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Synthesis of (2R; 4R,S)- and (2S; 4R,S)-dimethyldocosanoic acids

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Abstract

2,4-Dimethyldocosanoic acid is a major acyl component of 2,3-di-*O*-acyl-α,α-trehalose glycolipid antigens isolated from *Mycobacterium tuberculosis*. Racemic 2,4-dimethyldocosanoic acid has been synthesised and the chiral centre at C-2 resolved as an (R)-(−)-naphthylethylamide. An isomeric mixture of 3,5-dimethylcyclohexan-1-ols was oxidised to isomeric 3,5-dimethylcyclohexan-1-ones. This was subjected to Baeyer-Villiger oxidation to give isomeric 3,5-dimethylcaprolactones. Ring opening under alkaline conditions followed by phase-transfer catalysed esterification gave isomeric methyl 3,5-dimethyl-6-hydroxyhexanoates. Protection of the alcohol with triphenylmethyl chloride followed by lithium aluminium hydride reduction and pyridinium chlorochromate oxidation gave isomeric 3,5-dimethyl-6-triphenylmethyloxyhexanals. Coupling with hexadecyltriphenylphosphonium bromide, followed by trityl deprotection and hydrogenation of the remaining alkene, yielded isomeric 2,4-dimethyldocosanols. Oxidation with pyridinium dichromate in *N,N'*-dimethylformamide produced isomeric 2,4-dimethyldocosanoic acids, which were resolved at C-2 as diastereoisomeric (R)-(−)-naphthylethylamides.

Key words: (2R; 4R,S)- and (2S; 4R,S)-Dimethyldocosanoic acid; Mycosanoic acid; *Mycobacterium tuberculosis*; 2,3-Di-*O*-acyl-α,α-trehalose; (R)-(−)-Naphthylethylamides

1. Introduction

Attention is again turning to the problem of tuberculosis as, coupled with AIDS, this is becoming a resurgent disease and new drug-resistant strains are also developing [1–3]. These multi-drug-resistant strains are increasingly difficult to detect, and treatment is an obvious problem. Rapid detection methods are required to identify the presence of tuberculosis in the patients as quickly as possible, to facilitate effective control of the disease.

Mycobacterium tuberculosis produces several lipid antigens in its cell envelope [4]. Polar acyl trehaloses [5–7] are a series of characteristic relatively simple glycolipid antigens, initially identified by Minnikin et al. [8]. These lipids have been characterised fully as 2,3-di-*O*-acyl-α,α-trehaloses by Besra et al. [9] (Fig. 1). The synthesis of these 2,3-di-*O*-acyl-α,α-trehaloses could provide bulk quantities of reproducible, pure lipids for use in the rapid serodiagnosis of tuberculosis.

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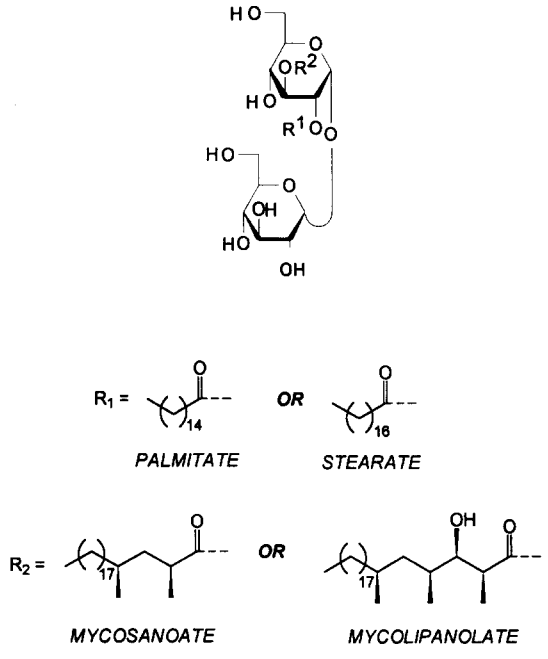


Fig. 1. 2,3-Di-*O*-acyl-α,α-trehaloses from *Mycobacterium tuberculosis.*

Diacyl trehaloses (DATs) from *M. tuberculosis* can be degraded to give a complex series of fatty acids. One major degradation component is the dimethylbranched mycosanoic acid (Fig. 2). A synthetic standard would be desirable for gas chromatographic (GC) analysis of such lipid antigen degradation products. A sample of mycosanoic acid is required to complete the synthesis of 2,3-di-*O*-acyl-α,α-trehaloses [10,11]. Such acids would also serve as intermediates in the synthesis

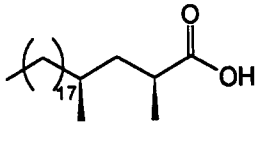


Fig. 2. Naturally ocurring (2S, 4S)-dimethyldocosanoic (mycosanoic) acid.

of the mycolipanolic acid components of the diacyl trehaloses (Fig. 1).

It is not known if the chirality of the fatty acyl chains of the diacyl trehaloses affects the antigenicity of these compounds. There is a need, therefore, for both racemic and optically pure fatty acids, which can be used to synthesise 2,3-di-*O*-acyl-α,α-trehaloses. In the present study, racemic 2,4-dimethyldocosanoic acid has been synthesised and partially resolved into its (2R; 4R,S)- and (2S; 4R,S)-isomers.

2. Experimental procedures

Solvents were dried according to standard literature procedures. Melting points (uncorrected) were recorded using a Kofler hot stage apparatus. Elemental analyses of crystalline solids were obtained using a Carlo-Erba Instrumentazione model 1106 CHN analyser. Infra-red spectra (cm^{-1}) were recorded on a Nicolet 20 SXB or a Nicolet 20 PC Fourier-Transform spectrometer; peaks are labelled 'br' (broad), 's' (strong), 'm' (medium) and 'w' (weak). Proton (^1H) and carbon (^{13}C) nuclear magnetic resonance (NMR) spectra (δ values/ppm) were obtained using solutions in deuteriochloroform on a Bruker WP 200 instrument; peaks are labelled 's' (singlet), 'd' (doublet), 't' (triplet), 'q' (quartet), 'quin' (quintet), 'sext' (sextet), 'm' (multiplet) and 'cm' (complex multiplet). ^{13}C-NMR spectra were assigned as far as possible.

Electron-impact and fast-atom bombardment mass spectra were recorded on AEI MS9 and Kratos MS 80RF spectrometers. Optical rotations were recorded on an NPL automatic polarimeter type 143D or on a PolAAr 2001 automatic polarimeter. Starting materials and chemical reagents were purchased from Aldrich, Fisons, Fluka and Lancaster Syntheses. Column chromatography was carried out at medium pressure using Merck 7736 grade silica gel. Fluka 60738 silica gel 60 or Fisons Matrex Silica 60 were used for flash column chromatography. Thin-layer chromatography (TLC) used Merck 5554 pre-coated silica-gel aluminium-backed sheets; all compounds were revealed by spraying with a 10% solution of molybdophosphoric acid (MPA) in ethanol

followed by heating at 180°C for 15 min. A 15% solution of ammonium molybdate in ethanol, followed by heating at 180°C for 15 min, was used for groups containing acid functionalities. Preparative thin-layer chromatography used Merck 5735 (Kieselgel 60 F_{254}) pre-coated silica-gel plastic-backed sheets; all compounds were revealed using 254-nm UV light. Reverse-phase high-performance thin-layer chromatography (HPTLC) used Merck 13-24 (RP-18 F_{254} s) pre-coated silica-gel glass-backed plates; all compounds were revealed using 254-nm UV light. Solvent system 1: petroleum ether (b.p. 40–60°C) : ethyl acetate (9:1). Solvent system 2: toluene : acetone (99:1). Solvent system 3: petroleum ether (b.p. 40–60°C) : ethyl acetate (5:1). Gas chromatography programme A: Pye Unicam Pye series 104 chromatograph containing a non-polar OV-17 column, 8 feet in length and 0.25 inches o.d. (3% OV-17 on Chromosorb). A temperature gradient of 80–280°C was used at 8°C per minute. Gas chromatography programme B: Packard model 427 chromatograph containing a BP1 capillary column, 25 m in length, i.d. 0.25 mm and with a film coating of 1 μm of methyl silicone BP1. A temperature gradient of 140–280°C was used at 10°C per minute.

2.1. Isomeric 3,5-dimethylcyclohexanones (2)

Sodium dichromate dihydrate (87.025 g, 292.0 mmol, 1.5 eq.) was dissolved in distilled water (200 cm^3) and concentrated sulphuric acid (50 cm^3) was added. The solution was further diluted with water (200 cm^3) and then cooled to 0°C. 3,5-Dimethylcyclohexanol (**1**) (25.031 g, 195.0 mmol, 1.0 eq.) was mixed with glacial acetic acid (50 cm^3) and carefully added to the dichromate solution. The resulting solution was then left to stir for 2 h, with warming to room temperature. Ice-water (200 cm^3) was then added and the product extracted with diethyl ether (4 × 250 cm^3). The combined ethereal layers were washed with saturated aqueous sodium chloride solution (2 × 250 cm^3), dried over anhydrous magnesium sulphate, filtered and concentrated in vacuo to yield a golden oil. Short-path distillation of this yielded the product (**2**) as a colourless oil (16.749 g, 67%). $R_f = 0.63$ in TLC solvent system 1; b.p. 39°C at 0.17 mm Hg; lit. b.p. 174–176°C at 760 mm Hg; IR (film): 1716.76 (s). Found M^+ 126.1053, $C_8H_{14}O$ requires 126.1980; ^{1}H-NMR (200 MHz) 0.93 [d, $J = 6.8$, (CH_3)-CH-, axial], 0.97 [d, $J = 5.9$, (CH_3)-CH-, equatorial], 1.55 [cm, $J = 5.7$, -CH-(CH_3)-, -CH(CH_3)-CH_2-CH(CH_3)-], 2.31 (m, br, -CH_2-CO-CH_2-), no integrals listed due to a mixture of *cis/trans* isomers; ^{13}C-NMR (50.3 MHz) 20.84 [(CH_3)-CH-, axial], 22.35 [(CH_3)-CH-, equatorial], 29.63 [(CH_3)-CH-, axial], 33.22 [(CH_3)-CH-, equatorial], 39.48 [-CH(CH_3)-CH_2-C′H(CH_3)-, equatorial-axial′ and axial-equatorial′], 42.60 [-CH(CH_3)-CH_2-CH′(CH_3)-, equatorial-equatorial′], 48.72 (-CH-CH_2-CO-CH_2-C′H-, equatorial-axial′; axial-equatorial′), 49.25 (-CH-CH_2-CO-CH_2-C′H-, equatorial-equatorial′), 176.60 (-CO-).

