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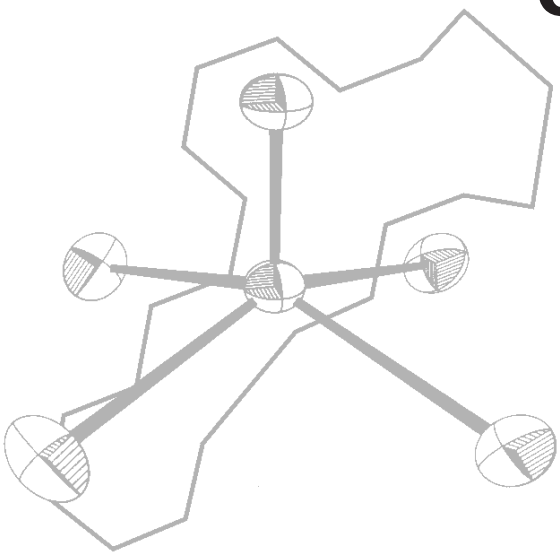

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Synthesis of Stereoisomerically Pure Monoether Lipids

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Four stereoisomerically pure lipids have been synthesized which contain chirally pure phytanol (obtained through resolution of a chiral amide) and chiral glycerol moieties. The lipids were obtained in 7–13% overall yield in eight steps from commercially available materials.

Introduction

Life in its simplest of forms, the prokaryotic cell, has existed on Earth for at least 3·5 billion years. During the course of evolution the eukaryotic cell became the basis for the development of the plant and animal kingdom. A third genotype, the Archaea, has however recently been classified as a new evolutionary lineage.[1] Organisms belonging to this family possess similarity to both prokaryotes in that they have no membrane-bound nucleus, and eukaryotes through ribosomal RNA genealogies.

Organisms belonging to the Archaea are widespread throughout Nature, and are remarkable for the extreme conditions in which they live. Archaea have recorded the highest known temperature for an environment that supports life, 113°C,[2] as well as some of the coldest, −1·8°C, in Antarctic waters.[3] They can thrive in water whose salt content is five times that of oceanic waters[4] and also where pH values range from below 5 for the acidophiles to above 9 for the alkaliphiles.[5]

The Archaea are able to exist and thrive in such extreme conditions through modifications of their cellular structure. Principally, modifications of their cellular membrane components provide them with stability to these harsh environments. The lipids of both eukaryotes and prokaryotes are typically comprised of straight chain partly unsaturated carboxylic acids linked to glycerol through an ester bond. Hydrolysis of the ester bond and/or oxidation of the unsaturated moieties under harsh environmental conditions probably leads ultimately to the destruction of the cell membrane. Lipids of the Archaea consist primarily of fully saturated, isoprenoid hydrocarbons attached to glycerol through an ether linkage, thus rendering them resistant to oxidative and hydrolytic degradation.

For example, the lipids of the archaeabacterium *Halobacterium cutiribrum* consist primarily of the chiral diether (1) where the configuration at the glycerol has been assigned *R*,[6,7] in contrast to conventional glycerolipids of the prokaryotes and eukaryotes where the stereochemistry of glycerol is reversed. The complete stereochemistry of the alkyl side chain shown in (1) has been assigned by degradation and comparison of the resultant products with materials of known optical configuration.[7] This alkyl chain, found in the membranes of many other Archaea, is derived from the C_{20} residue phytanol (2), found as either C 3 epimer in Nature.

There has been considerable interest in recent years relating to the use of the isoprenoid phytanol (2), and its phytanylated glycerol derivatives (1) and (3), in studies of membrane formation and function.[4,8–10] The majority of such publications have employed chirally impure compounds derived synthetically through the use of chirally impure phytanol, racemic glycerol, or both. Only one paper has employed chirally pure materials; however, in this work only one of the four* possible diastereoisomers of (3) was produced.[11] The effect that the chirality of both the glycerol and C 3 centres of phytanol has on membrane formation, packing and function has however never been studied.[10] In this paper we describe the synthesis of two diastereoisomers of phytanol (2) and their subsequent conversion

* In this work only changes at the epimeric centres at C 3 of phytanol and C 2 of glycerol are examined, while the stereocentres at C 7 and C 11 of phytanol remain unaltered (both *R*, derived from the starting material phytol). Therefore for the purpose of this paper the possible number of GMPE stereoisomers derived from phytol is considered as four.

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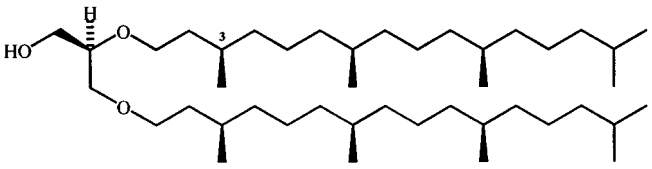
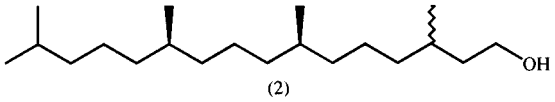
into four diastereomers of glycerol monophytanyl ether (GMPE) (3).



(1)



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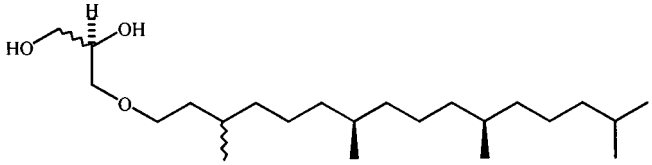


(3)

Results and Discussion

Our route to the four diastereomers of (3) involved the synthesis of chirally pure phytanols and subsequent etherification of the known protected, chiral glycerols (4) and (5)* (Scheme 1). Stereochemically pure phytanol has been obtained in small quantities from natural sources such as *Halobacterium cutiribrum* through a procedure involving a series of extractions and hydrolyses that break down the lipids to their corresponding subunits.[7] Chirally pure phytanol (e.e. >99%) has also been obtained synthetically by asymmetric hydrogenation of the commercially available allylic alcohol phytol (6).[13] The first method requires the extensive procedure of culturing the appropriate Archaea, extraction of the membranous material and then carrying out the necessary isolation and purification processes. The second method utilizes a chiral catalyst that requires independent multistep synthesis. For these reasons we opted for a more classical route to enantiomerically pure phytanol involving separation of appropriate diastereomeric derivatives.

