Synthesis of Methyl (1*R*,2*S*)-2-[(1'*Z*,4'*Z*,7'*Z*)-Hexadeca-1',4',7'-trienyl]cyclopropanecarboxylate: a Potential Inhibitor of the Enzyme 5-Lipoxygenase

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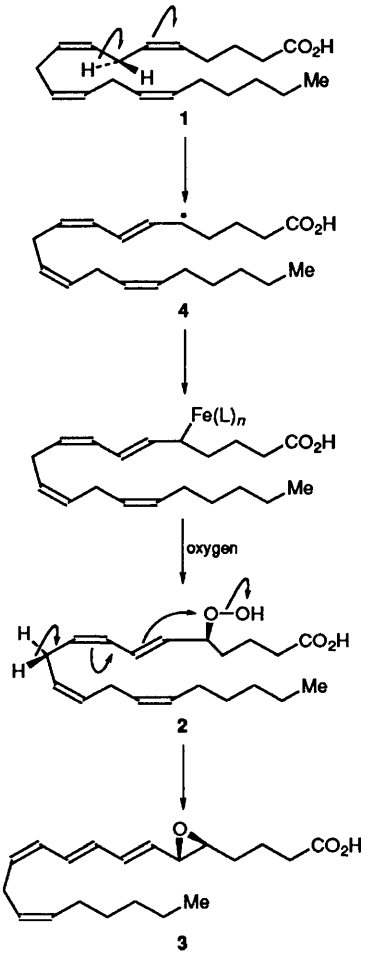
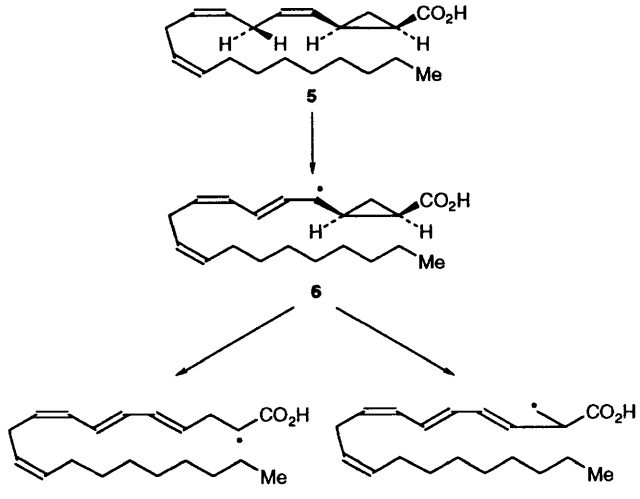
We describe the synthesis of a novel cyclopropyl analogue of arachidonic acid *via* a convergent synthesis that employed methyl (1*R*,2*S*)-2-formylcyclopropanecarboxylate in conjunction with the ylide from (3*Z*,6*Z*)-pentadeca-3,6-dienyl(triphenyl)phosphonium iodide. This compound was designed to inhibit the enzyme 5-lipoxygenase after reaction to form a putative α-cyclopropylmethylene radical.

The enzyme 5-lipoxygenase catalyses the conversion of arachidonic acid **1** into the hydroperoxide **2** and thence into leukotriene A_4 **3**[1] (Scheme 1). This then acts as a precursor



Scheme 1

of the other leukotrienes which are implicated in many inflammatory and allergic conditions. Clearly inhibitors of this enzyme may have therapeutic value in the treatment of such conditions, and much work has already been carried out in this area.[2] One impediment to the rational design of inhibitors is the lack of knowledge of the mechanism of the conversion of **1** into **2**. The mechanism shown in Scheme 1[3] is but one of several that have been proposed, and most envisage the initial formation of the allylic free radical **4**. In an attempt to provide some evidence for such a radical, we embarked upon a synthesis of the cyclopropyl analogue **5** of arachidonic acid. We reasoned that this compound accessible from (2*R*)-glyceraldehyde acetonide could divert the course of the reaction shown in Scheme 1 *via* formation of the α-cyclopropylmethylene radical **6** (Scheme 2).



Scheme 2

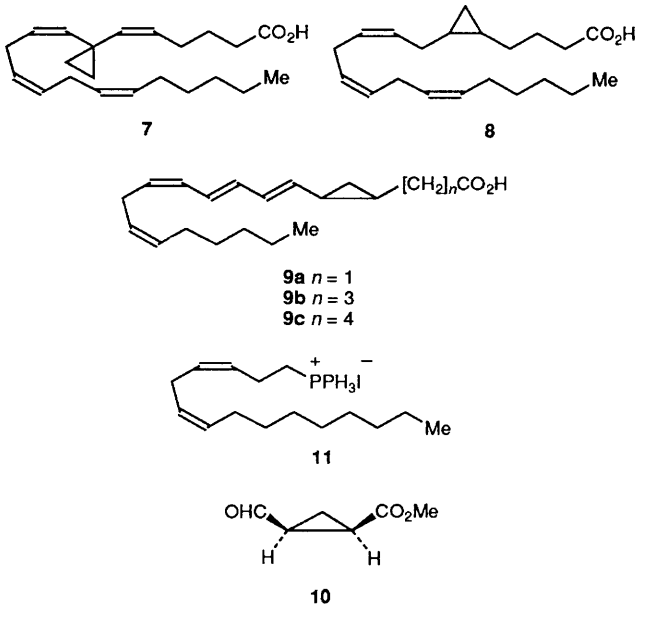
Others have prepared cyclopropyl analogues of arachidonic acid, *e.g.* compounds **7** to **9**,[4–6] and although compounds **7** and **8** were essentially devoid of inhibitory activity against 5-lipoxygenase, compounds **9a** and **9c** had IC_{50} values of around 20 μmol. Compound **5** had not, however, been prepared, and we can now report its synthesis from the two fragments **10** and **11**.

