (8 H, br m, 4 × CH_2), 2.32 (2 H, t, CH_2CO_2Et), 2.88 (2 H, t, CH_2COCl) and 4.14 (2 H, q, OCH_2Me); ν_{max}(film)/cm^{-1} 1800 (COCl), 1735 (ester C=O) and 730 (C–Cl).

Ethyl 8-*Hydroxy*-[8,8-$^2H_2$]*octanoate* **19**.—A solution of the acid **18** (9.89 g, 45.2 mmol) in anhydrous THF–dimethylformamide (DMF) (30 cm^3; 1:1) was added to a solution of sodium borodeuteride (1.13 g, 27 mmol; 98 atom %D) in the same mixed solvent (30 cm^3) at 0 °C under nitrogen, and the mixture was kept for 2 h at 0 °C and then for 1 h at room temperature. The product was poured into 1 mol dm^{-3} hydrochloric acid and worked up with diethyl ether in the usual way to give an oil, which was chromatographed on silica gel and eluted with ethyl acetate–hexane (3:7). Distillation gave *ethyl* 8-*hydroxy*-[8,8-$^2H_2$]*octanoate* **19** (5.4 g, 63%), b.p. 86–88 °C/0.1 mmHg (Found: *m/z*, 145.120. $C_{10}H_{18}D_2O_3 - C_2H_5O$ requires *m/z*, 145.120); δ_H(80 MHz) 1.25 (3 H, t, OCH_2Me), 1.10–1.70 (10 H, br m, 5 × CH_2), 2.03 (1 H, s, OH), 2.29 (2 H, t, CH_2CO_2Et) and 4.13 (2 H, q, OCH_2Me); ν_{max}(film)/cm^{-1} 3610–3100br (OH), 2190, 2090 (C–D) and 1730 (C=O).

Ethyl 7-([^{2}H]*Formyl*)*heptanoate* **20**.—A solution of ethyl 8-hydroxy-[8,8-$^2H_2$]octanoate **19** (5 g, 26 mmol) in dry dichloromethane (10 cm^3) was added to a solution of PDC (14.8 g, 39.5 mmol) in dichloromethane (100 cm^3) under nitrogen and stirred overnight. Dry diethyl ether (150 cm^3) was added and the mixture was triturated and filtered through a short Florisil column. Evaporation and distillation gave ethyl 7-([^{2}H]formyl)octanoate **20** (2.5 g, 51%), b.p. 62–64 °C/0.05 mmHg; δ_H(90 MHz) 1.25 (3 H, t, *J* 7, CH_2Me), 1.10–1.80 (8 H, m, 4 × CH_2), 2.35 (2 H, t, CH_2CO_2Et), 2.47 (2 H, t, *J* 7, CH_2CDO) and 4.18 (2 H, q, *J* 7, CH_2Me); ν_{max}(film)/cm^{-1} 2080 (C–D), 1740 (ester) and 1715 (aldehyde). The *semicarbazone* had m.p. 80–81 °C (Found: C, 54.45; H, 8.85; N, 17.5; *m/z*, 227.139. $C_{11}H_{20}DN_3O_3$ requires C, 54.1; H, 9.05; N, 17.2%; M − NH_3, 227.1404).