2.2. Isomeric 3,5-dimethylcaprolactones (3)

Isomeric 3,5-dimethylcyclohexanones (**2**) (3.549 g, 28.2 mmol, 1.0 eq.) were dissolved in dichloromethane (50 cm^3), and a catalytic quantity of *para*-toluenesulphonic acid (100 mg) was added. *meta*-Chloroperbenzoic acid (80%, 7.187 g, 41.8 mmol, 1.2 eq.) was added to the solution, which was left to stir for 16 h at room temperature. Solid sodium hydrogen carbonate was added and the resulting mixture stirred for 1 h at room temperature. The dense white precipitate of *meta*-chlorobenzoic acid was removed by filtration at reduced pressure and the solvent removed from the filtrate in vacuo. This yielded a crude oil that was purified by Kugelrohr distillation to afford **3** as a sweet-smelling, colourless oil (2.104 g, 53%). $R_f = 0.65$ in TLC solvent system 1; IR (film): 1732.30 (s), 1703.36 (s). Found M^+ 143.0618, $C_8H_{14}O_2$ requires 142.1974; ^{1}H-NMR (200 MHz) 0.84 [d, 3H, $J = 6.9$, (CH_3)-CH-CH_2-O-], 0.96 [d, 3H, $J = 6.8$, (CH_3)-CH-CH_2-CO-], 1.52–2.10 [m, 4H, -CH(CH_3)-CH_2-CH(CH_3)-], 2.42 (m, 2H, -CH_2-CO-O-), 3.93 (m, 2H, -CH_2-O-CO-).

2.3. Isomeric 3,5-dimethyl-6-hydroxyhexanoic acids (4)

Isomeric 3,5-dimethylcyclohexanones (**2**) (15.750 g, 125.0 mmol, 1.0 eq.) were dissolved in

dichloromethane (300 cm^3), and a catalytic quantity of *para*-toluenesulphonic acid (200 mg) was added. *meta*-Chloroperbenzoic acid (50–60%, 46.721 g, 150.0 mmol, 1.2 eq.) was added portionwise and the solution left to stir for 16 h at room temperature. The dense white precipitate of *meta*-chlorobenzoic acid was removed by filtration under reduced pressure and most of the solvent removed from the filtrate in vacuo. Dichloromethane (100 cm^3) was added, and 30% aqueous sodium hydroxide solution (350 cm^3) was then carefully added. This solution was rapidly stirred and left at room temperature for 16 h. The mixture was then carefully acidified to pH 1 by the dropwise addition of concentrated sulphuric acid. The aqueous solution was then extracted using dichloromethane (4 × 200 cm^3). The dichloromethane layers were dried over magnesium sulphate, filtered and concentrated in vacuo to yield a thick syrup. Purification by flash column chromatography using petroleum ether (b.p. 40–60°C) and ethyl acetate (9:1 to 5:1 to ethyl acetate gradient) gave **4** as a thick, colourless syrup (13.501 g, 86%). $R_f = 0.07$ in TLC solvent system 1; IR (film): 3387.43 (m, br),1707.22 (s). Found M^+ 160.1175, $C_8H_{16}O_3$ requires 160.2126; ^{1}H-NMR (200 MHz) 0.90 [d, 3H, $J = 6.6$, -(C*H*$_3$)CH-CH_2-COOH], 0.95 [d, 3H, $J = 6.4$, -(C*H*$_3$)CH-CH_2OH], 1.36 [quin, 2H, $J = 6.7$, -C*H*$_2$-CH-(CH_3)-CH_2-COOH], 1.67 [sext, 1H, $J = 6.6$, -C*H*(CH_3)-CH_2-COOH], 2.10 [m, 1H, -C*H*(CH_3)-CH_2OH], 2.31 ('q', 2H, $J = 5.9$, -C*H*$_2$-COOH), 3.46 [d, 2H, $J = 5.2$, -(CH_3)CH-C*H*$_2$OH], 7.35 (s, br, 1H, -CH_2O*H*); ^{13}C-NMR (50.3 MHz) 16.17, 17.42 [-(*C*H_3)CH-CH_2OH], 19.37, 20.49 [-(*C*H_3)CH-CH_2-COOH], 27.46, 27.57 [-(CH_3)*C*H-CH_2-COOH], 32.89 [-(CH_3)CH-*C*H_2-CH(CH_3)-], 40.10, 40.40 [-(CH_3)*C*H-CH_2OH], 41.31, 42.41 (-*C*H_2-COOH), 67.55, 68.50, (-*C*H_2OH), 178.69 (-*C*OOH).

2.4. *Isomeric methyl 3,5-dimethyl-6-hydroxyhexanoates (5)*

Isomeric 3,5-dimethyl-6-hydroxyhexanoic acids (**4**) (14.405 g, 76.0 mmol, 1.0 eq.) were dissolved in 15% aqueous tetrabutylammonium hydroxide (100 cm^3) and stirred at room temperature for 30 min. Dichloromethane (100 cm^3) was then added and the mixture stirred rapidly to ensure efficient mixing of the two phases. Iodomethane (11.800 g, 5.20 cm^3, 89.4 mmol, 1.2 eq.) was added and the mixture left to stir for 24 h at room temperature. The mixture was poured into water (100 cm^3) and the products extracted with dichloromethane (3 × 150 cm^3). The combined organic layers were washed with saturated aqueous sodium chloride solution (300 cm^3), dried over anhydrous magnesium sulphate, filtered and concentrated in vacuo. The crude product was purified by flash column chromatography using petroleum ether (b.p. 40–60°C) and ethyl acetate (9:1 to 5:1 to 2:1 gradient) to yield **5** as a colourless oil (10.789 g, 82%). $R_f = 0.11$ in TLC solvent system 1; IR (film): 3410.58 (w, br), 1718.79 (s); Found: MH^+ 175.1325, $C_9H_{18}O_3$ requires 174.2394, $C_9H_{19}O_3$ requires M 175.2473; ^{1}H-NMR (200 MHz) 0.89 [d, 3H, $J = 6.5$, -(C*H*$_3$)CH-CH_2-CO-O-CH_3], 0.92 [d, 3H, $J = 6.3$, -(C*H*$_3$)CH-CH_2OH], 1.33 [quin, 2H, $J = 6.7$, -C*H*$_2$-(CH_3)CH-CH_2-CO-O-CH_3], 1.65 [sext, 1H, $J = 6.5$, -C*H*(CH_3)-CH_2-CO-O-CH_3], 2.09 [m, 1H, -C*H*(CH_3)-CH_2OH], 2.30 ('q', 2H, $J = 5.8$, -C*H*$_2$-CO-O-CH_3), 3.45 (d, 2H, $J = 5.4$, -C*H*$_2$OH), 3.57 (s, 3H, -CO-O-C*H*$_3$), 7.40 (s, br, 1H, -CH_2O*H*); ^{13}C-NMR (50.3 MHz) 16.19, 17.44 [-(*C*H_3)CH-CH_2OH], 19.37, 20.51 [-(*C*H_3)CH-CH_2-CO-O-CH_3], 27.57, 27.68 [-(CH_3)*C*H-CH_2-CO-O-CH_3], 32.97 [-(CH_3)CH-*C*H_2-CH(CH_3)-], 40.20, 40.49 [-(CH_3)*C*H-CH_2OH], 41.28, 42.49 (-*C*H_2-CO-O-CH_3), 67.60, 68.37 (-*C*H_2-OH), 86.27 (-CO-O-*C*H_3), 173.81 (-*C*O-O-CH_3).