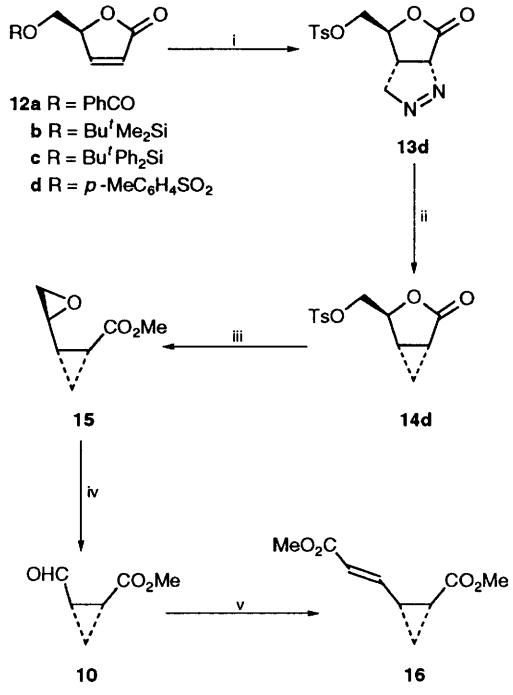
The stereochemically-defined fragment **10** was prepared *via* the route shown in Scheme 3. We have already described[7] an efficient method for the formation of the pyrazoline **13a** and its photolysis to provide the oxabicycle **14a**, but show in Table 1 the results of more extensive studies to determine the optimum reaction conditions. In the event, the tosyl derivative **14d** proved to be the most synthetically useful and was prepared routinely and reproducibly on the multigram scale. Reaction of this compound with sodium methoxide in methanol provided a near quantitative yield of the epoxy ester **15**. Oxidative cleavage of this using periodic acid in ether–1,4-dioxane (1:1) yielded the aldehyde ester **10** in somewhat disappointing yield (50%). All spectroscopic data were fully consistent with this structure, but further characterisation was obtained following reaction with the stabilised ylide (methoxycarbonylmethylidene)triphenyl-

Table 1

Protecting group[a]	Solvent conditions	Irradiation *t*/h	Yield of photo product (%)
Benzoyl	CH_2Cl_2	4	35
13a	Benzene–acetonitrile (1:1)–benzophenone (0.5 equiv.)	2	40–75
	CH_2Cl_2–benzophenone (0.5 equiv.)	2	64
TBDMS	CH_2Cl_2–benzophenone (0.5 equiv.)	8	*ca.* 50
13b	Benzene–acetonitrile (1:1)	8	—
TBDPS	Benzene–acetonitrile (1:1)–benzophenone (0.1 equiv.)	4	5
13c	CH_2Cl_2	2	12
Tosyl	CH_2Cl_2	2	45–54
13d	CH_2Cl_2–benzophenone (0.5 equiv.)	2	51
	Benzene–acetonitrile (1:1)–benzophenone (0.1 equiv.)	2	50

[a] Benzoyl = PhCO, TBDMS = *tert*-butyldimethylsilyl, TBDPS = *tert*-butyldiphenylsilyl, Tosyl = *p*-$MeC_6H_4SO_2$.





Scheme 3 *Reagents and conditions:* i, diazomethane, dichloromethane (DCM) (81%); ii, *hv*, DCM (54%); iii, NaOMe, MeOH (90%); iv, HIO_4, ether–1,4-dioxane (1:1) (50%); $Ph_3P{=}CHCO_2Me$, DCM

phosphorane to produce the *trans*-diester **16**. High field ^1H and ^{13}C NMR spectra obtained for this compound exhibited discrete signals with no evidence of stereoisomeric products.

Several routes were explored for the preparation of the requisite phosphonium salt **11**, but the one shown in Scheme 4 was ultimately used. The THP ether of but-1-yn-4-ol **17** was converted into its lithium salt and then treated sequentially with $BF_3{\cdot}Et_2O$ and ethylene oxide to provide the anticipated alcohol **18** in 80% isolated yield. Conversion of this into the toluene-*p*-sulfonate **19**, then the iodide **20**, and finally into the phosphonium salt **21** was accomplished in an overall yield of around 60% (for the three steps). A Wittig reaction of this with nonanal yielded the expected *cis*-alkene **22**, and this was selectively hydrogenated (Lindlar catalyst, THF) to produce the all-*cis*-dienol **23** in excellent yield (88%). Removal of the THP group was accomplished using aqueous acetic acid (86%) to provide the alcohol **24** which was converted into the iodide **25** (I_2–imidazole in DCM)[8] (73%), and thence into the desired phosphonium salt **11** (essentially quantitative).