(8S)-8-*Hydroxy*-[8-^{2}H]*octanoic Acid* **21**.—A solution of ethyl 7-([^{2}H]formyl)heptanoate **20** (1.7 g, 9 mmol) in ethanol (1 cm^3) was added to a rapidly fermenting mixture of bakers' yeast (*Saccharomyces cerevisiae*) [30 g, Sigma YSC type(II)] and glucose (30 g), in tap water (800 cm^3) at 30 °C, during 2 h. After the mixture had been stirred (16 h) and passed through a short Florisil column the filtrate was extracted with diethyl ether. The extracts were washed (water), dried ($MgSO_4$), evaporated and the oil thus produced was chromatographed on silica gel, with ethyl acetate–light petroleum (2:3, containing 0.2% acetic acid) as eluent. (8S)-8-*Hydroxy*-[8-^{2}H]*octanoic acid* **21** (0.85 g, 58%) was crystallised from ethyl acetate–light petroleum, m.p. 60–61 °C (Found: C, 59.7; H, 10.3. $C_8H_{15}DO_3$ requires C, 59.6; H, 10.15%); δ_H(250 MHz) 1.34 (6 H, s-m, 3 × CH_2), 1.54–1.65 (4 H, m, $CH_2CH_2CO_2H$ and CH_2CH-DOH), 2.33 (2 H, t, *J* 7, CH_2CO_2H), 3.62 (1 H, t, *J* 6.5,

C*H*DOH) and 6.77 (2 H, br s, CO_2H and OH); δ_D(40 MHz; $CHCl_3$) 3.64 (1 D, s, 8-D); δ_C(63 MHz) 24.7 (C-3), 25.5 (C-6), 28.7 and 29.0 (CH_2s), 32.5 (C-7), 34.1 (C-2), 62.5 (t, C-8) and 179.1 (C-1); ν_{max}(KBr)/cm^{-1} 3460 (OH), 3600–2700br (OH), 2160 (C–D) and 1690 (C=O). The *Mosher's ester derivative* of the methyl ester (CH_2N_2) had δ_H(400 MHz) 1.30 (6 H, m, 3 × CH_2), 1.59 (2 H, m, CH_2C$H_2$$CO_2$Me), 1.67 (2 H, m, OCHDC$H_2$), 2.29 (2 H, t, *J* 7.5, C$H_2$$CO_2$Me), 3.55 (3 H, t, *J* 1.2, OMe), 3.66 (3 H, s, CO_2Me), 4.31 (1 H, t, *J* 6.6, CHD), 7.39 (3 H, m, 3 × ArH) and 7.52 (2 H, m, 2 × ArH).

(8S)-8-(*p-Tolylsulfonyloxy*)-[8-^{2}H]*octanoic Acid* **22**.—(8*S*)-8-Hydroxy-[8-^{2}H]octanoic acid **21** (550 mg, 3.4 mmol) and toluene-*p*-sulfonyl chloride (683 mg, 3.6 mmol) were stirred in anhydrous pyridine (3 cm^3) at 0 °C for 2 h and the mixture was poured into 2 mol dm^{-3} hydrochloric acid. Extraction with diethyl ether and work-up gave, after chromatography on silica gel [eluent ethyl acetate–light petroleum (3:7)], the *acid* **22** (670 mg, 62%), m.p. 35–36 °C (Found: *m/z*, 297.113. $C_{15}H_{21}DO_5S$ – H_2O requires *m/z*, 297.115); δ_H(80 MHz) 1.20–1.80 (10 H, m, 5 × CH_2), 2.33 (2 H, t, *J* 7, C$H_2$$CO_2$H), 2.45 (3 H, s, Me), 4.00 (1 H, t, OCHD), 7.33 (2 H, d, *J* 8.1, 2 × ArH) and 7.78 (2 H, d, *J* 8.1, 2 × ArH); ν_{max}(film)/cm^{-1} 3600–2500br (OH), 3050 (Ar C–H), 2190 (C–D), 1700 (C=O) and 1595 (Ar).