It was our intention to oxidize the C 3 epimeric phytanols to the corresponding phytanic acids and then to prepare a diastereomeric mixture of secondary amide derivatives. Chromatographic separation followed by removal of the chiral auxiliary would then allow the chirally pure phytanols (7) and (8) to be obtained. Etherification of glycerol could then be effected by suitable derivatization of the chiral phytanols and reaction with the protected glycerols (4) and (5).

Hydrogenation of the allylic alcohol phytol (6) under an atmosphere of hydrogen with Raney nickel as catalyst[14] afforded the phytanols (2) epimeric at C 3 in quantitative yield (Scheme 2). The spectroscopic data for the mixture (2) were in full accord with literature data.[11] Moss and Fujita have reported that these diastereomers can be separated by column chromatography;[9] however, we were unable to effect any separation using either conventional column chromatography or h.p.l.c. The diastereomers could not be differentiated in the ^{1}H n.m.r. spectrum (at 400 MHz) nor in the ^{13}C n.m.r. spectrum (at 100 MHz).

Oxidation of the diastereomeric mixture of (2) with chromium trioxide in a 2 : 1 solution of acetone/acetic acid provided the phytanic acids (9) in 60% yield.[13] Again separation of the diastereomers was impossible by standard chromatographic techniques and the individual diastereomers could not be discerned in high-field n.m.r. spectra.

In Sita's work on the asymmetric hydrogenation of phytol,[13] the purity of the chiral products (chirally pure phytanol) was determined by conversion into chiral naphthylethylamide derivatives which exhibited subtle differences in chromatographic behaviour and spectroscopic data.[13] The absolute stereochemistry of the products was deduced from the mechanism of the chiral hydrogenation catalyst employed.[15]

The acid chloride was formed *in situ* from the acid (9), by using oxalyl chloride and catalytic dimethylformamide, and reaction with the commercially available chiral naphthylethylamine (10) furnished the 3*R* and 3*S* naphthylethylamide diastereomers (11) and (12) of phytanic acid in a 6 : 4 ratio† (respectively) in 71% overall yield (Scheme 2). This mixture could be partially separated by flash column chromatography (6 : 1 hexane/diethyl ether); however, material with >99% d.e. was obtained following h.p.l.c. The spectroscopic and physical data for (11) and (12) were in accord with those reported. Furthermore, the two compounds exhibited differences in their ^{13}C n.m.r. spectra and polarity on t.l.c. and h.p.l.c., thus allowing the absolute stereochemistry to be assigned by direct comparison with the data reported by Sita.[13]

All subsequent reactions were performed on the separated pure diastereomers, and all subsequent materials are assumed to be enantiomerically pure. Hydrolysis of the amides (11) and (12) to obtain the phytanic acids (13) and (14) proved difficult under standard conditions (Scheme 3). Treatment of the amide with concentrated sulfuric acid in tetrahydrofuran at reflux led over many days to cleavage at the N–alkyl bond of the chiral auxiliary, generating the primary amides (15) and (16) (in *c.* 70% yield) with little or no formation of the desired acids (13) and (14). Such cleavage in the hydrolysis of amides is rare, though has been reported previously.[16] The structures of the amides (15) and (16) were deduced from their spectroscopic data, in particular the infrared spectra where each exhibits

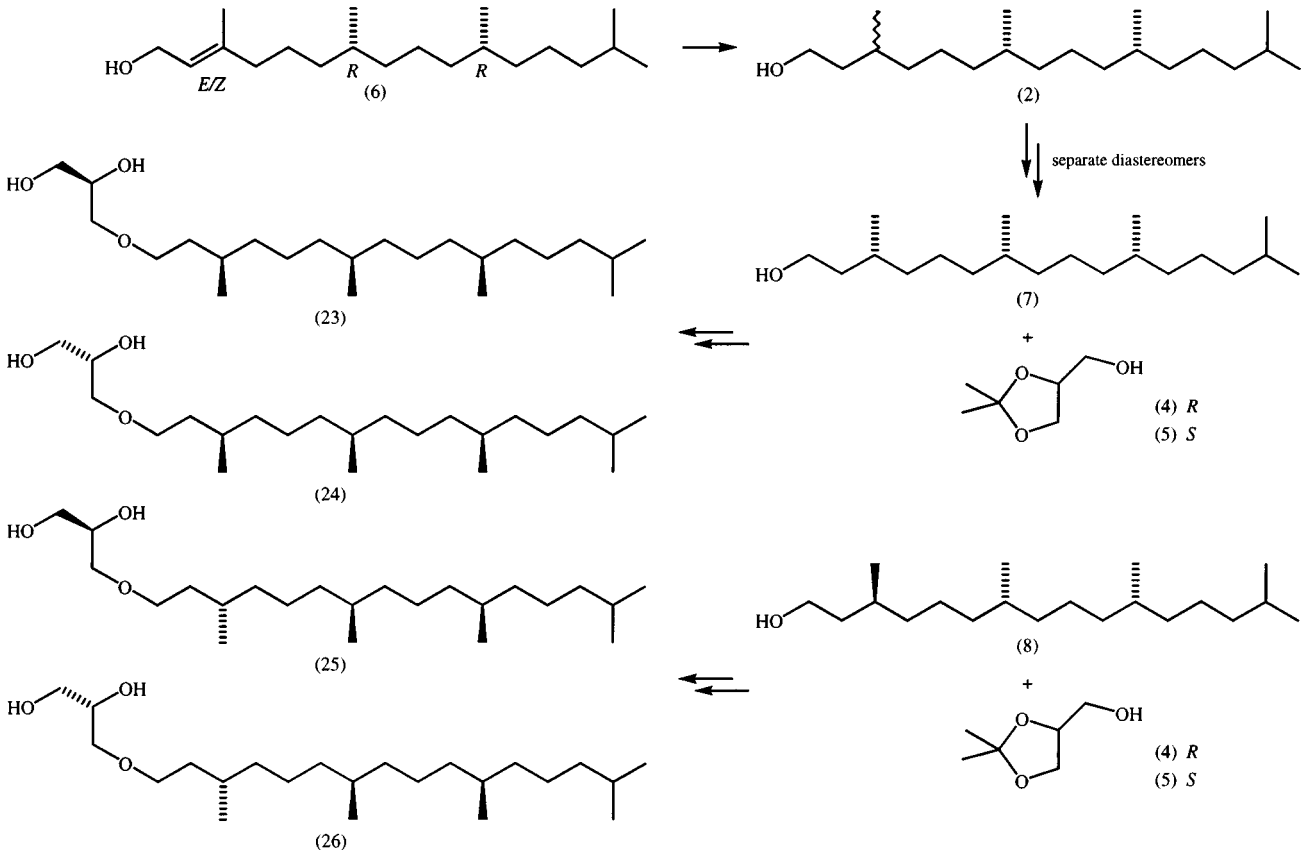
* Of the two protected glycerols employed in this work, *R* isopropylideneglycerol (4) is commercially available, whereas *S* isopropylideneglycerol (5) was formed following the procedures of Jung and Shaw,[12] and Jackson.[12]