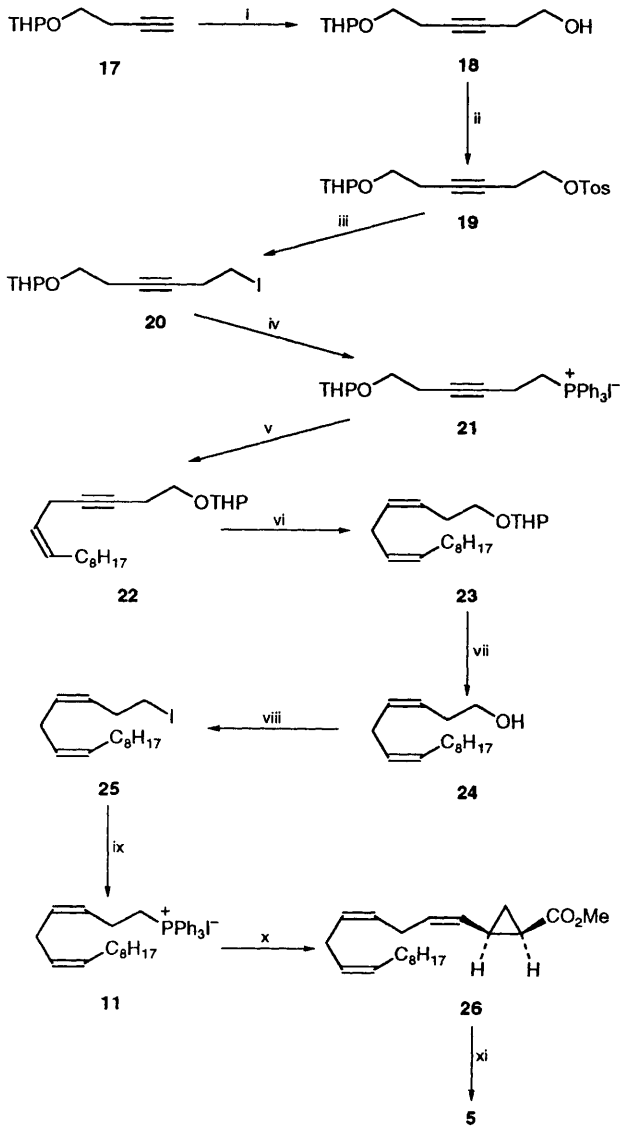
Finally, the key Wittig reaction between the phosphorus ylide formed from **11** using butyl lithium and the aldehyde **10** was accomplished to provide a poor yield of the ester **26** (12%). The desired cyclopropyl analogue **5** could then be obtained after treatment with methanolic LiOH (48%). The ^1H NMR of **26** exhibited discrete signals for the ester methoxy and C-19 methyl with no evidence for stereoisomeric products, though the NMR of **5** was too complex to provide definitive proof of stereochemical purity.

A preliminary bioassay was carried out in collaboration with Cascade Biochemicals Ltd. using commercially available 5-lipoxygenase from the coral *Plexaura homomalla* and also 5-LO prepared from potato skins. No evidence that the analogue was acting either as a substrate or an inhibitor was obtained, but it is hoped to carry out further experiments to confirm these negative results.

Although these initial results are disappointing, the methodology described here allows easy access to the key cyclopropyl compound **10**, which is a key intermediate in our current syntheses of novel amino acids containing cyclopropyl rings.

Experimental

IR spectra were recorded on a Perkin-Elmer 881 double beam grating spectrometer using sodium chloride cells and chloroform solutions unless otherwise stated. NMR spectra were recorded at 250 MHz using a Bruker WM250 spectrometer and



Scheme 4 *Reagents and conditions:* i, BuLi, tetrahydrofuran (THF), then BF_3·Et_2O, ethylene oxide (80%); ii, *p*-$MeC_6H_4SO_2Cl$, pyridine, DCM (70%); iii, NaI, acetone (90%); iv, Ph_3P, MeCN (96%); v, BuLi, THF, then nonanal (62%); vi, Lindlar catalyst, hydrogen, THF (88%); vii, HOAc–THF–water (4:2:1) (88%); viii, Ph_3P, I_2, imidazole, DCM (86%); ix, Ph_3P, MeCN (96%); x, BuLi, THF, then compound **10** (12%); xi, LiOH, THF (48%)

at 300 MHz using a Bruker WM300 instrument. *J*-Values are given in Hz. M.p.s were determined using an Electrothermal digital apparatus and are uncorrected. Reactions using anhydrous conditions were carried out under a static nitrogen atmosphere using oven-dried glassware. THF (tetrahydrofuran) was dried over molecular sieves and distilled from sodium–benzophenone prior to use. All other reagents and solvents were used as supplied without further purification. Flash chromatography was performed using Sorbosil C60 silica. Ether refers to diethyl ether and petrol to light petroleum, boiling point range 40–60 °C unless otherwise stated. All optical rotations were measured at 20 °C, and are given in units of 10^{-1} deg cm^2 g^{-1}.