(8*R*)-[8-^{2}H]*Dec-9-ynoic Acid* **23**.—A mixture of the tosate **22** (800 mg, 2.5 mmol) in anhydrous DMSO (2 cm^3) was added to a suspension of lithium acetylide–ethylenediamine complex (600 mg, 6.2 mmol) in DMSO (2 cm^3) and the whole was stirred at room temperature for 2 h. The product was poured into ice–2 mol dm^{-3} hydrochloric acid and extracted with diethyl ether. Work-up and bulb-to-bulb distillation gave (8*R*)-[8-^{2}H]dec-9-ynoic acid **23** (296 mg, 70%), b.p. 95–100 °C/0.1 mmHg as an oil, which was solid at 0 °C; δ_H(80 MHz) 1.20–1.80 (10 H, m, 5 × CH_2), 1.93 (1 H, d, *J* 2.6, HC≡C), 2.18 (1 H, m, CHD), 2.35 (2 H, t, *J* 7.1, CH_2CO_2H) and 9.80 (1 H, br s, CO_2H); ν_{max}(film)/cm^{-1} 3300 (terminal HC≡C), 3600–2700br (OH), 2120 (C≡C) and 1720 (C=O).

(8R)-[8-^{2}H]*Octadeca-9,12-diynoic Acid* **25**.—A solution of (8*R*)-[8-^{2}H]*dec-9-ynoic acid* **23** (132 mg, 0.78 mmol) in anhydrous THF (1 cm^3) was added to a stirred solution of ethylmagnesium bromide (2.0 mol dm^{-3} in THF; 0.78 cm^3) in THF (2 cm^3) at 0 °C under nitrogen. The mixture was then heated at 50 °C for 30 min, cooled and copper(I) bromide–dimethyl sulfide complex (20 mg) was added to the stirred mixture during 10 min. The reaction mixture was cooled to 0 °C and a solution of 1-(*p*-tolylsulfonyloxy)oct-2-yne **24** (200 mg, 0.72 mmol) in THF (2 cm^3) was added: the mixture was stirred at 0 °C (1 h) and then at 20 °C for 3 h. Saturated aq. ammonium chloride was added and the product was worked up with diethyl ether, and washed as usual. Chromatography on silica gel [eluent ethyl acetate–light petroleum (1:9 with 0.1% acetic acid)] gave the diyne acid **25** (82 mg, 42%) as a crystalline solid, which was used immediately in the reduction (below).

Methyl (8R)-[8-^{2}H]*Octadeca-9(Z),12(Z)-dienoate* **5**, R = Me. —The procedure for semi-hydrogenation of the diyne acid **25** was as described above and gave (8*R*)-[8-^{2}H]octadeca-9(*Z*),12(*Z*)-dienoic acid **5**, R = H in 80% yield (65 mg). The acid was esterified with diazomethane to give the *title ester* **5**, R = Me (55 mg, 81%) (Found: M^+, 295.263. $C_{19}H_{33}DO_2$ requires *M*, 295.262); δ_H(400 MHz) 0.93 (3 H, t, Me), 1.31 (16 H, m, 8 × CH_2), 1.62 (2 H, m, CH_2C$H_2$$CO_2$Me), 2.06 (3 H, m, 8-H and 14-H_2), 2.30 (2 H, t, *J* 7.5, C$H_2$$CO_2$Me), 2.77 (2 H, t, *J* 6.2, 11-H_2), 3.66 (3 H, s, OMe) and 5.35 (4 H, m, olefinic 9-, 10-, 12-, 13-H); δ_D(40 MHz) 2.05 (1 D, s, 8-D); δ_C(100 MHz) 14.07 (C-18), 22.58 (C-17), 24.96 (C-3), 25.64 (C-11), 26.84 (C-8, t), 27.21 (C-14), 29.08, 29.13, 29.16, 29.36 and 29.51 (CH_2s), 31.54 (C-16), 34.11 (C-2), 51.43 (OMe), 127.92 (C-12), 128.08 (C-10), 130.00 (C-9), 130.21 (C-13) and 174.30 (C-1); ν_{max}(film)/cm^{-1} 3020 (olefinic C–H), 2150 (C–D), 1740 (C=O) and 1650 (C=C).