2.5. *Isomeric methyl 3,5-dimethyl-6-triphenylmethyloxyhexanoates (6)*

Isomeric methyl 3,5-dimethyl-6-hydroxyhexanoates (**5**) (10.789 g, 62.0 mmol, 1.0 eq.) were dissolved in dry dichloromethane (150 cm^3) under nitrogen. The solution was cooled to 0°C before the dropwise addition of dry pyridine (5.908 g, 6.04 cm^3, 74.70 mmol, 1.2 eq.). The resulting solution was stirred for 30 min at 0°C before the addition of triphenylmethyl chloride (20.669 g, 74.0 mmol, 1.2 eq.). The solution was allowed to warm to room temperature, and stirring was continued for a further 16 h. The dichloromethane solvent

was removed in vacuo and the gummy residue resuspended in diethyl ether (200 cm^3). This was poured onto saturated aqueous ammonium chloride solution (250 cm^3) and the products extracted with diethyl ether (3 × 150 cm^3). The ethereal layers were combined and washed with saturated aqueous sodium chloride solution, dried over anhydrous magnesium sulphate, filtered and concentrated in vacuo. Purification by flash column chromatography using petroleum ether (b.p. 40–60°C) and ethyl acetate (95:5 to 9:1 gradient) gave **6** as a thick syrup (22.525 g, 87%). R_f = 0.63 in TLC solvent system 1; IR (film): 1736.16 (s). Found M^+ 416.2274, $C_{28}H_{32}O_3$ requires 416.5590; ^{1}H-NMR (200 MHz) 0.87 [d, 3H, J = 6.3, -(CH_3)CH-CH_2-CO-O-CH_3], 0.95 [d, 3H, J = 6.6, -(CH_3)CH-CH_2-OTr], 1.34 [cm, 2H, J = 6.7, -CH_2-CH(CH_3)-CH_2-CO-O-CH_3], 1.76 [sext, 1H, J = 6.6, -CH(CH_3)-CH_2-CO-O-CH_3], 2.07 [m, 1H, -CH(CH_3)-CH_2OTr], 2.29 ('q', 2H, J = 9.3, -CH_2-CO-O-CH_3), 2.90 (cm, 2H, -CH_2OTr), 3.62 (s, 3H, -CO-O-CH_3); ^{13}C-NMR (50.3 MHz) 16.94, 18.12 [-(CH_3)CH-CH_2-OTr], 19.23, 20.47 [-(CH_3)CH-CH_2-CO-O-CH_3], 27.82, 27.93 [-(CH_3)CH-CH_2-CO-O-CH_3], 30.99, 31.46 [-(CH_3)-CH-CH_2-CH(CH_3)-], 40.87, 41.27 [(CH_3)CH-CH_2-OTr], 41.53, 42.62 [-CH_2-CO-O-CH_3], 68.25, 68.95 (-CH_2-OTr), 86.22 (-CO-O-CH_3), 126.87, 127.31, 127.72, 127.98, 128.79 (trityl), 144.54 (Ph_3C-O-), 173.73 (-C-O-CH_3).

2.6. *Isomeric 3,5-dimethyl-6-triphenylmethyloxyhexan-1-ols (7)*

Isomeric methyl 3,5-dimethyl-6-triphenylmethyloxyhexanoates (**6**) (22.525 g, 49.0 mmol, 1.0 eq.) were dissolved in dry tetrahydrofuran (300 cm^3) under nitrogen and cooled to 0°C. Lithium aluminium hydride (2.611 g, 68.8 mmol, 1.7 eq.) was added in portions to the solution and the mixture allowed to warm steadily to room temperature over a 2 h period. The reaction was quenched by the careful addition of saturated aqueous ammonium chloride solution (200 cm^3). The resulting suspension was allowed to stir for a further 1 h before the products were extracted using diethyl ether (3 × 150 cm^3). The ethereal layers were combined, washed with saturated aqueous sodium chloride solution (250 cm^3) and dried over anhydrous magnesium sulphate.. The organic layer was filtered and concentrated in vacuo to give a crude, thick syrup. Purification by flash column chromatography using petroleum ether (b.p. 40–60°C) and ethyl acetate (9:1 to 5:1 gradient) gave **7** as a colourless syrup (8.530 g, 41%). R_f = 0.18 in TLC solvent system 1; IR (film): 3385.50 (m, br). Found M^+ 388.2401, $C_{27}H_{32}O_2$ requires 388.5486; ^{1}H-NMR (200 MHz) 0.82 [d, 3H, J = 6.3, -(CH_3)CH-CH_2-CH_2OH], 0.97 [d, 3H, J = 6.7, -(CH_3)CH-CH_2-OTr], 1.33 [quin, 2H, J = 6.6, -CH_2-CH(CH_3)-CH_2-CH_2OH], 1.40–1.58 [cm, 4H, -CH_2-CH(CH_3)-CH_2OTr, -CH_2-CH_2-OH], 1.82 [sext, 1H, J = 5.7, -(CH_3)CH-CH_2-CH_2OH], 2.00 [m, 1H, -(CH_3)CH-CH_2OTr], 2.94 (cm, 2H, -CH_2-OTr), 3.60 (cm, -CH_2OH), 7.16–7.36 [9H, m, C(3)H, C(4)H, C(5)H of trityl], 7.40–7.46 [6H, m, C(2)H, C(6)H of trityl]; ^{13}C-NMR (50.3 MHz) 17.35, 17.75 [-(CH_3)CH-CH_2-OTr], 18.50, 20.62 [-(CH_3)CH-CH_2-CH_2OH], 26.70, 26.94 [-(CH_3)CH-CH_2-CH_2OH)], 30.99, 31.46 [-CH_2-(CH_3)CH-CH_2-OTr], 40.70 [-(CH_3)CH-CH_2-OTr], 41.45, 41.86 [-CH_2-CH_2OH], 61.86 [-CH_2-OH], 68.29, 69.02 [-CH_2-OTr], 126.91, 127.77, 128.05, 128.86 (trityl), 144.62 (Ph_3C-O-).

2.7. *Isomeric 3,5-dimethyl-6-triphenylmethyloxyhexanals (8)* [12]

Isomeric 3,5-dimethyl-6-triphenylmethyloxyhexan-1-ols (**7**) (8.313 g, 19.40 mmol, 1.0 eq.) were dissolved in dry dichloromethane (180 cm^3) under nitrogen. Activated 4 Å molecular sieves were added and the mixture stirred for 30 min at room temperature before the addition of pyridinium chlorochromate (PCC) (9.517 g, 44.0 mmol, 2.3 eq.). The resulting solution was stirred for 2 h at room temperature. The solvent was removed in vacuo and the tacky, brown precipitate resuspended in diethyl ether (150 cm^3). The suspension was filtered through a silica gel pad which was rinsed with further aliquots of diethyl ether (3 × 100 cm^3). The solvent was evaporated from the filtrate in vacuo and the crude syrup purified by flash column chromatography using petroleum ether (b.p. 40–60°C) and ethyl acetate (95:5 to 9:1 gradient) to give **8** as a colourless, thick syrup (4.859 g,

59%). $R_f = 0.44$ in TLC solvent system 1; IR (film): 3420.23 (w, br), 1724.58 (s). Found M^+ 386.2306, $C_{27}H_{30}O_2$ requires 386.5328; ^{1}H-NMR (200 MHz) 0.81 [d, 3H, $J = 6.4$, -(CH_3)CH-CH$_2$-CHO], 0.90 [d, 3H, $J = 6.7$, -(CH_3)CH-CH$_2$-OTr], 1.31 [quin, 2H, $J = 6.8$, -CH_2-CH(CH$_3$)-CH$_2$-CHO], 1.69 [sext, 1H, $J = 6.5$, -(CH$_3$)CH-CH$_2$-CHO], 1.81 [sext, 1H, $J = 5.6$, -(CH$_3$)CH-CH$_2$-OTr], 2.06 (dd, 2H, $J = 2.8$, 8.2, -CH_2-CHO), 2.96 (cm, 2H, -CH_2-OTr), 7.08–7.40 [m, 9H, C(3)H, C(4)H, C(5)H of trityl], 7.50–7.76 [m, 6H, C(2)H, C(6)H of trityl], 9.58 (t, 1H, $J = 2.8$, -CHO); ^{13}C-NMR (50.3 MHz) 17.27, 18.19 [-(CH_3)CH-CH$_2$-OTr], 19.74, 20.65 [-(CH_3)CH-CH$_2$-CHO], 25.47, 25.76 [-(CH$_3$)CH-CH$_2$-CHO], 30.98, 31.46 [-(CH$_3$)CH-CH_2-CH(CH$_3$)-], 40.47 [-(CH$_3$)CH-CH$_2$-OTr], 41.16, 41.48 (-CH_2-CHO), 68.07, 68.73 (-CH_2-OTr), 126.95, 127.05, 127.31, 127.80, 128.00, 128.76 (trityl), 144.48 (Ph$_3$C-O-), 203.10 (-CHO).

2.8. *1-Hexadecyltriphenylphosphonium bromide (**24**)*

Triphenylphosphine (28.821 g, 0.11 mol, 1.0 eq.) and 1-bromohexadecane (33.668 g, 0.11 mol, 1.0 eq.) were admitted to a dry flask fitted with a reflux condenser and drying tube. Dry toluene (300 cm^3) was added to dissolve the solids, and the mixture was heated to reflux for 24 h. The toluene was removed in vacuo and the resulting syrup allowed to cool. Diethyl ether (400 cm^3) was added, causing precipitation of the product. This was filtered under reduced pressure and washed with further diethyl ether (3 × 100 cm^3).The product was then dried further using high-vacuum apparatus to yield **24** as a white solid (56.820 g, 91%). M.p. 102–104°C. Found M^+ 567.7928, $C_{34}H_{48}PBr$ requires 567.6310; ^{1}H-NMR (200 MHz) 0.82 (t, 3H, $J = 6.3$, CH_3-CH$_2$-CH$_2$-), 1.16 [s, -(CH_2)$_n$-], 1.54 (t, 2H, $J = 6.2$, -CH_2-CH$_2$-PPh$_3$-), 3.63 (m, 2H, -CH_2-PPh$_3$-); ^{13}C-NMR (50.3 MHz) 14.15 (CH_3-CH$_2$-CH$_2$-CH$_2$-), 22.68 (CH$_3$-CH_2-CH$_2$-CH$_2$-), 29.22, 29.37, 29.67, [-(CH_2)$_n$-], 31.90 CH$_3$-CH$_2$-CH_2-CH$_2$-), 117.46 (-CH_2-PPh$_3$-), 119.15 [C(1), C(1′), C(1″) of PPh$_3$], 130.44, 130.69 [C(3), C(3′), C(3″), C(5), C(5′), C(5″) of PPh$_3$], 133.56, 133.75 [C(2), C(2′), C(2″), C(6), C(6′), C(6″) of PPh$_3$], 135.09 [C(4), C(4′), C(4″) of PPh$_3$].