5-*Tosyloxymethylfuran*-2(5H)-*one* **12d**.—To a solution of the (*S*)-5-hydroxymethylfuran-2(5*H*)-one (0.34 g, 0.002 mol) in dichloromethane (12 cm^3, dry) and pyridine (5 cm^3, dry) under nitrogen at 0 °C was added toluene-*p*-sulfonyl chloride (0.50 g, 0.0026 mol) in dichloromethane (4 cm^3, dry), dropwise by syringe. The solution was stirred for 1 h at 0 °C and then transferred to the freezer and stored overnight (*ca.* 16 h) at −20 °C. The reaction mixture was then washed with 10% aqueous citric acid solution (3 × 50 cm^3), the combined organic layers were dried (Na_2SO_4) and concentrated under reduced pressure to yield a colourless oil. This was dissolved in a minimum of dichloromethane and to this was added *ca.* 5 times the volume of petrol and the solution cooled slowly to −78 °C to initiate crystallisation. The yellow-white solid which formed was washed with cold ethanol and the solid dried under reduced pressure to yield the title compound as a white crystalline solid (0.34 g, 54%), R_f (9:1 ether–ethyl acetate) 0.33; m.p. 54.5–55.0 °C; ν_{max}/cm^{-1} 1776 (C=O, lactone) 1675, 1599, 1370, 1191, 1160, 1093, 994 and 814; $\delta(CDCl_3$; 250 MHz) 2.48 (s, 3 H, $ArCH_3$), 4.26 (d, 2 H, $J_{1',5}$ 6, 1'-H_2), 5.20–5.28 (m, 1 H, 5-H), 6.22 (dd, 1 H, $J_{3,4}$ 6, $J_{3,5}$ 1.5, 3-H), 7.38 (d, 2 H, 2 × Ar-H), 7.48 (dd, 1 H, $J_{4,3}$ 6, $J_{4,5}$ 1.5, 4-H) and 7.76 (d, 2 H, 2 × Ar-H); $[\alpha]_D$ −52 (*c* 1.8, $CHCl_3$) (Found: C, 53.4; H, 4.6. $C_{12}H_{12}O_5S$ requires C, 53.72; H, 4.51%).

2-*Tosyloxymethyl*-7-*oxa*-2,3-*diazabicyclo*[3.3.0]*oct*-2-*en*-8-*one* **13d**.—To a solution of the furanone **12d** (1.25 g, 0.005 mol) in dichloromethane (20 cm^3) was added a solution of diazomethane (*ca.* 2.0 equiv.) in ether, and the solution was allowed to stand overnight at −20 °C (*ca.* 16 h). The excess diazomethane was destroyed with glacial acetic acid and the white crystalline precipitate formed was filtered from the solution and dried under reduced pressure to yield the pyrazoline as a white crystalline solid, which was purified by recrystallisation from ethanol to yield the title compound (1.18 g, 81%), R_f (9:1 ether–ethyl acetate) 0.21, m.p. (112 °C); ν_{max}/cm^{-1} 1793 (C=O, lactone), 1599, 1452, 1431, 1370, 1223, 1192, 1179, 1095 and 957; $\delta(CDCl_3$; 250 MHz) 2.46 (s, 3 H, Ar-CH_3), 2.90–3.04 (m, 1 H, 5-H), 4.16–4.20 (m, 3 H, 1'-H_2, 6-H), 4.80–4.92 (m, 2 H, 4-H_2), 5.64 (dt, 2 H, $J_{1,5}$ 9, 1-H), 7.38 (d, 2 H, 2 × Ar-H) and 7.76 (d, 2 H, 2 × Ar-H); $[\alpha]_D$ −246 (*c* 2.4, CH_3Cl) (Found: C, 50.2; H, 4.6; N, 8.9. $C_{13}H_{14}O_4N_2S$ requires C, 50.32; H, 4.55; N, 9.03%).

4-*Tosyloxymethyl*-3-*oxabicyclo*[3.1.0]*hexan*-2-*one* **14d**.—A solution of the pyrazoline **13d** (1.10 g, 0.0035 mol) in dichloromethane (120 cm^3) was degassed with nitrogen for 30 min and then irradiated with a medium pressure mercury lamp for 2 h in a Pyrex cell. The resultant solution was concentrated under reduced pressure to yield the crude product as a yellow oil, which was purified by flash column chromatography (9:1 ether–ethyl acetate as eluent) to yield the title compound as a white crystalline solid (0.54 g, 54%), R_f (9:1 ether–ethyl acetate) 0.43, m.p. (89.9–90.1 °C); ν_{max}/cm^{-1} 3020, 1785 (C=O, lactone), 1371, 1190 and 1177; $\delta(CDCl_3$; 250 MHz) 0.84–0.92 (m, 1 H, 6-H_a), 1.22–1.36 (m, 1 H, 6-H_b), 2.06–2.26 (m, 2 H, 1-H, 5-H), 2.46 (s, 3 H, Ar-CH_3), 4.10–4.26 (m, 2 H, 1'-H_2), 4.50 (t, 1 H, $J_{4,1'}$ 7.5, 4-H), 7.38 (d, 2 H, *J* 9, 2 × Ar-H) and 7.78 (d, 2 H, *J* 9, 2 × Ar-H); $[\alpha]_D$ +53 (*c* 2.2, $CHCl_3$) (Found: C, 55.3; H, 5.00. $C_{13}H_{14}O_5S$ requires C, 55.31; H, 5.00%).

Methyl (1R,2S,2'S)-2-(*Oxiran*-2'-*yl*)*cyclopropanecarboxylate* **15**.—To a stirred ice-cooled solution of sodium methoxide (0.40 g, 0.0073 mol) in methanol (30 cm^3, dry) under nitrogen, was added dropwise a solution of the toluene-*p*-sulfonate **14d** (1.35 g, 0.0048 mol). The solution was stirred for 1.5 h and the pH adjusted to pH 7 by careful addition of glacial acetic acid. Water (50 cm^3) and dichloromethane (50 cm^3) were then added and the organic layer washed and dried (Na_2SO_4), and the solvents removed under reduced pressure to yield an orange oil which was purified by flash column chromatography (1:1 ether–petrol as eluent) to yield the title compound as a colourless oil, 0.61 g (90%), R_f (1:1 ether–petrol) 0.32; ν_{max}/cm^{-1} 1724 (C=O, ester), 1179 and 1132; $\delta(CDCl_3$, 250 MHz) 1.09–1.29 (m, 3 H,

2-H, 3-H_2), 1.88 (m, 1 H, 1-H), 2.71 (dd, 1 H, $J_{3'a,3'b}$ 9, $J_{3'a,2}$ 2.1, 3'-H_a), 2.85 (t, 1 H, $J_{3'a,3'b}$ 9, 3'-H_b), 3.09–3.17 (m, 1 H, 2'-H) and 3.76 (s, 3 H, OCH_3) (Found: M^+, 142.063. $C_7H_{10}O_3$ requires M^+, 142.063).