Methyl (8R)-8-*Formyloxy*-[8-^{2}H]*octanoate* **26**.—Methyl (8*S*)-8-hydroxy-[8-^{2}H]octanoate (30 mg, 0.17 mmol), triphenylphosphine (54 mg, 0.20 mmol), and anhydrous formic acid (3 drops) were treated in anhydrous THF (2 cm^3) with a solution of DEAD (35 mg, 0.20 mmol) in THF (0.5 cm^3) under nitrogen at 0 °C. The mixture was stirred (4 h), concentrated under reduced pressure and extracted with hexane to give, after work-up and chromatography on silica gel [eluent ethyl acetate–light petroleum (1:9)], *the title ester* **26** (30 mg, 81%) (Found: *m/z*, 172.108. $C_{10}H_{17}DO_4$ – CH_3O requires *m/z*, 172.108); δ_H(80 MHz) 1.20–1.70 (10 H, m, 5 × CH_2), 2.31 (2 H, t, C$H_2$$CO_2$Me), 3.67 (3 H, s, OMe), 4.14 (1 H, m, CHD) and 8.05 (1 H, s, HCO_2); ν_{max}(film)/cm^{-1} 3030 (formate C–H), 2200 (C–D) and 1725 (C=O).

Methyl (8R)-8-*Hydroxy*-[8-^{2}H]*octanoate* **27**.—A solution of the formyloxy compound **26** (30 mg, 0.17 mmol) in methanol (3 cm^3) was treated with conc. aq. ammonia (36%; 1 drop) for 2 h at room temperature. Work-up gave the title ester **27** (24 mg, 80%); δ_H(80 MHz) 1.20–1.70 (10 H, m, 5 × CH_2), 2.30 (2 H, t, C$H_2$$CO_2$Me), 3.55 (1 H, m, C*H*DOH) and 3.66 (3 H, s, OMe); ν_{max}(film)/cm^{-1} 3600–3100br (OH), 2150 (C–D) and 1725 (C=O). The *Mosher's ester derivative* [δ_H(400 MHz) 4.27 (1 H, t, *J* 6.6, CHDO] was prepared by the usual method (Found: *m/z*, 158.129. $C_{18}H_{24}DF_3O_5$ – $C_9H_8O_3F_3$ requires *m/z*, 158.129) (See Table 1).

(8R)-9-*Oxo*-[8-^{2}H]*decanoic Acid* **28** *by Acetylene Hydration*.—(8*R*)-[8-^{2}H]Dec-9-ynoic acid **23** (71 mg, 0.42 mmol) was added to a stirred mixture of mercury(II) acetate (530 mg, 1.6 mmol), acetic acid (3 cm^3), and water (0.5 cm^3) at 70 °C and the mixture was stirred at this temperature for 2 h. Water (15 cm^3) was added and the pH was adjusted to 7 by the addition of sodium acetate, when sodium borohydride (150 mg, 4 mmol) was added to the solution kept at 0 °C and the whole was stirred for 30 min. The product was poured into water, acidified (pH 2) and extracted into diethyl ether. Work-up gave an oil, which was dissolved in dichloromethane (1 cm^3) and added to a solution of PDC (180 mg, 0.47 mmol) in dichloromethane (1 cm^3), and the mixture was stirred under nitrogen at 20 °C for 3 h. Diethyl ether (5 cm^3) was added and the mixture was filtered through flash silica, the filtrate being concentrated under reduced pressure to yield (8*R*)-9-oxo-[8-^{2}H]decanoic acid **28** (35 mg, 44%); δ_H(80 MHz) 1.20–1.75 (10 H, m, 5 × CH_2), 2.19 (3 H, s, 9-H_3), 2.25–2.55 (3 H, m, C$H_2$$CO_2$H and COCHD) and 9.70 (1 H, br, s, CO_2H); ν_{max}($CHCl_3$)/cm^{-1} 1715 (acid and ketone).