† Commercially available phytol is supplied as an approximately 65 : 35 mixture of *E* and *Z* stereoisomers.

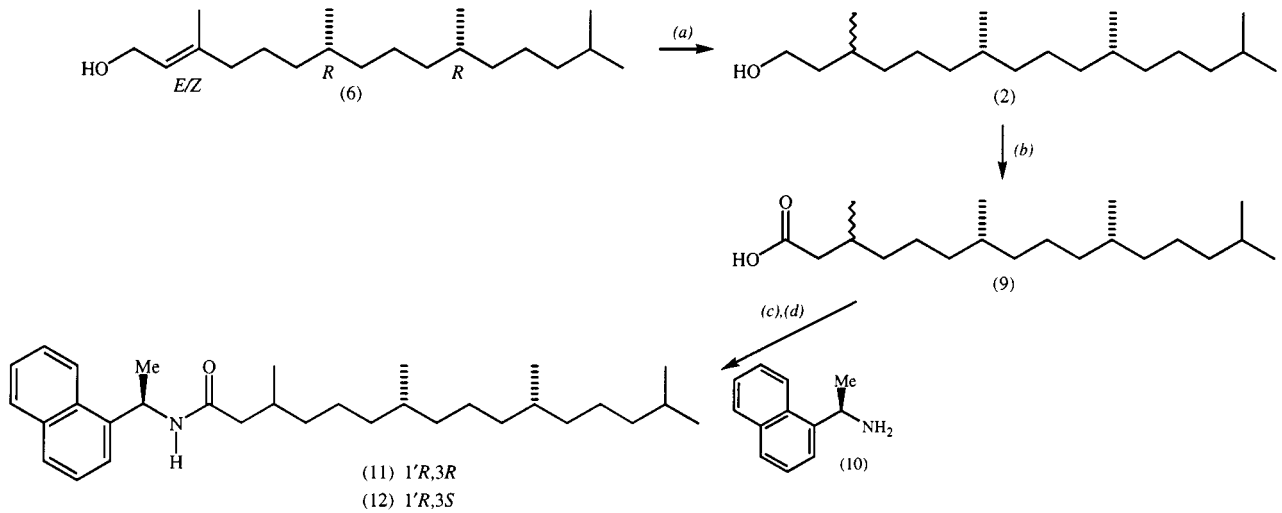
stretches at 3529, 3410 and 1677 cm^{-1}, consistent with a primary amide, as well as peaks at δ 175·5 in the ^{13}C n.m.r. spectra, again indicative of an amide carbonyl. Independent synthesis of the amides from 3*RS* phytanic acid (9) (by treatment of the derived acid chloride with 33% aqueous ammonia) gave material indistinguishable (by t.l.c. and ^{1}H n.m.r.) from that derived from the attempted amide hydrolysis.

Hydrolysis of the primary amides (15) and (16) was achieved with 10 M sulfuric acid in tetrahydrofuran at reflux; however, yields for this reaction, and time for completion of the reaction, were variable ranging from 30–70% and 6–14 days.

Hydrolysis of (11) and (12) under milder acidic conditions (6 M HCl, EtOH), and with 'anhydrous t-butoxide',[17] both failed to generate the desired pro-



Scheme 1. Proposed route to the four diastereomers of GMPE from resolved phytanol.



Scheme 2. *(a)* Raney nickel/H_2, EtOH. *(b)* CrO_3, H_2O, 2 : 1 acetone/acetic acid. *(c)* Oxalyl chloride, catalytic dimethylformamide, diethyl ether. *(d)* (*R*)-(+)-1-(1-Naphthyl)ethylamine (10), triethylamine, CH_2Cl_2.

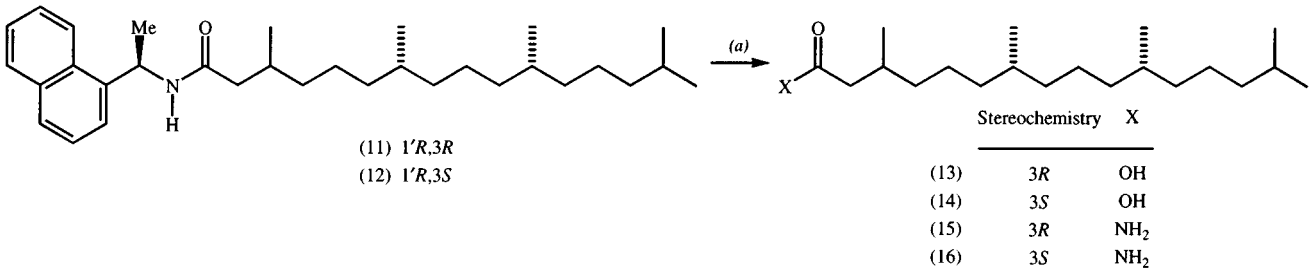
ducts, while attempted reductive removal with DIBAL[18] returned only starting material. The method of Evans and coworkers[19]* for the hydrolysis of secondary amides, involving the *in situ* formation of the active *N*-nitrosamide followed by hydrolysis with lithium hydroperoxide, gave the best and most reproducible method (50% overall yield) for the preparation of the acids (13) and (14) (Scheme 4).