Methyl (1R,2S)-2-*Formylcyclopropanecarboxylate* **10** *and Methyl* (1R,2S)-2-[(E)-2'-*Methoxycarbonylvinyl*]*cyclopropanecarboxylate* **16**.—To a solution of periodic acid (0.96 g, 0.0041 mol) in 1:1 ether–1,4-dioxane solution at room temperature under nitrogen was added a solution of the epoxide **15** (0.51 g, 0.0036 mol) and the resultant mixture was then stirred for 1 h. The reaction mixture was then extracted with 1:1 petrol–ether, the organic layers combined, dried (Na_2SO_4) and concentrated under reduced pressure to yield a yellow oil **10** (0.41 g, 88%), which was used immediately. The product was derivatised for characterisation purposes as follows. Compound **10** was immediately taken up into dichloromethane (10 cm^3), cooled to 0 °C, and to this was added (in portions) methoxycarbonylmethylene(triphenyl)phosphorane (1.46 g, 0.0036 mol). This solution was then stirred for 1 h and then the solvents were removed under reduced pressure to yield a white crystalline solid, which was extracted with boiling petrol (3 × 50 cm^3), the extracts filtered and the filtrate concentrated under reduced pressure to yield a colourless oil, which was purified by flash column chromatography to yield the product **16** as a colourless oil (0.47 g, 75%), R_f (4:1 petrol–ether) 0.35; δ($CDCl_3$; 250 MHz) 0.69 (td, 1 H, $J_{3a,3b}$ 4.6, $J_{3a,1}$ 3.6, $J_{3a,2}$ 3.6, 3-H_a), 1.27 (td, 1 H, $J_{3a,3b}$ 4.6, $J_{3b,1}$ 1.7, $J_{3b,2}$ 1.7, 3-H_b), 1.33 (m, 1 H, 2-H), 1.66 (m, 1 H, 1-H), 3.28 (s, 3 H, OCH_3), 3.41 (s, 3 H, OCH_3), 6.01 (d, 1 H, $J_{2',1'}$ 15.6, 2'-H) and 7.30 (dd, 1 H, $J_{1',2'}$ 15.6, $J_{1',2}$ 5.1, 1'-H) (Found: M^+, 184.074. $C_9H_{12}O_4$ requires M^+, 184.074).

6-(*Tetrahydropyran*-2'-*yloxy*)*hex*-3-*yn*-1-*ol* **18**.—To a solution of 4-(tetrahydropyran-2'-yloxy)but-1-yne **17** (2.7 g, 0.018 mol) in dry THF (110 cm^3) at −78 °C under nitrogen was added dropwise a solution of butyllithium (1.6 mol dm^{-3} in hexanes; 12 cm^3, 0.0192 mol) and the resultant solution stirred for 10 min prior to the addition of boron trifluoride–diethyl ether (2.5 cm^3, 0.02 mol). The resultant solution was stirred again for 10 min before the addition of ethylene oxide (2.0 equiv.), and this solution was then stirred for 30 min at −78 °C. The reaction was then quenched with saturated aqueous ammonium chloride, the products extracted with ether (2 × 100 cm^3). The combined organic extracts were dried ($MgSO_4$) and concentrated under reduced pressure to yield a colourless oil, which was purified by flash column chromatography (9:1 petrol–ether as eluent) to yield the title compound as a colourless oil (2.8 g, 80%), R_f (9:1 ether–petrol) 0.38; ν_{max}/cm^{-1} 2941, 1468, 1454, 1442, 1354, 1158, 1034, 973 and 870; δ($CDCl_3$; 250 MHz) 1.42–1.94 (m, 6 H, 3'-H_2, 4'-H_2, 5'-H_2), 2.38–2.52 (m, 4 H, 2-H_2, 5-H_2), 2.75 (br s, OH), 3.46–3.58 (m, 2 H, 6-H_2 or 6'-H_2), 3.68 (t, 2 H, J 7, 1-H_2), 3.75–3.98 (m, 2 H, 6'-H_2 or 6-H_2) and 4.65 (t, J 4, 2'-H) (Found: M^+, 198.126. $C_{11}H_{18}O_3$ requires M^+, 198.126).