(8R)-8-*Acetoxy*-[8-^{2}H]*octanoic Acid* **29**.—Trifluoroacetic anhydride (3.16 cm^3, 22 mmol) in dry dichloromethane (3 cm^3) was added to 85% hydrogen peroxide (0.435 cm^3) in dichloromethane (8 cm^3) at 0 °C during 30 min under nitrogen, and the mixture was stirred for 1 h to form 1.0 mol dm^{-3} peroxytrifluoroacetic acid. The latter solution (0.5 cm^3) was added during 10 min to a mixture of the oxo acid **28** (35 mg, 0.19 mmol) and anhydrous disodium hydrogen phosphate (350 mg) in dichloromethane (2 cm^3) at room temperature under nitrogen, and the mixture was stirred for 2 h. Water was added and the mixture was worked up with diethyl ether to give (8*R*)-8-acetoxy-[8-^{2}H]octanoic acid **29** (24 mg, 63%); δ_H(80 MHz) 1.20–1.75 (10 H, m, 5 × CH_2), 2.04 (3 H, s, MeCO), 2.35 (2 H, t, CH_2CO_2H) and 4.04 (1 H, t, C*H*D); ν_{max}($CHCl_3$)/cm^{-1} 1720 (ester and acid C=O).

Methyl (8R)-8-*Hydroxy*-[8-^{2}H]*octanoate* **27** *from the Baeyer–Villiger Sequence.*—The acetoxyoctanoic acid **29** (24 mg, 0.12 mmol) in ethanol (0.5 cm^3) was hydrolysed at 20 °C by the addition of 40% aq. potassium hydroxide (2 cm^3) and ethanol (4 cm^3), and the mixture was kept for 18 h. Work-up by acidification to pH 2 at 0 °C and extraction with ethyl acetate gave the acid parent of compound **27** (12 mg, 64%), which was esterified with diazomethane in diethyl ether and chromatographed on silica gel with ethyl acetate–light petroleum (3:7) as eluent to give methyl (8*R*)-8-hydroxy-[8-^{2}H]octanoate **27** [δ_H(400 MHz) 4.27 (1 H, t, *J* 6.6, CHDO)] except for optical properties spectroscopically identical with the sample made by the Mitsunobu method above. Examination as the Mosher ester showed 92.5% (8*R*) and 7.5% (8*S*) isomers.

Ethyl [2,2-$^2H_2$]*Non*-3-*ynoate* **35.**—Potassium carbonate (700 mg, anhydrous) in deuterium oxide (15 cm^3, 99.8 atom%) was vigorously stirred as a two-phase system with ethyl diazoacetate (4.3 g, 38 mmol) in anhydrous diethyl ether (15 cm^3) for 12 h. The aqueous layer was removed and the exchange was repeated. After separation the aqueous layer was extracted with diethyl ether and the combined ether layers were dried ($MgSO_4$), and evaporated under reduced pressure to give ethyl deuteriodiazoacetate (2.8 g, 65%), δ_H(90 MHz) 1.30 (3 H, t, Me) and 4.29 (2 H, q, CH_2). Deuterium incorporation was ≥atom 98%.

A solution of hept-1-yne (3.93 cm^3, 30 mmol) in anhydrous THF (30 cm^3) was stirred at −5 °C under nitrogen and treated with butyllithium (1.6 mol dm^{-3} in hexane; 19 cm^3) during 10 min and the mixture was then stirred at 0 °C for 1 h. After cooling to −40 °C, the mixture was treated with boron trifluoride–diethyl ether (4.92 cm^3, 40 mmol) in THF (5 cm^3) dropwise and the mixture was stirred at −40 °C for 30 min. Ethyl deuteriodiazoacetate (2.5 g, 22 mmol) was added during 1 h and the stirred mixture was maintained at −40 °C for 2 h before warming to room temperature. Deuterium oxide (15 cm^3; 99.8 atom%) was added and the mixture was stirred overnight. Extraction with diethyl ether, work-up and distillation gave *ethyl* [2,2-$^2H_2$]*non*-3-*ynoate* **35** (1.67 g, 90%), b.p. 60–62 °C/0.1 mmHg (lit.,[23] for unlabelled material b.p. 90–91 °C/2.5 mmHg) (Found: M$^+$ −29, 155.106. $C_9H_{11}D_2O_2$ requires *m/z*, 155.104); δ_H(400 MHz) 0.90 (3 H, t, Me), 1.28 (3 H, t, OCH_2Me), 1.33 (4 H, m, 2 × CH_2), 1.51 (2 H, m, CH_2), 2.19 (2 H, t, $CH_2C{\equiv}CCD_2$) and 4.18 (2 H, q, OCH_2Me). The isotopic purity at C-2, as judged by ^{1}H NMR spectroscopy, was >96 atom%; δ_C(100 MHz) 14.0 (Me), 14.2 (Me), 18.8 (C-5), 22.3 (C-8), 25.7 (vestigial m, C-2), 28.4 (C-6), 31.1 (C-7), 61.4 (OCH_2), 71.4 (C-4), 83.9 (C-3) and 169.1 (C-1); ν_{max}(film)/cm^{-1} 2260 (C≡C), 2150 (C–D) and 1740 (C=O).