The material obtained from this reaction was difficult to purify by conventional methods, and analytically pure material was obtained only after h.p.l.c. These materials were identical in all respects (excepting optical rotation) to the mixture of diastereomeric acids (9) obtained by oxidation of phytanol (2) discussed above.

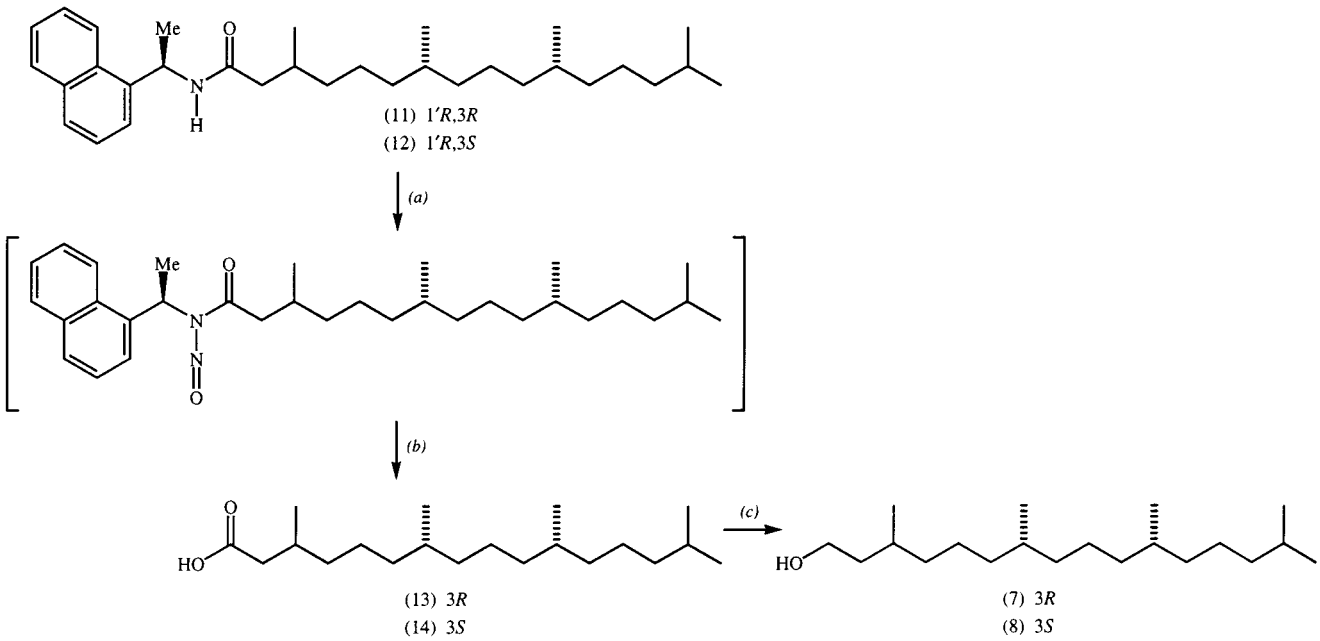
3*R* and 3*S* Phytanic acids (13) and (14), respectively, were reduced to the corresponding phytanols (7) and (8) with $LiAlH_4$. The reduction proceeded in excellent yield, with spectroscopic data and optical rotations in agreement with those reported for materials synthesized by asymmetric hydrogenation of phytol.[13]

Bromination of the phytanols (7) and (8) using aqueous 48% HBr generated the phytanyl bromides (17) and (18) (81 and 76% respectively) required for the coupling reactions with the protected glycerols (4) and (5)[12] (Scheme 5). Etherification of the protected glycerols (4) and (5) was effected by using KOH in refluxing toluene under Dean–Stark conditions, to furnish the ethers (19)–(22) in approximately 55% yield. The compounds exhibited spectroscopic data in full accord with their proposed structures.

Acid hydrolysis of the ketals using a catalytic amount of acid in 10 : 1 dioxan/water afforded the four isomers of glycerol monophytanyl ether (23)–(26) in excellent yield (90% average yield). Spectroscopic data for each compound were in full accord with their proposed structures, with compound (23) having spectroscopic data in agreement with those reported previously for this material.[11] The optical rotation of material in this work ($[\alpha]_D = +0{\cdot}49$) was different to that reported ($[\alpha]_D = -0{\cdot}94$).



Scheme 3. *(a)* Conc. H_2SO_4, tetrahydrofuran, reflux 14 days. Preferentially form (15) and (16) instead of (13) and (14).



Scheme 4. *(a)* Sodium nitrite, 2 : 1 acetic anhydride/acetic acid, 0°C to room temperature. *(b)* $LiOH/H_2O_2$, 3 : 1 tetrahydrofuran/H_2O, 0°C to room temperature. *(c)* $LiAlH_4$, tetrahydrofuran.

* Many thanks to Dr Percy H. Carter for additional information relating to this reaction.