6-(*Tetrahydropyran*-2'-*yloxy*)-1-*tosyloxyhex*-3-*yne* **19**.—To a solution of toluene-*p*-sulfonyl chloride (3.76 g, 0.0197 mol) in pyridine (40 cm^3, dry) was added a solution of alcohol **18** (3.30 g, 0.0171 mol) in dichloromethane (10 cm^3, dry) and the resultant solution left overnight at −20 °C. The reaction mixture was then diluted with dichloromethane (100 cm^3), washed with aqueous 10% citric acid solution (3 × 100 cm^3) and the organic layer separated, dried ($MgSO_4$) and then concentrated under reduced pressure to yield the crude product as a yellow oil. This was then purified by flash column chromatography (1:1 petrol–ether as eluent) to yield the title compound as a colourless oil (4.10 g, 70%), R_f (1:1 petrol–ether) 0.35; ν_{max}/cm^{-1} 2942, 2801, 1598, 1454, 1442, 1364, 1190, 1178, 1122, 1034, 904 and 622; δ($CDCl_3$; 250 MHz) 1.42–1.94 (m, 6 H, 3'-H_2, 4'-H_2, 5'-H_2), 2.38–2.58 (m, 4 H, 2-H_2, 5-H_2), 2.45 (s, 3 H, Ar-CH_3), 3.46–3.58 (m, 2 H, 6-H_2 or 6'-H_2), 3.75–3.98 (m, 2 H, 6-H_2 or 6'-H_2), 4.06 (t, 2 H, 1-H_2), 4.65 (t, J 4, 2'-H), 7.34 (d, J 6, 2 × Ar-H-*o*) and 7.80 (d, J 6, 2 × Ar-H-*m*) (Found: C, 61.3; H, 6.8. $C_{18}H_{24}O_5S$ requires C, 61.34; H, 6.86%).

1-*Iodo*-6-(*tetrahydropyran*-2'-*yloxy*)*hex*-3-*yne* **20**.—To a solution of the toluene-*p*-sulfonate **19** (2.80 g, 0.008 mol) in acetone (120 cm^3 AnalaR) was added sodium iodide (1.5 equiv., AnalaR) and the reaction mixture refluxed for 6 h. The reaction mixture was then taken up into dichloromethane (50 cm^3), and the organic layer washed with aqueous sodium thiosulfate (50 cm^3), water (50 cm^3) and then dried ($MgSO_4$), and concentrated under reduced pressure to yield the title compound as a colourless oil (2.23 g, 90%), R_f (9:1 petrol–ether) 0.23; ν_{max}/cm^{-1} 2937, 2869, 1436, 1353, 1249, 1122, 969, 907 and 814; δ($CDCl_3$; 250 MHz) 1.44–1.92 (m, 6 H, 3'-H_2, 4'-H_2, 5'-H_2), 2.40–2.54 (m, 2 H, 5-H_2), 2.68–2.82 (m, 2 H, 2-H_2), 3.20 (t, 2 H, J 7, 1-H_2), 3.46–3.52 (m, 2 H, 6-H_2 or 6'-H_2), 3.76–3.98 (m, 2 H, 6-H_2 or 6'-H_2) and 4.64 (t, J 4, 2'-H) (Found: C, 42.9; H, 5.5. $C_{11}H_{17}O_2I$ requires C, 42.87; H, 5.56%).

(6Z)-1-(*Tetrahydropyran*-2'-*yloxy*)*pentadec*-6-*en*-3-*yne* **22**.—To a solution of the iodide **20** (2.2 g, 0.0072 mol) in acetonitrile (80 cm^3, dry) was added triphenylphosphine (1.9 g, 0.0073 mol), and the resultant solution refluxed for 36 h. The reaction mixture was then cooled, and the acetonitrile removed under reduced pressure to yield a yellow oil. This was taken up into dichloromethane (80 cm^3), and 2,3-dihydropyran (1.0 equiv.) and Amberlyst-15 ion exchange resin (as catalyst) was added, and the mixture refluxed for 30 min. The catalyst was then filtered off and the filtrate concentrated under reduced pressure to yield a pale yellow oil, which was washed with ether (2 × 50 cm^3), and the ethereal washings decanted. The excess solvent was removed under reduced pressure to yield the phosphonium iodide as a sticky white solid (3.9 g, 96%). To a solution of this phosphonium iodide (3.9 g, 0.0069 mol) in 9:1 THF–hexamethylphosphoramide (HMPA) solution (50 cm^3) under nitrogen at −78 °C was added dropwise a solution of butyllithium (1.6 mol dm^{-3} in hexanes; 4.7 cm^3, 1.2 equiv.) and the resultant red solution stirred at −78 °C for 30 min and then at 0 °C for a further 10 min. The reaction was then cooled to −78 °C again and a solution of nonanal (1.0 g, 0.007 mol) in THF (15 cm^3, dry) added dropwise and the resultant solution stirred for 30 min at −78 °C and then at 0 °C for a further 90 min. The reaction was then quenched with water (50 cm^3), extracted with ether (2 × 100 cm^3), and the ethereal extracts were dried ($MgSO_4$) and concentrated under reduced pressure to yield a yellow oil. This was then purified by flash column chromatography (9:1 petrol–ether as eluent) to yield the title compound as a colourless oil (1.3 g, 62%), R_f (9:1 petrol–ether) 0.42; ν_{max}/cm^{-1} 2928, 2801, 1493, 1193, 1010 and 907; δ($CDCl_3$; 250 MHz) 0.87 (t, 3 H, J 7, alkyl-CH_3), 1.25 (s, 12 H, 6 × alkyl-CH_2), 1.42–1.85 (m, 6 H, 3'-H_2, 4'-H_2, 5'-H_2), 1.98–2.03 (m, 2 H, 8-H_2), 2.42–2.49 (m, 2 H, 2-H_2), 2.85–2.89 (m, 2 H, 5-H_2), 3.46–3.55 (m, 2 H, 1-H_2 or 6'-H_2), 3.75–3.90 (m, 2 H, 1-H_2 or 6'-H_2), 4.63 (t, 1 H, J 4, 2'-H) and 5.34–5.46 (m, 2 H, 6-H, 7-H) (Found: M^+, 292.24. $C_{19}H_{34}O_2$ requires M^+, 292.24).