[2,2-$^2H_2$]*Non*-3-*yn*-1-*ol and its Acetate* **36.**—Lithium aluminium hydride (1.0 mol dm^{-3} in THF; 6 cm^3) was added to a solution of ethyl [2,2-$^2H_2$]non-3-ynoate (1.71 g, 9.2 mmol) in anhydrous THF (20 cm^3) at −5 °C under nitrogen, and the mixture was stirred (20 min). Decomposition with ethyl acetate and work-up gave [2,2-$^2H_2$]*non*-3-*yn*-1-*ol* (985 mg, 75%), b.p. 42–44 °C/0.1 mmHg (Found: M$^+$, 142.130. $C_9H_{14}D_2O$ requires *M*, 142.133); δ_H(80 MHz) 0.90 (3 H, t, Me), 1.2–1.7 (6 H, br m, 3 × CH_2), 1.93 (1 H, s, OH), 2.15 (2 H, t, $CH_2C{\equiv}C$) and 3.66 (2 H, s, CH_2OH); ν_{max}(film)/cm^{-1} 3650–3110 (OH), 2205 (C≡C) and 2130 (C–D).

The alcohol (0.92 g, 6.5 mmol) and DMAP (50 mg) in a mixture of anhydrous pyridine (0.8 cm^3, 9.7 mmol) and acetic anhydride (0.92 cm^3, 9.7 mmol) were stirred at room temperature under nitrogen for 2 h. Work-up gave [2,2-$^2H_2$]non-3-ynyl acetate **36** (1.13 g, 95%); δ_H(80 MHz) 0.90 (3 H, t, Me), 1.20–1.70 (6 H, br m, 3 × CH_2), 2.05 (3 H, s, COMe), 2.13 (2 H, t, $CH_2C{\equiv}C$) and 4.12 (2 H, s, CH_2OAc); ν_{max}(film)/cm^{-1} 2245 (C≡C), 2120 (C-D) and 1745 (C=O).