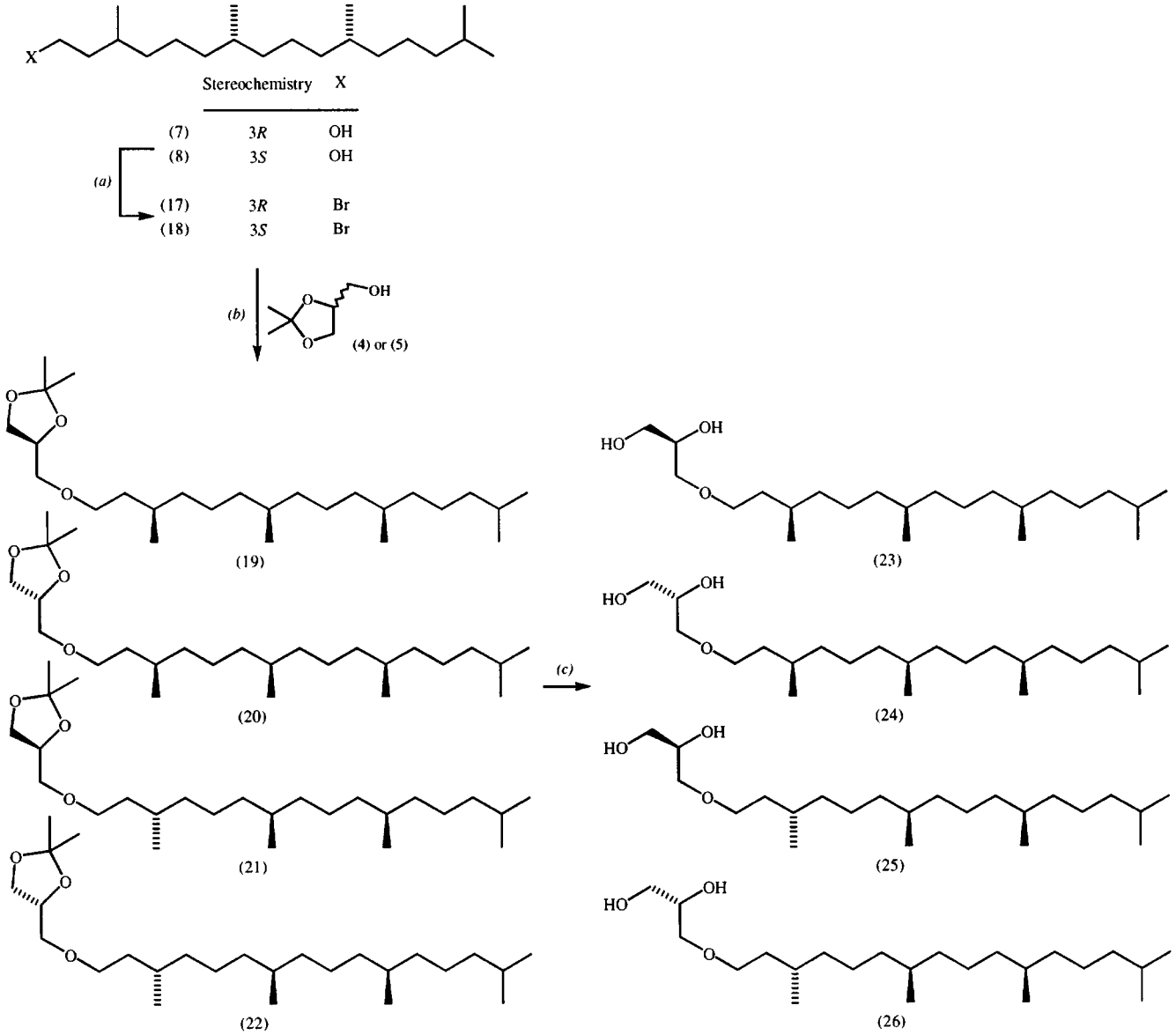
Conclusion

The total synthesis of the four diastereomers of glycerol monophytanyl ether (3) in chirally pure form has been achieved in eight steps from commercially available phytol (6) in 7–13% overall yield. Identification of a reproducible route to both diastereomers of phytanol also allows the preparation of other phytanylated membrane-forming materials, such as 1,2-diphytanyl-*sn*-glycerol[20] and its phosphatidylcholine derivative,[21] in chirally pure form. Studies on the formation and properties of membranes from these and related materials are currently under way.[22]

Experimental

^{1}H and ^{13}C n.m.r. spectra were recorded on Bruker AC-200 (200 MHz) and AMX-400 (400 MHz) spectrometers with samples dissolved in $CDCl_3$ containing tetramethylsilane as internal reference. Electron ionization (e.i.) mass spectra were recorded on a modified Kratos mass spectrometer calibrated with perfluorokerosene (PFK). Chemical ionization (c.i.) mass spectra were recorded on an AEI Ms30 spectrometer with methane as the ionizing gas. Microanalyses were performed by the Campbell Microanalytical Laboratory, Department of Chemistry, University of Otago, Dunedin, New Zealand. DEPT experiments were conducted to assist with the assignment of ^{13}C n.m.r. spectra. Purifications were achieved by flash chromatography on silica gel (0·040–0·063 mm) unless otherwise stated. Retention factors (R_F) of compounds were determined by thin-layer chromatography on Merck 60 F_{254} plates. Optical rotations were recorded on an Optical Activity Polaar 2001 spectrometer at ambient temperature.

All reactions were performed under an atmosphere of dry nitrogen. Anhydrous tetrahydrofuran was obtained by distillation from sodium benzophenone ketyl immediately prior to use. All other solvents were distilled and all reagents were commercial grade.



Scheme 5. *(a)* 48% HBr, conc. H_2SO_4. *(b)* *R* or *S* Isopropylideneglycerol (4) or (5), KOH, toluene. *(c)* Catalytic HCl, 10 : 1 dioxan/H_2O.