(3Z,6Z)-1-(*Tetrahydropyran*-2'-*yloxy*)*pentadeca*-3,6-*diene* **23**.—To a solution of the acetylene **22** (1.3 g, 0.0043 mol) in THF (55 cm^3, dry) was added Lindlar catalyst (0.2 g) and the resultant solution transferred to a Parr apparatus and agitated under an atmosphere of hydrogen for 4 h. The hydrogen gas was then removed on a water pump, the reaction vessel flushed with

nitrogen, and the catalyst removed over Celite. The residual filtrate was then concentrated under reduced pressure to yield a yellow oil which was purified by flash column chromatography (9:1 petrol–ether as eluent) to give the title compound as a colourless oil (1.15 g, 88%); R_f (9:1 petrol–ether) 0.37; ν_{max}/cm^{-1} 3012, 2956, 2872, 1049, 1047 and 668; $\delta(CDCl_3$; 250 MHz) 0.88 (t, 3 H, J 7, alkyl-CH_3) 1.26 (s, 12 H, 6 × alkyl-CH_2), 1.48–1.92 (m, 6 H, 3′-H_2, 4′-H_2, 5′-H_2), 1.98–2.12 (m, 2 H, 8-H_2), 2.34–2.48 (m, 2 H, 2-H_2), 2.78–2.94 (m, 2 H, 5-H_2), 3.36–3.56 (m, 2 H, 1-H_2 or 6′-H_2), 3.68–3.96 (m, 2 H, 1-H_2, 6′-H_2), 4.63 (t, 1 H, J 4, 2′-H) and 5.26–5.52 (m, 4 H, 3-H, 4-H, 6-H, 7-H) (Found: M^+, 294.256. $C_{19}H_{36}O_2$ requires M^+, 294.256).

(3Z,6Z)-*Pentadeca*-3,6-*dien*-1-*ol* **24**.—A solution of diene **23** (1.1 g, 0.0036 mol) in 4:2:1 glacial acetic acid–THF–water was heated to 55 °C in a water bath for 4 h with stirring. The resultant solution was neutralised with saturated aqueous sodium hydrogen carbonate, and the resultant reaction mixture extracted with ether (2 × 100 cm^3). The ethereal layer was washed with water (100 cm^3), dried ($MgSO_4$) and concentrated under reduced pressure to yield a colourless oil, which was purified by flash column chromatography (1:1 petrol–ether as eluent) to give the title compound as a colourless oil (0.7 g, 86%), R_f (1:1 petrol–ether) 0.33; ν_{max}/cm^{-1} 3330 (OH), 3012, 2956, 2925, 2872, 1049, 1047, 668 and 649; $\delta(CDCl_3$; 250 MHz) 0.88 (t, 3 H, alkyl-CH_3), 1.25 (s, 12 H, 6 × alkyl-CH_2), 1.58 (br s, 1 H, OH), 2.00–2.16 (m, 2 H, 8-H_2), 2.36–2.40 (m, 2 H, 2-H_2), 2.78–2.84 (m, 2 H, 5-H_2), 3.64 (t, 2 H, J 7, 1-H_2) and 5.2–5.60 (m, 4 H, 3-H, 4-H, 6-H, 7-H).

(3Z,6Z)-1-*Iodopentadeca*-3,6-*diene* **25**.—To a solution of iodine (0.79 g, 0.0031 mol) and imidazole (0.22 g, 0.0032 mol) in dichloromethane (50 cm^3, dry) at 0 °C under nitrogen was added (in portions) triphenylphosphine (0.83 g, 0.0032 mol) and the resultant solution stirred for 10 min prior to the addition of the alcohol **24** (0.85 g, 0.0038 mol) and the resultant solution stirred for 4 h at room temperature. The reaction was then quenched with water (50 cm^3) and the organic layer separated, dried ($MgSO_4$), and then concentrated under reduced pressure to yield a yellow oil, which was taken up into dichloromethane (10 cm^3) and filtered over a silica pad, eluting with petrol, to yield the title compound as a pale yellow oil (0.74 g, 73%), R_f (9.5:0.5 petrol–ether) 0.57; ν_{max}/cm^{-1} 3009, 2922, 1460, 1241 and 968; $\delta(CDCl_3$; 250 MHz) 0.88 (t, 3 H, J 7, alkyl-CH_3), 1.28 (s, 12 H, 6 × alkyl-CH_2), 1.94–2.12 (m, 2 H, 8-H_2), 2.60–2.84 (m, 4 H, 2-H_2, 5-H_2), 3.14 (t, 2 H, J 7, 1-H_2) and 5.26–5.60 (m, 4 H, 3-H, 4-H, 6-H, 7-H) (Found: M^+, 334.156. $C_{15}H_{27}I$ requires M^+, 334.116).