[2,2-$^2H_2$]*Non*-3(Z)-*enyl Acetate.*—2-Methyl-but-2-ene (0.34 cm^3) was added to a stirred mixture of sodium borohydride (50 mg, 1.3 mmol) in dry diglyme (4 cm^3). After the mixture had been cooled to 0 °C, boron trifluoride–diethyl ether (0.21 cm^3, 1.7 mmol) was added and the mixture was maintained at 0 °C (2 h). A solution of [2,2-$^2H_2$]non-3-ynyl acetate **36** (218 mg, 1.2 mmol) in diglyme (0.5 cm^3) was added and the mixture was stirred for 30 min at 0 °C, then at 20 °C for 2 h. After the mixture had been cooled to 0 °C, ethylene glycol (0.2 cm^3) and then acetic acid (1 cm^3) were added. The mixture was stirred (15 h) and water (20 cm^3) and 2 mol dm^{-3} hydrochloric acid (to attain pH 1) were added. Work-up with hexane gave an oil, which was treated with 30% hydrogen peroxide (3 cm^3) in THF (5 cm^3) containing sodium acetate (500 mg). Work-up with hexane and chromatography on silica gel [eluent ethyl acetate–hexane (1:9)] gave the *title acetate* (140 mg, 60%) (Found: *m/z*, 126.136. $C_{11}H_{18}D_2O_2 - C_2H_4O_2$ requires *m/z*, 126.134); δ_H(80 MHz) 0.89 (3 H, t, Me), 1.20–1.70 (6 H, br m, 3 × CH_2), 2.03 (3 H, s, COMe), 2.10 (2 H, m, CH_2), 4.05 (2 H, s, CH_2OAc) and 5.2–5.7 (2 H, m, 3- and 4-H); ν_{max}(film)/cm^{-1} 3030 (olefinic C-H), 2205, 2120 (C–D), 1740 (C=O), 1660 (C=C) and 745.

[2,2-$^2H_2$]*Non*-3(Z)-*en*-1-*ol* **37.**—A solution of the above acetate (140 mg, 0.75 mmol) in dry THF (2 cm^3) was treated with lithium aluminium hydride (1.0 mol dm^{-3} in THF; 1 cm^3) under nitrogen at 0 °C. Work-up gave [2,2-$^2H_2$]*non*-3(Z)-*en*-1-*ol* **37** (92 mg, 85%) (Found: M+, 144.148. $C_9H_{16}D_2O$ requires *M*, 144.148); δ_H(400 MHz) 0.89 (3 H, t, Me), 1.23–1.43 (6 H, br m, 3 × CH_2), 1.63 (1 H, br s, OH), 2.05 (2 H, m, 5-H_2), 3.63 (2 H, s, CH_2OH), 5.35 (1 H, d, *J* 10.8, 3-H) and 5.57 (1 H, dt, $J_{4,3}$ 10.8, $J_{4,5}$ 7.3, 4-H); δ_D(40 MHz) 2.31 (2 D, s, 2-D_2); δ_C(100 MHz) 13.8 (C-9), 22.4 (C-8), 27.2 (C–5), 29.2 (C-6), 30.0 (m, C-2), 31.3 (C-7), 62.1 (C-1), 124.7 (C-4) and 133.3 (C-3).

[2,2-$^2H_2$]*Non*-3(Z)-*enal* **6.**—A solution of the above nonenol **37** (60 mg, 0.42 mmol) in deacidified dichloromethane (0.5 cm^3) was added to a stirred suspension of Dess–Martin periodinane (265 mg, 0.62 mmol) in dichloromethane (2 cm^3) at 20 °C under nitrogen. The mixture was stirred (1 h) and diluted with diethyl ether (20 cm^3). The organic solution was stirred (15 min) with saturated aq. sodium hydrogen carbonate (10 cm^3) containing sodium thiosulfate (1.5 g) and was then washed (water), dried ($MgSO_4$), and evaporated under reduced pressure to give [2,2-$^2H_2$]*non*-3(Z)-*enal* **6** (50 mg, 83%) (Found: M$^+$, 142.130. $C_9H_{14}D_2O$ requires *M*, 142.126). As prepared the sample contained 90% of the aldehyde and 10% of the unchanged alcohol by GLC. The pure aldehyde is unstable and it is recommended that it is prepared and purified by GLC as required from a stock of the alcohol. The aldehyde had δ_H(80 MHz) 0.89 (3 H, t, Me), 1.10–1.70 (6 H, m, 3 × CH_2), 2.03 (2 H, m, 5-H_2), 5.30–5.90 (2 H, m, 3- and 4-H) and 9.65 (1 H, s, CHO); ν_{max}(film)/cm^{-1} 3020 (olefinic C–H), 2720 (aldehyde C–H), 2220, 2120 (C–D), 1720 (C=O) and 1650 (C=C).

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