2.9. *Isomeric (6E,Z)2,4-dimethyl-1-triphenylmethyloxydocos-6-enes (**9a**, **9b**)*

1-Hexadecyltriphenylphosphonium bromide (**24**) (18.068 g, 31.0 mmol, 3.0 eq.) was suspended in dry tetrahydrofuran (150 cm^3) under an atmosphere of nitrogen. *n*-Butyl lithium (2.5 M in hexanes) (13.4 cm^3, 33.5 mmol, 3.3 eq.) was added dropwise at room temperature and the resulting red solution allowed to stir for 1 h at room temperature. 3,5-Dimethyl-6-triphenylmethyloxyhexanals (**8**) (3.906 g, 10.0 mmol, 1.0 eq.) were dissolved in dry tetrahydrofuran (50 cm^3) and this solution slowly added to the Wittig reagent, prepared above. The solution was stirred for 3 h at room temperature before quenching with acetone (50 cm^3). The solvent was removed in vacuo and the gummy residue resuspended in diethyl ether (250 cm^3). This was washed through a silica gel pad and rinsed with further aliquots of diethyl ether (3 × 150 cm^3). The solvent was removed from the filtrate in vacuo, to give a crude syrup. Purification of this using flash column chromatography with petroleum ether (b.p. 40–60°C) and ethyl acetate (95:5 to 9:1 gradient) gave a mixture of **9a** and **9b** as a colourless oil (4.042 g, 68%). Compounds **9a** and **9b** showed $R_f = 0.72$ and $R_f = 0.69$, respectively, in TLC solvent system 1; IR (film): 1647.42 (s). Found M^+ 539.6126, $C_{43}H_{61}O$ requires 539.9543; ^{1}H-NMR (200 MHz) 0.88 [m, 6H, -(CH_3)CH-CH$_2$-CH=CH-, CH_3-CH$_2$-CH$_2$-], 0.97 [d, 3H, $J = 6.7$, -(CH_3)CH-CH$_2$-OTr], 1.24 [s, 34H, -(CH_2)$_n$-, -CH_2-CH(CH$_3$)-CH$_2$-CH=CH-], 1.61 [m, 1H, -CH(CH$_3$)-CH$_2$-CH=CH-], 2.09 [m, 1H, -CH(CH$_3$)-CH$_2$OTr], 2.41 [dt, 4H, -CH_2-CH=CH-CH_2-], 2.83 (cm, 2H, -CH_2-OTr), 5.35 (2H, m, $J = 5.6$, -CH=CH-CH$_2$-); ^{13}C-NMR (50.3 MHz) 14.22 (CH_3-CH$_2$-CH$_2$-CH$_2$-), 17.23, 18.48 [-CH(CH_3)-CH$_2$-CH=CH-CH$_2$-], 20.25, 20.66 [-(CH_3)CH-CH$_2$-OTr], 22.78 (CH$_3$-CH_2-CH$_2$-CH$_2$-), 27.45 [-CH_2-CH=CH-CH$_2$-(CH$_3$)-], 29.47, 29.75 [-(CH_2)$_n$-], 30.69 [-CH=CH-CH$_2$-CH(CH$_3$)-CH_2-], 31.53 [-CH$_2$-

$CH(CH_3)$-CH_2-], 32.01 (CH_3-CH_2-CH_2-CH_2-), 34.29 [-CH_2-CH=CH-CH_2-$CH(CH_3)$-], 37.26, 38.24 [-CH=CH-CH_2-$CH(CH_3)$-], 40.8, 41.31 [-$(CH_3)CH$-CH_2-OTr], 68.39 (-CH_2-OTr), 126.86, 127.73 (trityl), 128.52 [olefinic, C(6)], 128.64, 128.79 (trityl), 130.88 [olefinic, C(7)], 144.63 (Ph_3C-O-).

2.10. Isomeric (6E,Z)2,4-dimethyldocos-6-en-1-ols (10a, 10b)

The isomers of (6E,Z)2,4-dimethyl-1-triphenylmethyloxydocos-6-ene (**9a**, **9b**) (4.042 g, 6.81 mmol, 1.0 eq.) were dissolved in dichloromethane (60 cm^3). Trifluoroacetic acid (5 cm^3) was carefully added and the solution stirred rapidly for 1 min before the addition of water (50 cm^3). The excess acid was then neutralised by the addition of solid sodium hydrogen carbonate. The biphasic mixture was poured into water (100 cm^3) and extracted with dichloromethane (3 × 75 cm^3). The organic layers were combined, dried over anhydrous magnesium sulphate, filtered and concentrated in vacuo to yield a white solid/oil mixture. Precipitation of the trityl alcohol from cold petroleum ether (b.p. 40–60°C) and subsequent removal from the mixture by filtration gave a colourless solution that was concentrated in vacuo to yield a viscous oil. Flash column chromatography using petroleum ether (b.p. 40–60°C) and ethyl acetate (9:1 to 7:1 gradient) gave **10a** and **10b** (mixture) as a viscous oil (2.062 g, 86%).

Compounds **10a** and **10b** both showed $R_f = 0.34$ in solvent system 1; IR(film): 3346.92 (m, br), 1653.21 (m). Found M^+ $C_{24}H_{52}O$ requires 356.6742; ^{1}H-NMR (200 MHz) 0.88 [cm, 9H, -(CH_3)CH-CH_2-$CH(CH_3)$-, CH_3-CH_2-CH_2-CH_2-], 1.23 [s, 34H, -$(CH_2)_n$-], 1.49 [m, 1H, -$(CH_3)CH$-CH_2], 1.53–1.87 [cm, 3H, -$(CH_3)CH$-CH_2-$CH(CH_3)$-CH=CH-], 3.41 [m, 2H, -CH_2-$CH(CH_3)$-CH_2OH], 5.35 (m, 2H, $J = 5.7$, -CH=CH-CH_2-); ^{13}C-NMR (50.3 MHz) 14.15 (CH_3-CH_2-CH_2-CH_2-), 16.38, 17.34 [-$CH(CH_3)$-CH_2OH], 20.33, 21.06 [-$CH(CH_3)$-CH_2-], 22.74 (CH_3-CH_2-CH_2-CH_2-), 27.41 [-CH_2-CH=CH-CH_2-(CH_3)-], 29.40, 29.63, 29.73, [-$(CH_2)_n$-], 30.76 [-CH_2-$CH(CH_3)$-CH_2OH], 31.97 (CH_3-CH_2-CH_2-CH_2-), 34.11 [-CH_2-$CH(CH_3)$-CH_2-], 37.12 [-CH_2-$CH(CH_3)$-CH_2-], 40.65, 41.10 [-$CH(CH_3)$-CH_2OH], 68.32 (-CH_2OH), 127.99 [olefinic, C(6)], 131.06 [olefinic, C(7)].

2.11. Isomeric 2,4-dimethyldocosanols (11)

Isomeric (6E,Z)2,4-dimethyldocos-6-enes (**10a**, **10b**) (1.324 g, 3.741 mmol, 1.0 eq.) were dissolved in diethyl ether (30 cm^3), and palladium-charcoal (10%) (0.132 g, 0.1 eq.) was added. The solution was evacuated several times and placed under an atmosphere of hydrogen gas. The suspension was rapidly stirred at room temperature for 16 h, after which the correct quantity of hydrogen gas (83.8 cm^3) had been absorbed. The suspension was filtered through a pad of Celite™, and this was washed with further aliquots of diethyl ether (4 × 25 cm^3). The solution was concentrated in vacuo to yield a crude oil. Purification by flash column chromatography using petroleum ether (b.p. 40–60°C) and ethyl acetate (95:5 to 9:1 gradient) gave **11** as a white solid. This was recrystallised from methanol to give a white solid (1.099 g, 83%). $R_f = 0.37$ in TLC solvent system 1; m.p. 42–44°C; IR (KBr disc): 3470.38 (m, br). Found M^+ 354.4721, $C_{24}H_{50}O$ requires 354.6584; ^{1}H-NMR (200 MHz) 0.85 [cm, 9H, CH_3-CH_2-CH_2-, -$CH(CH_3)$-CH_2OH, -$CH(CH_3)$-CH_2-CH-(CH_3)-], 1.23 [s, 34H, -$(CH_2)_n$-], 1.49 [m, 1H, -$CH(CH_3)$-CH_2-$CH(CH_3)$-], 1.63 [m, 3H, -CH_2-$CH(CH_3)$-CH_2OH, -$CH(CH_3)$-CH_2-CH-(CH_3)-], 3.37 (cm, 2H, -CH_2OH); ^{13}C-NMR (50.3 MHz) 14.15 (CH_3-CH_2-CH_2-CH_2-), 16.35, 17.32 [-$CH(CH_3)$-CH_2-], 19.41, 20.37 [-$CH(CH_3)$-CH_2OH], 22.71 (CH_3-CH_2-CH_2-CH_2-), 26.94, 27.09 [-$CH(CH_3)$-CH_2-$CH(CH_3)$-], 29.40, 29.73, 30.06 [-$(CH_2)_n$-], 30.73 [-CH_2-$CH(CH_3)$-CH_2OH], 31.95 (CH_3-CH_2-CH_2-CH_2-), 36.68 [-CH_2-CH_2-$CH(CH_3)$-CH_2-], 38.04, 38.82 [-CH_2-CH_2-$CH(CH_3)$-], 40.67, 41.09 [-$CH(CH_3)$-CH_2OH], 68.04 (-CH_2OH). Found C, 81.39, H, 15.24%, $C_{24}H_{50}O$ requires C, 81.27, H, 15.17%.