*(3*RS*,7*R*,11*R*)-3,7,11,15-Tetramethylhexadecan-1-ol (2)*

A solution of phytol (49·3 g, 167 mmol) in ethanol (250 ml) was stirred under an atmosphere of hydrogen over Raney nickel (10·3 g, 50% slurry in water) for 60 h. The catalyst was removed by filtration through Celite^{R} and the filtrate concentrated under reduced pressure to give (2) (49·3 g, 100%) as a colourless oil: ^{1}H n.m.r. (200 MHz) δ ($CDCl_3$) 0·83–0·91, m, 15H; 1·03–1·59, m, 24H; 3·64–3·72, m, 2H, H1. ^{13}C n.m.r. (50 MHz) δ ($CDCl_3$) 19·59–19·74, overlapping signals, non-terminal CH_3 groups; 22·6, CH_3; 22·7, CH_3; 24·4, CH_2; 24·5, CH_2; 24·8, CH_2; 28·0, CH; 29·5, CH; 32·8, CH; 37·3–37·5, overlapping signals, CH_2 groups; 39·4, CH_2; 39·9, CH_2; 40·0, CH_2; 61·2, CH_2OH. Mass spectrum (c.i.) *m/z* 298 (M^+, 100%), 281 (41), 226 (15), 211 (22), 196 (26), 183 (28), 168 (30), 155 (31).

*(3*RS*,7*R*,11*R*)-3,7,11,15-Tetramethylhexadecanoic Acid (9)*

Chromium trioxide (0·80 g, 8·0 mmol) in water (1 ml) was added dropwise to a solution of (2) (1 g, 3·3 mmol) in acetone (40 ml) and acetic acid (20 ml) at room temperature. Stirring was continued for 1 h and water (50 ml) was added, followed by enough solid sodium metabisulfite to destroy the excess oxidant. The majority of the solvent was removed in vacuum, the residue taken up in water (60 ml) and extracted with diethyl ether (3×20 ml). The combined organic extracts were washed with water (50 ml), dried (Na_2SO_4) and the solvent was removed under reduced pressure to give the crude material as a pale yellow oil. Purification with 2:1 hexane/diethyl ether as eluent gave (9) (625 mg, 61% yield) as a colourless oil: R_F 0·70 (1:1 hexane/diethyl ether). ν_{max} 1709 cm^{-1}. ^{1}H n.m.r. (200 MHz) δ ($CDCl_3$) 0·83–0·88, m, 12H; 0·97, d, *J* 6·6 Hz, 3H; 1·03–1·40, m, 20H; 1·49, sept, *J* 6·5 Hz, 1H, H3; 1·90–2·05, m, 1H; 2·08–2·20, m, 1H; 2·31–2·41, m, 1H. Mass spectrum (c.i.) *m/z* 312 (M^+, 3%), 157 (21), 139 (18), 111 (25), 97 (46), 87 (87), 71 (69), 57 (93), 43 (100).

*(3*RS*,7*R*,11*R*)-3,7,11,15-Tetramethyl*-N-*[(1′*R*)-1′-(1-naphthyl)ethyl]hexadecanamide*

N,N-Dimethylformamide (1 drop) followed by oxalyl chloride (482 mg, 3·80 mmol) was added to a solution of (9) (1·08 g, 3·46 mmol) in diethyl ether (20 ml). The reaction mixture was stirred for 1 h under nitrogen at room temperature, the solvent removed under reduced pressure and the residue dissolved in dichloromethane (15 ml). A solution of (*R*)-(+)-1-(1-naphthyl)ethylamine (0·65 g, 3·8 mmol) in dichloromethane (5 ml) and triethylamine (1·01 g, 10 mmol) was added and the stirring continued under nitrogen for an additional 2 h. The solvent was removed under reduced pressure, the residue dissolved in hexane and filtered through a short pad of Celite^{R}. After removal of the solvent in vacuum, the crude product was purified by using 2:1 hexane/diethyl ether as eluent to provide the title 3*RS* amide (1·41 g, 87·8%) as a pale yellow gum: R_F 0·36 (2:1 hexane/diethyl ether). ^{1}H n.m.r. (200 MHz) δ ($CDCl_3$) 0·82–0·88, m, 15H; 1·00–1·40, m, 20H; 1·51, sept, *J* 6·5 Hz, 1H, H3; 1·68, d, *J* 6·8 Hz, 3H; 1·90–2·05, m, 2H; 2·14–2·24, m, 1H; 5·63, d, *J* 8·1 Hz, 1H; 5·95, m, 1H; 7·38–7·55, m, 4H; 7·80, d, *J* 8·0 Hz, 1H; 7·87, d, *J* 8·0 Hz, 1H; 8·14, d, *J* 8·1 Hz, 1H. ^{13}C n.m.r. (50 MHz) δ ($CDCl_3$) 19·56, 19·66, 20·46, 22·53, 22·62, 24·25, 24·37, 24·71, 27·89, 30·85, 32·70, 37·07, 37·22, 37·33, 37·38, 39·29, 44·35, 44·61, 122·47, 123·53, 125·03, 125·82, 126·47, 128·33, 128·63, 131·13, 133·87, 138·17, 171·37. Mass spectrum (c.i.) *m/z* 466 (M^+, 100%), 312 (10), 155 (21).

*(3*R*,7*R*,11*R*)-3,7,11,15-Tetramethyl*-N-*[(1′*R*)-1′-(1-naphthyl)ethyl]hexadecanamide (11) and its (3*S*,7*R*,11*R*) Isomer (12)*

H.p.l.c. of the 3*RS* amide (described in the previous paragraph) with 8·8:1·2 hexane/ethyl acetate as eluent (Whatman Partisil 10 column, 13·5 ml/min flow rate) gave firstly the 3*R* isomer (11): R_F 0·32 (2:1 hexane/diethyl ether) (Found: C, 82·4; H, 10·9; N, 2·8. Calc. for $C_{32}H_{51}NO$: C, 82·5; H, 11·0; N, 3·0%). $[\alpha]_D$ +28·7° (*c*, 2·6 in $CHCl_3$) (lit.[13] $[\alpha]_D^{27}$ +40·75°). ^{1}H n.m.r., ^{13}C n.m.r. identical to those of the 3*RS* amide. Further elution provided the 3*S* isomer (12): R_F 0·27 (2:1 hexane/diethyl ether) (Found: C, 82·5; H, 11·1; N, 2·8. Calc. for $C_{32}H_{51}NO$: C, 82·5; H, 11·0; N, 3·0%). $[\alpha]_D$ +28·4° (*c*, 2·3 in $CHCl_3$) (lit.[13] $[\alpha]_D^{27}$ +43·39°). ^{1}H n.m.r., ^{13}C n.m.r. identical to those of the 3*RS* amide.