Methyl (1R,2S)-2-[(1′Z,4′Z,7′Z)-*Hexadeca*-1′,4′,7′-*trienyl*]*cyclopropanecarboxylate* **26**.—To a solution of the iodide **25** (0.74 g, 0.0022 mol) in acetonitrile (50 cm^3, dry), was added triphenylphosphine (0.59 g, 0.0025 mol) and the resultant solution refluxed for 36 h. The reaction was then cooled to room temperature and the solvent removed under reduced pressure to yield a pale yellow oil, which was washed with ether (40 cm^3) and the excess solvent removed under reduced pressure to yield the phosphonium iodide as a pale yellow, viscous oil (1.30 g, 97%). This was then dissolved in THF (30 cm^3, dry), cooled to −78 °C under nitrogen, and to this was added butyllithium (1.6 mol dm^{-3} in hexane; 1.8 cm^3, 1.3 equiv.). The resultant red solution was stirred for 20 min at −78 °C and at 0 °C for a further 10 min. The reaction was then cooled to −78 °C again, and a solution of the aldehyde **10** (0.17 g, 0.001 33 mol) in THF (10 cm^3, dry) was added dropwise and the resultant solution stirred for 15 min at −78 °C and then allowed to warm to 0 °C over 90 min. The reaction was then quenched with water (50 cm^3), extracted with ether (2 × 50 cm^3), the combined organic extracts were then dried ($MgSO_4$), and concentrated under reduced pressure to yield a brown oil, which was purified by flash column chromatography (9:1 petrol–ether as eluent) to yield the title compound as a colourless oil (0.048 g, 12%), R_f (19:1 petrol–ether) 0.33; ν_{max}/cm^{-1} 3009, 2923, 1734 (C=O), 1649, 1384, 1199, 1172 and 679; $\delta(CDCl_3$; 250 MHz) 0.88 (t, 3 H, J 7, alkyl-CH_3), 1.18–1.46 (m, 14 H, 6 × alkyl-CH_2, 3-H_2), 1.88–2.18 (m, 4 H, 1-H, 2-H, 9′-H_2), 2.70–2.96 (m, 4 H, 3′-H_2, 6′-H_2), 3.68 (s, 3 H, OMe), 5.28–5.50 (m, 6 H, 1′-H, 2′-H, 4′-H, 5′-H, 7′-H, 8′-H) [Found: $(M + NH_4)^+$, 336.290. $C_{21}H_{38}O_2N$ requires $(M + NH_4)^+$, 336.290].

(1R,2S)-2-[(1′Z,4′Z,7′Z)-*Hexadeca*-1′,4′,7′-*trienyl*]*cyclopropanecarboxylic Acid* **5**.—To a solution of the methyl ester **26** (0.048 g, 0.000 15 mol) in THF (4 cm^3, dry) was added LiOH (2 mol dm^{-3}; approximately 2 equiv.) and the resultant solution stirred at room temperature for 5 days. The reaction mixture was then neutralised with glacial acetic acid and the solution extracted with ether (2 × 25 cm^3). The organic layer was washed with water (1 × 20 cm^3), dried ($MgSO_4$) and concentrated under reduced pressure to yield a colourless oil, which was purified by flash column chromatography (1:1 petrol–ether as eluent) to give the title compound as a colourless oil (22 mg, 48%), R_f (1:1 petrol–ether) 0.44; ν_{max}/cm^{-1} 3001, 1700 (C=O), 1411 and 973; $\delta(CDCl_3$; 250 MHz) 0.88 (t, 3 H, J 7, alkyl-CH_3), 1.20–1.41 (m, 14 H, 6 × alkyl-CH_2, 3-H_2), 1.90–2.26 (m, 5 H, 1-H, 2-H, 9′-H_2), 2.78–2.99 (m, 4 H, 3′-H_2, 6′-H_2), 5.24–5.58 (m, 6 H, 1′-H, 2′-H, 4′-H, 5′-H, 7′-H, 8′-H) and 11.04 (br s, 1 H, CO_2H) [Found: $(M + NH_4)^+$, 322.275. $C_{20}H_{32}O_2$ requires $(M + NH_4)^+$, 322.275].

(5S,15S)-5,15- *And* (8S,15S)-8,15-*Dihydroxyicosatetraenoic Acids* (*DIHETE*): *Standard Assay*.—To a solution of arachidonic acid (10 mg, 3.28 × 10^{-5} mol) in phosphate buffer (0.2 mol dm^{-3}; 130 cm^3, pH 6.8) was added soybean lipoxygenase (4 mg) and the progress of the reaction monitored by UV analysis at 270 nm. After 60 min the rate of increase of absorbance at 270 nm had stopped and the reaction mixture was acidified to pH 3 with HCl (1 mol dm^{-3}). The reaction was then extracted with ether (2 × 50 cm^3), the ethereal layers combined and treated with sodium borohydride (2 mg, 5.29 × 10^{-5} mol). The reaction mixture was then washed with water (2 × 50 cm^3), dried ($MgSO_4$) and concentrated under reduced pressure. The residue was then taken up into methanol (1 cm^3, HyperSolv grade) and stored under argon at −78 °C. HPLC analysis eluting with 80:20:0.1 methanol–water–acetic acid revealed two component peaks, which corresponded to authentic samples of 5,15-DIHETE (R_T 15.4 min) and 8,15-DIHETE (R_T 17.1 min) respectively. UV analysis revealed two λ_{max} peaks corresponding to the conjugated diene (240 nm) and the conjugated triene (270 nm) respectively.

Acknowledgements

We thank the SERC and Eli Lilly and Company for a CASE award for P. I. B.

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Paper 3/07048D
Received 29*th November* 1993
Accepted 4*th February* 1994