2.12. Isomeric 2,4-dimethyldocosanoic acids (12)

Isomeric 2,4-dimethyldocosanols (**11**) (0.475 g, 1.35 mmol, 1.0 eq.) were dissolved in anhydrous *N,N'*-dimethylformamide (30 cm^3) under nitrogen.

Activated 4 Å molecular sieves were added and the mixture stirred for 30 min before the additon of pyridinium dichromate (PDC) (4.277 g, 11.4 mmol, 8.5 eq.). The resulting solution was then stirred for 48 h at room temperature, before being poured onto saturated aqueous ammonium chloride solution (100 cm^3). The product was extracted with dichloromethane (4 × 75 cm^3) and the organic extracts combined. The organic layer was then washed with saturated aqueous sodium chloride solution (200 cm^3), dried over anhydrous magnesium sulphate, filtered and concentrated in vacuo. Purification of the residue by flash column chromatography using petroleum ether (b.p. 40–60°C) and ethyl acetate (9:1 to 2:1 gradient) as eluent gave a white solid. Recrystallisation of the product from acetone gave **12** as a white solid (0.227 g, 46%). R_f = 0.27 in TLC solvent system 1 (streak); m.p. 46–48°C; IR (KBr disc): 3424.08 (w, br), 1709.15 (s). Found M^+ 368.3672, $C_{24}H_{48}O_2$ requires 368.6420; ^{1}H-NMR (200 MHz) 0.86 ['t', 6H, J = 5.7, -(CH_3)CH-CH_2-, CH_3-CH_2-CH_2-], 1.17 [d, 3H, J = 6.9, -CH(CH_3)-COOH], 1.23 [s, 34H, (-CH_2)$_n$-], 1.42 [m, 2H, -(CH_3)CH-CH_2-CH(CH_3)-COOH], 1.73 [m, 1H, -CH(CH_3)-CH_2-CH(CH_3)-COOH], 2.54 [m, 1H, -(CH_3)CH-COOH]; ^{13}C-NMR (50.3 MHz) 14.19 (*C*H_3-CH_2-CH_2-CH_2-), 16.84, 17.84 [-(*C*H_3)CH-CH_2-], 19.37, 19.63 [-(*C*H_3)CH-COOH], 22.75 (CH_3-*C*H_2-CH_2-CH_2-), 26.83 [-*C*H_2-CH_2-CH(CH_3)-], 29.44, 29.77, 30.02 [-(*C*H_2)$_n$-], 30.76 [-CH(CH_3)-*C*H_2-CH(CH_3)-COOH], 32 01 (CH_3-CH_2-*C*H_2-CH_2-), 37.06, 37.20 [-*C*H_2-CH(CH_3)-CH_2-CH(CH_3)-COOH], 37.34 [-*C*H(CH_3)-CH_2-CH(CH_3)-COOH], 40.84, 41.31 [-*C*H(CH_3)-COOH], 183.53 (-*C*OOH).

*2.13. Isomeric methyl 2,4-dimethyldocosanoates (**13**)*

Isomeric 2,4-dimethyldocosanoic acids (**12**) (16.9 mg, 0.46 mmol) were dissolved in 15% aqueous tetrabutylammonium hydroxide (2 cm^3) in a screw-top tube to which dichloromethane (2 cm^3) and iodomethane (0.331 g, 0.1 cm^3, 2.17 mmol, 5.4 eq.) were added, and the contents of the tube were mixed for 16 h. The solution was poured into saturated aqueous ammonium chloride solution (10 cm^3) and the organic products extracted with dichloromethane (3 x 5 cm^3). The organic layers were combined, dried over anhydrous magnesium sulphate, filtered and concentrated in vacuo to give an oil. Purification using flash column chromatography with petroleum ether (b.p. 40–60°C) and ethyl acetate (95:5) gave **13** as a colourless oil (15.4 mg, 90%). R_f = 0.74 in TLC solvent system 1. Found M^+ 382.3792, $C_{25}H_{50}O_2$ requires 382.6688; ^{1}H-NMR (200 MHz) 0.85 ['t', 6H, J = 5.6, -CH(CH_3)-CH_2-(CH_3)CH-CO-, CH_3-CH_2-CH_2-] 1.13 [d, 3H, J = 6.9, -CH(CH_3)-CH_2-CH(CH_3)-CO-], 1.23 [s, 34H, (-CH_2)$_n$-], 1.48–1.81 [m, 3H, -CH_2-CH(CH_3)-COO-CH_3, -(CH_3)CH-CH_2-(CH_3)CH-COO-CH_3], 2.50 [m, 1H, -(CH_3)CH-COO-CH_3], 3.62 (s, 3H, -COO-CH_3).

*2.14. (2R)-N-2-Naphthylethyl-(2R; 4R,S)-dimethyldocosamide (**14**) and (2R)-N-2-naphthylethyl-(2S; 4R,S)-dimethyldocosamide (**15**)* [13,14]

Isomeric 2,4-dimethyldocosanoic acids (65.5 mg, 0.178 mmol, 1.0 eq.) were dissolved in an excess of oxalyl chloride (2 cm^3) in a screw-top tube. The contents were heated at 70°C for 1 h before removal of the excess oxalyl chloride in vacuo to give the crude acid chloride. 4-Dimethylaminopyridine (26.1 mg, 0.214 mmol, 1.2 eq.) and (R)-(−)-1-(1-naphthyl)ethylamine (46.5 mg, 0.292 mmol, 1.2 eq.) were each dissolved in dry dichloromethane (2 cm^3) and added, under an atmosphere of nitrogen, to the acid chloride. The tube contents were then mixed for 16 h at room temperature and the reaction was quenched by pouring the mixture into saturated aqueous ammonium chloride solution (15 cm^3). The organic products were extracted using dichloromethane (3 × 10 cm^3). The organic layers were combined and washed with saturated aqueous sodium chloride solution (25 cm^3), dried over anhydrous magnesium sulphate, filtered and concentrated in vacuo to give a crude solid. Purification using preparative TLC (3× in solvent system 2) gave **14** (13.9 mg, 15%) and **15** (21.1 mg, 22%) as white solids.

Compound **14** showed R_f = 0.50 in solvent system 2; $[\alpha]_D^{16}$ (c 1.0, $CHCl_3$) −23.6°; IR (KBr disc): 3283.26 (s, br), 2918.67 (s), 2851.15 (s), 1718.79 (m), 1633.91 (s), 1541.91 (m), 1466.09 (m), 1383.14 (m), 1097.04 (s), 800.56 (m), 777.41 (m), 700.25 (m), 464.90 (m). Found M^+ 521.4648, $C_{36}H_{59}NO$

requires 521.8682; ^{1}H-NMR (200 MHz) 0.86 [t, 6H, J = 6.6, CH_3-CH$_2$-CH$_2$-, -CH(CH_3)-CH$_2$-CH(CH$_3$)-CO-], 1.07 [d, 3H, J = 6.8, -NH-CH(CH_3)-], 1.23 [s, 34H, -(CH_2)$_n$-], 1.66 [d, 3H, J = 6.75, -CH(CH_3)-CO-NH-], 1.76 (m, 3H, -CH(CH$_3$)-CH_2-CH(CH$_3$)-CO-), 2.20 [m, 1H, -CH(CH$_3$)-CO-NH-], 5.59 (d, 1H, J = 8.4, -CO-NH-), 6.78 [m, 1H, J = 6.8, -CO-NH-CH(CH$_3$)-], 7.41–7.56 (m, 4H, naphthyl), 7.78–7.88 (m, 2H, naphthyl), 8.04–8.10 (m, 1H, naphthyl).

Compound **15** showed R_f = 0.33 in solvent system 2; $[\alpha]_D^{16}$ (c 1.0, CHCl$_3$): −16.0°; IR (KBr disc): 3261.79 (s, br), 2918.52 (s), 2851.02 (s), 1717.53 (m), 1632.16 (s), 1540.65 (m), 1466.09 (m), 1381.41 (m), 1097.04 (s), 799.79 (m), 776.71 (m), 700.25 (m), 464.90 (m). Found M$^+$ 521.4621, $C_{36}H_{59}NO$ requires 521.8682; ^{1}H-NMR (200 MHz) 0.86 [t, 6H, J = 6.1, CH_3-CH$_2$-CH$_2$-, -CH(CH_3)-CH$_2$-CH(CH$_3$)-CO-], 1.11 [d, 3H, J = 6.8, -NH-CH(CH_3)-], 1.23 [s, 34H, -(CH_2)$_n$-], 1.65 [d, 3H, J = 6.7, -CH(CH_3)-CO-NH-], 1.76 (m, 3H, -CH-(CH$_3$)-CH_2-CH(CH$_3$)-CO-), 2.22 [m, 1H, -CH(CH$_3$)-CO-NH-], 5.58 (d, 1H, J = 8.0, -CO-NH-), 5.88 [m, 1H, J = 6.8, -CO-NH-CH(CH$_3$)-], 7.39–7.51 (m, 4H, naphthyl), 7.76–7.97 (m, 2H, naphthyl), 8.02–8.10 (m, 1H, naphthyl).

*2.15. (2R; 4R,S)- and (2S; 4R,S)-dimethyldocosanoic acids (**16** and **17**)*

The amide was dissolved in 1,4-dioxane (2 cm^3) in separate screw-top tubes, and equal volumes of 3N aqueous sulphuric acid (2 cm^3) were added. The tubes were heated to 110°C for 16 h, cooled, and the contents poured onto saturated aqueous ammonium chloride solution (20 cm^3). The products were extracted using dichloromethane (3 × 10 cm^3) and the organic layers combined. The organic layers were washed with saturated aqueous sodium chloride solution (25 cm^3), dried over anhydrous magnesium sulphate, filtered and concentrated in vacuo to give black oils. Purification by flash column chromatography yielded the acids as yellow oils.