*(3*R*,7*R*,11*R*)-3,7,11,15-Tetramethylhexadecanamide (15)*

Concentrated H_2SO_4 (5 ml) was added to a solution of (11) (0·98 g, 2·10 mmol) in tetrahydrofuran (25 ml) and the solution heated at reflux for 14 days under nitrogen. After cooling to room temperature the solvent was removed under reduced pressure, water (100 ml) was added and the solution extracted with dichloromethane (3×75 ml). The combined organic extracts were washed with brine (175 ml), dried (Na_2SO_4) and filtered through a pad of silica. After removal of the solvent in vacuum the resultant dark brown oil was purified by using 3:1 to 1:1 hexane/ethyl acetate as eluent to provide (15) (273 mg, 41·7%) as a rust-coloured wax. Analytically pure *material* was obtained by h.p.l.c. purification with 1:1 hexane/ethyl acetate as eluent (Whatman Partisil 10 column, 13·5 ml/min flow rate): R_F 0·15 (1:1 hexane/ethyl acetate) (Found: C, 76·9; H, 13·2; N, 4·4. $C_{20}H_{41}NO$ requires C, 77·1; H, 13·3; N, 4·5%). $[\alpha]_D$ +2·4° (*c*, 3·3 in $CHCl_3$). ν_{max} 3529, 3410, 1677 cm^{-1}. ^{1}H n.m.r. (200 MHz) δ ($CDCl_3$) 0·83–0·87, m, 12H; 0·95, d, *J* 6·2 Hz, 3H; 1·06–1·40, m, 22H; 1·52, sept, *J* 6·6 Hz, 1H, H3; 1·90–2·28, m, 1H; 5·70, br s, 2H, NH. ^{13}C n.m.r. (50 MHz) δ ($CDCl_3$) 19·58, CH_3; 19·65, CH_3; 19·73, CH_3; 22·61, CH_3; 22·70, CH_3; 24·39, CH_2; 24·45, CH_2; 24·78, CH_2; 27·96, CH; 30·74, CH; 32·76, CH; 37·14, CH_2; 37·21, CH_2; 37·28, CH_2; 37·39, CH_2; 37·44, CH_2; 39·36, CH_2; 43·61, CH_2; 175·53, amide CO. Mass spectrum (c.i.) *m/z* 312 ($M+H^+$, 100%).

*(3*S*,7*R*,11*R*)-3,7,11,15-Tetramethylhexadecanamide (16)*

The *amide* (16) was prepared in an identical fashion to that used for the preparation of (15), with (12) as the starting material to afford (16) (Found: C, 77·1; H, 13·2; N, 4·4. $C_{20}H_{41}NO$ requires C, 77·1; H, 13·3; N, 4·5%). $[\alpha]_D$ −2·2° (*c*, 2·2 in $CHCl_3$). R_F, i.r. ^{1}H n.m.r., and ^{13}C n.m.r. data were identical to those of (15).

*(3*R*,7*R*,11*R*)-3,7,11,15-Tetramethylhexadecanoic Acid (13)*

A solution of (11) (595 mg, 1·28 mmol) in a 2:1 mixture of acetic anhydride/acetic acid (20 ml) was chilled to 0°C and sodium nitrite (881 mg, 12·8 mmol) was added. The reaction mixture was allowed to warm to room temperature and stirred under nitrogen for 4 h after which time t.l.c. (1:1 hexane/diethyl ether) showed the starting material had been consumed. After adding diethyl ether (25 ml) and chilling the solution to 0°C, saturated $NaHCO_3$ (50 ml) was added and the mixture stirred for 10 min. Diethyl ether (50 ml) and saturated $NaHCO_3$ (25 ml) were added and the organic layer was separated, washed with saturated $NaHCO_3$ (3×50 ml), brine (50 ml), dried (Na_2SO_4) and the solvent removed in vacuum. The crude material was used immediately in the next reaction. The dark yellow liquid was dissolved in tetrahydrofuran (15 ml) and water (5 ml), chilled to 0°C and lithium hydroxide (306 mg, 12·8 mmol) added followed by 30% hydrogen peroxide (2·6 ml). The reaction was allowed to warm to room temperature and stirring continued under nitrogen for 24 h after which time sodium metabisulfite (2·7g, 10% excess) was added to the recooled solution. Water (25 ml) was added and the reaction mixture stirred for 5 min at 0°C after

which time the tetrahydrofuran was removed under reduced pressure and water (20 ml) added. The aqueous phase was extracted with ethyl acetate (3×30 ml), the combined organic extracts were washed with brine (50 ml), dried (Na_2SO_4) and the solvent was removed in vacuum to give a yellow liquid. The residue was partially purified with 2 : 1 hexane/diethyl ether as eluent to give a mixed polar fraction containing the acid along with an unknown impurity (373 mg, 93%). Analytically pure material was obtained by h.p.l.c. purification (Whatman Partisil 10 column, 13·5 ml/min flow rate) with a mixture of 0·2% acetic acid and 3% ethyl acetate in hexane to give (13) (335 mg, 42·1%) as a pale yellow oil: $[\alpha]_D$ +4·6° (*c*, 6·7 in $CHCl_3$) (lit.[13] $[\alpha]_D^{27}$ +4·50°). R_F, i.r. and ^{1}H n.m.r. data were identical to those of (9).

*(3*S,*7*R,*11*R*)-3,7,11,15-Tetramethylhexadecanoic Acid (14)*

The acid (14) was prepared in an identical manner to that used for the preparation of (13), with (12) (1·19 g, 2·55 mmol) as starting material to afford (14) (405 mg, 50·7%): $[\alpha]_D$ −4·9° (*c*, 7·8 in $CHCl_3$) (lit.[13] $[\alpha]_D^{27}$ −4·45°). R_F, ^{1}H n.m.r., and i.r. data were identical to those for (9).