Compound **16** (7.0 mg, 75%) showed R_f = 0.25 in TLC solvent system 1 (streak). Found M$^+$ 368.8162, $C_{24}H_{48}O_2$ requires M 368.6420; ^{1}H-NMR (200 MHz) 0.85 ['t', 6H, J = 5.3, -(CH_3)CH-CH$_2$-CH(CH$_3$)-, CH_3-CH$_2$-CH$_2$-], 1.15 [d, 3H, J = 6.7, -CH(CH_3)-CO-], 1.23 [s, 34H, (-CH_2)$_n$-], 1.42 [m, 1H, -(CH$_3$)CH-CH$_2$-CH(CH$_3$)-COOH], 1.74 [m, 3H, -CH_2-CH(CH$_3$)-COOH, -(CH$_3$)CH-CH$_2$-(CH$_3$)CH-COOH], 2.56 [m, 2H, -(CH$_3$)CH-COOH].

Compound **17** (10.5 mg, 71%) showed R_f = 0.26 in TLC solvent system 1 (streak). Found M$^+$ 368.7984, $C_{24}H_{48}O_2$ requires M 368.6420; ^{1}H-NMR (200 MHz) 0.86 ['t', 6H, J = 5.1, -(CH_3)CH-CH$_2$-CH(CH$_3$)-CO-, CH_3-CH$_2$-CH$_2$-], 1.17 [d, 3H, J = 6.9, -CH(CH_3)-CH$_2$-], 1.23 [s, 34H, (-CH_2)$_n$-], 1.43 [m, 1H, -(CH$_3$)CH-CH$_2$-CH(CH$_3$)-COOH], 1.74 [m, 3H, -CH_2-CH(CH$_3$)-COOH, -(CH$_3$)CH-CH$_2$-(CH$_3$)CH-COOH], 2.56 [m, 1H, -(CH$_3$)CH-COOH].

*2.16. Methyl (2R; 4R,S)- and (2S; 4R,S)-2,4-dimethyldocosanoates (**18** and **19**)*

The acids **16** and **17** were dissolved in 15% aqueous tetrabutylammonium hydroxide (2 cm^3) in separate screw-top tubes, and dichloromethane (2 cm^3) and iodomethane (0.10 cm^3, 0.332 g, 2.17 mmol) were added to each tube and the contents of the tubes mixed for 16 h at room temperature. The reaction mixtures were poured into saturated aqueous ammonium chloride solution (20 cm^3) and the products extracted with dichloromethane (3 × 10 cm^3). The organic layers were combined, washed with saturated aqueous sodium chloride solution (20 cm^3), dried over anhydrous magnesium sulphate and concentrated in vacuo. Purification by flash column chromatography, using petroleum ether (b.p. 40–60°C) and ethyl acetate (9:1) yielded **18** (2.3 mg, 66%) and **19** (2.7 mg, 77%) as colourless oils.

Compound **18** showed R_f = 0.75 in TLC solvent system 1; $[\alpha]_D^{16}$ (c 0.5, CHCl$_3$): −2.4°. Found M$^+$ 382.4126, $C_{25}H_{50}O_2$ requires 382.6688; ^{1}H-NMR (200 MHz) 0.86 ['t', 6H, J = 6.8, -CH(CH_3)-CH$_2$-(CH$_3$)CH-CO-, CH_3-CH$_2$-CH$_2$-], 1.16 [d, 3H, J = 7.0, -CH(CH_3)-CO-], 1.23 [s, 34H, (-CH_2)$_n$-], 1.42–1.73 [m, 3H, -CH_2-CH(CH$_3$)-COO-CH$_3$, -(CH$_3$)CH-CH$_2$-(CH$_3$)CH-COO-CH$_3$], 2.57 [m, 1H, -(CH$_3$)CH-COO-CH$_3$], 3.65 (s, 3H, -COO-CH_3).

Compound **19** showed R_f = 0.74 in solvent sys-

tem 1; $[\alpha]_D^{16}$(c 0.5, $CHCl_3$): +3.2°. Found 382.4054, $C_{25}H_{50}O_2$ requires 382.6688; ^{1}H-NMR (200 MHz) 0.86 ['t', 6H, J = 6.8, -(C*H*$_3$)CH-CH$_2$-CH(CH$_3$)-CO-, C*H*$_3$-CH$_2$-CH$_2$-], 1.14 [d, 3H, J = 6.9, -CH(C*H*$_3$)-CO-], 1.23 [s, 34H, (-C*H*$_2$)$_n$-], 1.41–1.72 [m, 3H, -C*H*$_2$-CH(CH$_3$)-COO-CH$_3$, -(CH$_3$)C*H*-CH$_2$-(CH$_3$)CH-COO-CH$_3$], 2.51 [m, 1H, -(CH$_3$)C*H*-COO-CH$_3$], 3.61 (s, 3H, -COO-C*H*$_3$).

2.17. *2,3,4,5,6-pentafluorobenzyl-(2R;4R,S)- and 2,3,4,5,6-pentafluorobenzyl-(2S;4R,S)-2,4-dimethyldocosanoate (**20** and **21**)*

The acids were dissolved in 15% aqueous tetrabutylammonium hydroxide (2 cm^3), and dichloromethane (2 cm^3) and 2,3,4,5,6-pentafluorobenzyl bromide (0.1 cm^3) were added and the contents of the tube mixed at room temperature for 16 h. The reaction mixtures were poured onto saturated aqueous ammonium chloride solution (15 cm^3) and extracted with diethyl ether (3 × 10 cm^3). The ethereal layers were combined and dried over anhydrous magnesium sulphate, filtered and concentrated in vacuo. Purification by preparative TLC in solvent system 1 yielded **20** (3.8 mg, 24%) and **21** (5.6 mg, 36%) as colourless oils.

Compound **20** showed R_f = 0.76 in solvent system 1; $[\alpha]_D^{16}$ (c 0.5, $CHCl_3$): −3.2°. Found M$^+$ 548.9185, $C_{31}H_{49}F_5O_2$ requires 548.7189; ^{1}H-NMR (200 MHz) 0.84 ['t', 6H, J = 6.7, -(C*H*$_3$)CH-CH$_2$-CH(CH$_3$)-CO-, C*H*$_3$-CH$_2$-CH$_2$-], 1.17 [d, 3H, J = 6.9, -CH(C*H*$_3$)-COO-], 1.23 [s, 34H, (-C*H*$_2$)$_n$-], 1.41–1.71 [m, 3H, -C*H*$_2$-CH(CH$_3$)-COO-CH$_2$-, -(CH$_3$)C*H*-CH$_2$-(CH$_3$)CH-COO-CH$_2$-], 2.32 [m, 1H, -(CH$_3$)C*H*-COO-CH$_2$-], 5.31 (s, 2H, -COO-C*H*$_2$-).

Compound **21** showed R_f = 0.75 in solvent system 1; $[\alpha]_D^{16}$ (c 0.5, $CHCl_3$): +4.8°. Found M$^+$ 548.9273, $C_{31}H_{49}F_5O_2$ requires 548.7189; ^{1}H-NMR (200 MHz) 0.85 ['t', 6H, J = 6.8, -(C*H*$_3$)CH-CH$_2$-CH(CH$_3$)-CO-, C*H*$_3$-CH$_2$-CH$_2$-], 1.16 [d, 3H, J = 6.9, -CH(C*H*$_3$)-CH$_2$-], 1.23 [s, 34H, (-C*H*$_2$)$_n$-], 1.42–1.73 [m, 3H, -C*H*$_2$-CH(CH$_3$)-COO-CH$_2$-, -(CH$_3$)C*H*-CH$_2$-(CH$_3$)CH-COO-CH$_2$-], 2.35 [m, 1H, -(CH$_3$)C*H*-COO-CH$_2$-], 5.31 (s, 3H, -COO-C*H*$_2$-).

3. Results and discussion

The acid was disconnected according to the scheme shown in Fig. 3. This route was designed so as to permit the coupling of different long-chain Wittig reagents in future syntheses (Scheme 1).

The oxidation of racemic 3,5-dimethylcyclohexan-1-ol (**1**) to the cyclic ketone (**2**) proceeded smoothly. Preparation of the caprolactone (**3**) needed care because of the volatility of the product. Eventually, the scheme was modified to include a crude work-up of the lactone (**3**) and then immediate ring-opening without further purification. Aqueous sodium hydroxide gave the best yield in the preparation of the hydroxy acid (**4**) , by the ring-opening reaction of the lactone (**3**). Acid hydrolysis gave a number of undesired products. A basic phase-transfer esterification procedure, using 15% aqueous tetrabutylammonium hydroxide and iodomethane, gave the desired hydroxy ester (**5**). Esterification with methanol in the presence of acid

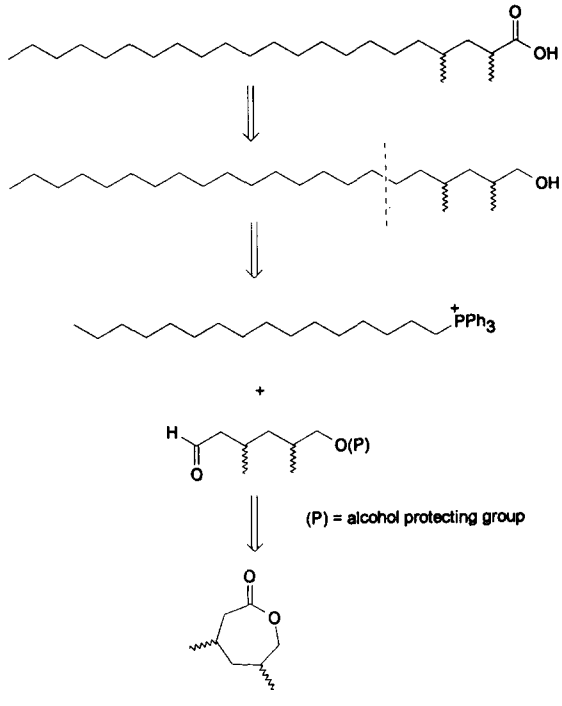
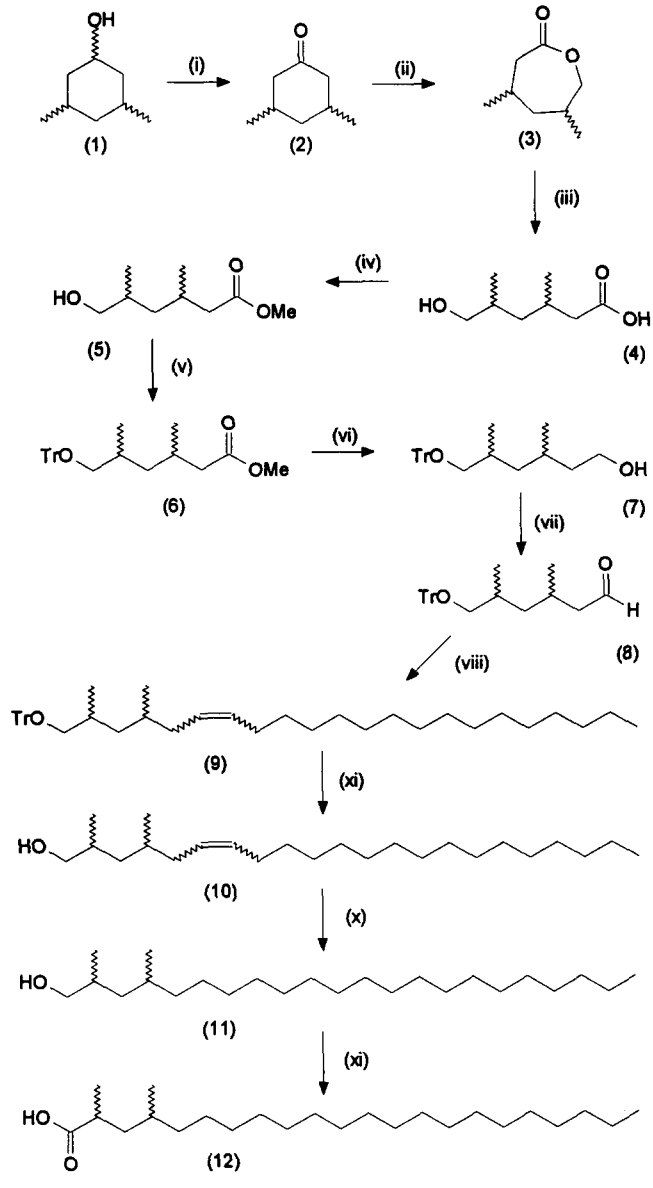


Fig. 3. Disconnection strategy for (2R,S; 4R,S)-dimethyldocosanoic acid.



Scheme 1. Synthesis of isomeric 2,4-dimethyldocosanoic acids. (i) Sodium dichromate dihydrate, concentrated sulphuric acid, glacial acetic acid, water; (ii) *meta*-chloroperbenzoic acid, *para*-toluenesulfonic acid, dichloromethane; (iii) 30% aqueous sodium hydroxide solution, dichloromethane; (iv) 15% aqueous tetrabutylammonium hydroxide, dichloromethane, iodomethane; (v) triphenylmethyl chloride, dry pyridine, dry dichloromethane; (vi) lithium aluminium hydride, dry tetrahydrofuran; (vii) pyridinium chlorochromate, dry dichloromethane, 4 Å molecular sieves; (viii) hexadecyltriphenylphosphonium bromide, *n*-butyl lithium, dry tetrahydrofuran; (ix) trifluoroacetic acid, dichloromethane, water, sodium hydrogen carbonate; (x) 10% palladium on charcoal, hydrogen gas, diethyl ether; (xi) pyridinium dichromate, dry *N,N'*-dimethylformamide, 4 Å molecular sieves.

catalysts gave inferior yields and several by-products.

The protection of the primary alcohol group of the hydroxy ester (**5**), as a triphenylmethyl ether, proceeded under standard conditions to give the trityl ether (**6**) in 87% yield. Other protecting groups were considered, but the trityl ether was favoured to reduce the volatility of the protected alcohol. Li-

thium aluminium hydride reduction of the ester (**6**) gave the desired alcohol (**7**). Despite several attempts at recrystallisation of this compound, no crystalline product could be obtained.

Pyridinium chlorochromate (PCC) oxidation of the alcohol (**7**), catalysed by activated 4 Å molecular sieves, formed the aldehyde (**8**). The sixteen-carbon Wittig reagent (**24**) was prepared under standard conditions. The chain extension of the aldehyde (**8**) with this Wittig reagent proved to be quite difficult. Initially, dry dichloromethane was used as the solvent with *n*-butyl lithium as the base, but only low yields of the desired product (**9**) were isolated. Several other solvent/base systems were attempted, including potassium hexadimethylsilazane (KHMDS) in dry toluene and KHMDS in dry dichloromethane, but the best yield was gained by the use of *n*-butyl lithium in dry tetrahydrofuran (THF). The Wittig reagent was initially only partially soluble in the THF, but upon formation of the phosphorus ylide, the characteristic deep red colour of the deprotonated Wittig reagent was observed. The amount of Wittig reagent used in the reaction was also crucial, the optimum quantity being 3.3 equivalents of the phosphorus ylide. The ratio of possible (E) and (Z) isomers, (**9a** and **9b**), could not be determined by GC. The 200-MHz NMR spectrum showed that a mixture of (E) and (Z) isomers was present, but it was difficult to make a quantitative estimate. Predicted results using a non-stabilised ylide are for a mixture of (E) and (Z) isomers. The ^{1}H-NMR spectrum of **9a/9b** and **10a/10b** showed a complex multiplet for the olefinic and allylic protons, with a range of coupling constants corresponding to both (E) and (Z) isomers.

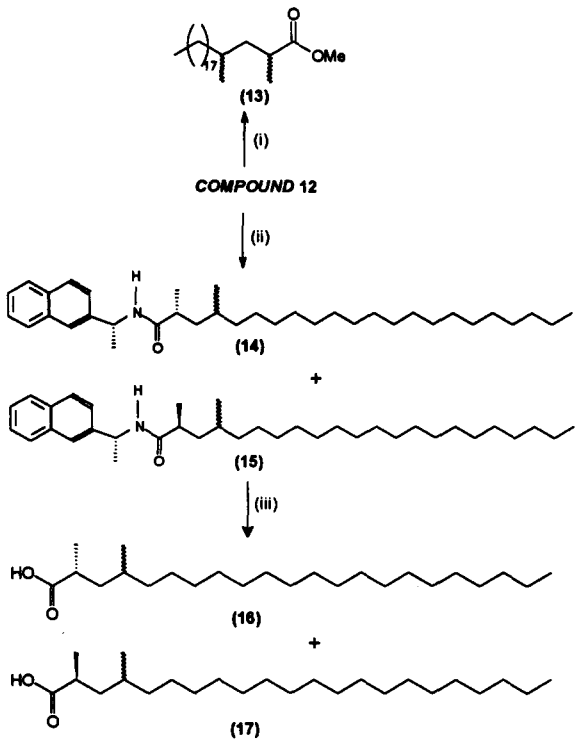
Deprotection of the triphenylmethyl ether protecting group of the **9a/9b** mixture occurred in good yield, using brief treatment with trifluoroacetic acid (TFA). Upon the addition of the TFA to the dichloromethane solution, a deep yellow colour was observed due to the formation of the trityl cation (Ph_3P^+). When water was added to the mixture as a quench, the colour quickly faded. The double bond of the vinyl alcohol (**10a**, **10b**) was easily removed by hydrogenation. The trityl protecting group was removed before the hydrogenation stage because of possible interference of the aromatic groups with the surface of the catalyst. The crystalline product (**11**) was oxidised to the carboxylic acid (**12**) by the use of pyridinium dichromate (PDC), again in the presence of activated 4 Å molecular sieves.

Assignments of the ^{13}C-NMR spectra were made by comparison with other racemic and enantiomerically pure methyl-branched compounds, such as tuberculostearic acid [15]. The CH_3 groups are paired together according to this data. For compound **4**, the CH_3 group at C-3 exhibits shifts of 19.37 and 20.49 ppm and the CH_3 at C-5 similar shifts of 16.17 and 17.42 ppm (see Table 1 for further details).

DEPT ^{13}C-NMR studies were performed on compounds **4–12** and clearly distinguished CH_3 and CH groups from the CH_2 groups present. Table 1 displays a correlation between the major carbon atoms in each compound and their chemical shifts (δ/ppm).

The methyl ester of (2R,S; 4R,S)-dimethyldocosanoic acid (**13**) was formed by phase transfer catalysis using aqueous tetrabutylammonium hydroxide (TBAH) and iodomethane. GC analysis of the racemic ester, using GC programme A, with the standard fatty acid methyl esters (C_{16} to C_{18}), showed an equivalent chain-length (ECL) value of 20.89. No separation of diastereoisomers was observed upon TLC analysis or with reverse-phase HPTLC. A partial separation of diastereoisomers was observed using GC programme B, the two isomers having ECL values of 20.87 and 20.91.

The racemic acid (**12**) was resolved at the C-2 chiral centre by the use of (R)-(−)-naphthylethylamides [13,14] (Scheme 2). Conversion of **12** to the acid chloride (**13**) and then reaction with (R)-(−)-naphthylethylamine in the presence of 4-dimethylaminopyridine gave a mixture of derivatives. On normal-phase TLC, a clear separation was observed with development, three times, in solvent system 2. The relative stereochemistry of the chiral centre at C-2 was assigned according to the rules of Helmchen et al. [13,14]. These state that where an (R)-resolving agent is used, i.e. (R)-(−)-naphthylethylamine, then the (R)-isomer of the C-2 chiral centre will bind less strongly to a silica gel stationary phase compared with the (S)-isomer. The (2R; 4R,S)-isomer (**14**) showed $R_f = 0.50$ and the (2S; 4R,S)-isomer (**15**) showed $R_f = 0.33$ (both in solvent system 2). Reverse-phase high-performance thin-layer chroma-



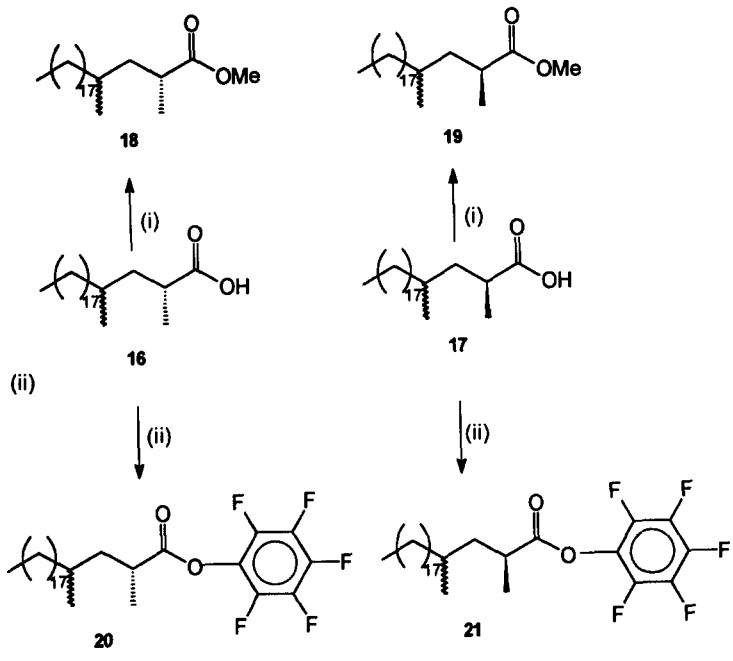
Scheme 2. Resolution of isomeric 2,4-dimethyldocosanoic acid. (i) 15% Aqueous tetrabutylammonium hydroxide, methyl iodide, dichloromethane; (ii) oxalyl chloride, (R)-(−)-2-naphthylethylamine, 4-dimethylaminopyridine, dry dichloromethane; (iii) 1,4-dioxane, 3 N aqueous sulfuric acid.

tography (HPTLC) of both of these isomers was attempted in order to attempt to separate each into the corresponding pairs of enantiomers. When acetonitrile:tetrahydrofuran (2:1) was used as solvent during reverse-phase HPTLC (8×), a slight separation of the enantiomers was observed, but not sufficient to allow complete resolution.

The two diastereoisomeric naphthylethylamides (**14** and **15**) were hydrolysed with 1,4-dioxane and 3N aqueous sulphuric acid, a method causing no racemisation [13,14]. Hence, **14** gave the (2R; 4R,S)-acid (**16**) and **15** gave the (2S; 4R,S)-acid (**17**). The liberated acids (**16** and **17**) were produced in yields of 15 and 22%, respectively. Unpublished work in this laboratory had shown that 2,3,4,5,6-pentafluorobenzyl (PFB) esters of multimethyl branched acids showed some slight separation on 25-m capillary GC columns. Hence, the two acids **16** and **17** were derivatised as PFB esters (Scheme 3) and analysed by GC. Using GC programme B, only a very slight separation was observed for the diastereomeric pairs of compounds. Compounds **16** and **17** were also derivatised as methyl esters (Scheme 3) using phase-transfer esterification. GC retention times using GC programme A showed ECL values of 20.92 for compound **20** and 20.93 for compound **21**. When a capillary column was employed (programme B), a

Table 1
^{13}C-NMR data for carbon atoms 1–6 (C 1–6) and methyl branch carbon atoms

Compound	C-1	C-2	C-3	C-4	C-5	C-6	Me at C-2/C-3	Me at C-4/C-5
4	178.69	41.31	27.46	32.89	40.10	67.55	19.37	16.17
		42.41	27.57	—	40.40	68.50	20.49	17.42
5	173.81	41.28	27.57	32.97	40.20	67.60	19.37	16.19
		42.49	27.68	—	40.49	68.37	20.51	17.44
6	173.73	41.53	27.82	30.99	40.87	68.25	19.23	16.94
		42.62	27.93	31.46	41.27	68.95	20.47	18.12
7	61.86	41.45	26.70	31.46	40.70	68.29	18.50	17.35
		41.86	26.94	33.02	—	69.02	20.62	17.75
8	203.10	41.16	25.47	30.98	40.47	68.07	19.74	17.27
		41.48	25.76	31.46	—	68.73	20.65	18.19
9	68.39	40.86	30.69	37.06	34.29	128.52	17.23	20.25
		41.31	—	38.24	—	—	18.48	20.66
10	68.32	40.65	30.76	37.12	34.11	127.99	16.38	20.33
		41.10	—	38.36	—	—	17.34	21.06
11	68.04	40.67	30.73	38.04	36.68	27.09	16.35	19.41
		41.09	—	38.82	—	26.94	17.32	20.37
12	183.53	40.84	30.76	37.34	37.06	26.83	16.84	19.37
		41.31	—	—	37.20	—	17.84	19.63



Scheme 3. Derivatisation of (2R; 4R,S)- and (2S; 4R,S)-2,4-dimethyldocosanoic acids. (i) 15% aqueous tetrabutylammonium hydroxide, iodomethane, dichloromethane; (ii) 15% aqueous tetrabutylammonium hydroxide, 2,3,4,5,6-pentafluorobenzyl bromide, dichloromethane.

partial separation of diastereisomers was observed in the case of compound **13**. For the pairs of enantiomers contained within compound **20**, or compound **21**, then only a very small separation was observed on GC.

An efficient synthetic route to 2,4-dimethyldocosanoic acid has been achieved via a novel, convergent synthesis. If it was possible to obtain *cis*- or *trans*-3,5-dimethylcyclohexanone, resolution of the 2-methyl branch at the completion of such a synthesis would permit the assignment of the absolute stereochemistry at C-2 and allow the assignment of the relative stereochemistry at C-4, depending on whether pure *cis* or pure *trans* starting material was used. The *cis*-isomer of 3,5-dimethylcyclohexanol is accessible by hydrogenation of 3,5-xylenol [16], but facilities for this reaction are not currently available in this laboratory.

The current synthetic route provides access to a range of dimethyl and multi-methyl branched fatty acids which are components of a range of mycobacterial lipids [1,4].

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References

1 P.J. Brennan (1980) in: C. Ratledge and S.G. Wilkinson (Eds.), Microbial Lipids, Vol. 1, Academic Press, Orlando, pp. 203–296.
2 F. Festenstein and J.M. Grange (1991) J. Appl. Bacteriol. 71, 19–30.
3 B.R. Bloom and C.J.C. Murry (1992) Science 257, 1055–1064.

4 D.E. Minnikin (1982) in: C. Ratledge and J.L. Stanford (Eds.), The Biology of the Mycobacteria, Vol. 1, Academic Press, London, pp. 95–184.
5 M.E. Hamid, J.L. Fraser, P.A. Wallace, G.S. Besra, M. Goodfellow, D.E. Minnikin and M. Ridell (1993) Lett. Appl. Microbiol. 16, 132–135.
6 A. Lemassu, M.-A. Lanélle and M. Daffé (1991) FEMS Microbiol. Lett. 78, 171–176.
7 H.H. Baer (1993) Carbohydr. Res. 240, 1–22.
8 D. E Minnikin, G. Dobson, D. Sesardic and M. Ridell (1985) J. Gen. Microbiol. 131, 1369–1374.
9 G.S. Besra, R.C. Bolton, M.R. McNeil, M. Ridell, K.E. Simpson, J. Glushka, H. van Halbeek, P.J. Brennan and D.E. Minnikin (1992), Biochemistry 31, 9832–9837.
10 P.A. Wallace and D.E. Minnikin (1993) J. Chem. Soc., Chem. Commun., 1292–1293.
11 P.A. Wallace, D.E. Minnikin and M. Ridell (1994) J. Chem. Soc., Chem. Commun., 329–330.
12 E.J. Corey, H.E. Ensley and J.W. Suggs (1976) J. Org. Chem. 41, 380–382.
13 G. Helmchen, G. Nill, D. Flochers, W. Schule and M.J.K. Jough (1979) Angew. Chem. Int. Ed. Engl. 18, 62–67.
14 G. Helmchen (1974) Tetrahedron Lett. 16, 1527–1529.
15 P. A Wallace and D.E. Minnikin (1994), Chem. Phys. Lipids 71, 145–162.
16 H.E. Ungade and A.D. McLaren (1944) J. Am. Chem. Soc. 66, 118–122.