*(3*R,*7*R,*11*R*)-3,7,11,15-Tetramethylhexadecan-1-ol (7)*

A Soxhlet apparatus containing $LiAlH_4$ (1·25 g, 32·9 mmol) immersed in dry tetrahydrofuran (10 ml) was attached to a round-bottomed flask charged with (13) (333 mg, 1·07 mmol) in tetrahydrofuran (30 ml). The solution was heated at reflux under nitrogen for 3 h after which time the contents were cooled to room temperature and a mixture of tetrahydrofuran and water (5 ml, 3 : 1) was added. The solvent was removed under reduced pressure, 3 M HCl (50 ml) was added and the product extracted into diethyl ether (3×30 ml). The combined organic extracts were washed with brine (25 ml), dried (Na_2SO_4) and the solvent was removed under reduced pressure to give the crude material as a clear yellow oil. Purification with 95 : 5 hexane/ethyl acetate as eluent provided (7) (312 mg, 98%) as a clear oil: R_F 0·29 (9 : 1 hexane/ethyl acetate). $[\alpha]_D$ +2·9° (*c*, 6·2 in $CHCl_3$) (lit.[13] $[\alpha]_D^{27}$ +2·70°). ^{1}H n.m.r. and ^{13}C n.m.r. spectra were identical to those of (2).

*(3*S,*7*R,*11*R*)-3,7,11,15-Tetramethylhexadecan-1-ol (8)*

The alcohol (8) was prepared in an identical manner to that used for the preparation of (7), with (14) (395 mg, 1·26 mmol) as the starting material to afford (8) (368 mg, 97·6%): $[\alpha]_D$ −2·3° (*c*, 8·3 in $CHCl_3$) (lit.[13] $[\alpha]_D^{27}$ −2·72°). ^{1}H n.m.r. and ^{13}C n.m.r. spectra were identical to those of (2).

*(3*R,*7*R,*11*R*)-1-Bromo-3,7,11,15-tetramethylhexadecane (17)*

A mixture of (7) (312 mg, 1·05 mmol), aqueous 48% HBr (50 ml) and conc. H_2SO_4 (0·5 ml) were heated at reflux under nitrogen for 24 h. Upon cooling to room temperature the mixture was extracted with hexane (3×25 ml). The combined organic extracts were dried (Na_2SO_4) and the solvent was removed in vacuum to give the crude product as a dark brown oil. Purification with hexane as eluent afforded (17) (306 mg, 81·0%) as a colourless oil: R_F 0·75 (hexane). $[\alpha]_D$ −3·3° (*c*, 6·1 in $CHCl_3$) (lit.[13] $[\alpha]_D^{27}$ −2·90°). ^{1}H n.m.r. (200 MHz) δ ($CDCl_3$) 0·82–0·90, m, 15H; 1·0–1·95, m, 24H; 3·34–3·50, m, 2H, H 1. ^{13}C n.m.r. (50 MHz) δ ($CDCl_3$) 18·95, CH_3; 19·00, CH_3; 19·71, CH_3; 22·64, CH_3; 22·73, CH_3; 24·23, CH_2; 24·48, CH_2; 24·82, CH_2; 27·98, CH; 29·71, CH; 31·68, CH; 32·12, CH_2; 32·78, CH; 36·80 CH_2; 36·85, CH_2; 37·31, CH_2; 37·40, CH_2; 39·39, CH_2; 40·06, CH_2; 40·13, CH_2. Mass spectrum (c.i.) *m/z* 361 ($M+H^+$, 29%), 359 (M^+, 26), 347 (15), 281 (71), 223 (34), 211 (57), 197 (54), 183 (60), 169 (71), 155 (76), 141 (67), 127 (95), 113 (100).

*(3*S,*7*R,*11*R*)-1-Bromo-3,7,11,15-tetramethylhexadecane (18)*

The *bromide* (18) was prepared in an identical fashion to that used for the preparation of (17), with (8) as the starting material to afford (18) (385 mg, 76%) (Found: C, 66·4; H, 11·6. $C_{20}H_{41}Br$ requires C, 66·5; H, 11·4%). $[\alpha]_D$ +3·1° (*c*, 7·7 in $CHCl_3$). R_F, ^{1}H n.m.r. and ^{13}C n.m.r. data were identical to those for (17).

(S)*-2,2-Dimethyl-1,3-dioxolan-4-methanol (5)*

The glycerol derivative (5) was prepared from 1,2:5,6-di-*O*-isopropylidene-D-mannitol by established procedures.[12] The material was used, without further purification, in subsequent reactions. ^{1}H n.m.r. (200 MHz) δ ($CDCl_3$) 1·37, s, 3H, CH_3; 1·43, s, 3H, CH_3; 2·16, br s, 1H, OH; 3·58, dd, *J* 11·7, 5·0 Hz, 1H, H 1; 3·73, dd, *J* 11·7, 3·8 Hz, 1H, H 1; 3·78, dd, *J* 8·2, 6·6 Hz, 1H, H 3; 4·03, dd, *J* 8·2, 6·7 Hz, 1H, H 3; 4·18–4·29, m, 1H (glycerol numbering has been used for the n.m.r. assignments).

*(2*S*)-1,2-Isopropylidene-3-[(3′*R,*7′*R,*11′*R*)-3′,7′,11′,15′-tetramethylhexadecyl]glycerol (19)*

A round-bottomed flask containing a solution of (5) (62·5 mg, 0·47 mmol) and powdered KOH (188 mg, 4·7 mmol) in toluene (15 ml) was fitted with a Dean–Stark apparatus and heated at reflux under nitrogen for 1 h. After cooling to room temperature, (17) (217 mg, 0·60 mmol) in toluene (5 ml) was added and the mixture heated at reflux for 24 h. The cooled solution was diluted with diethyl ether (35 ml), washed sequentially with water (25 ml), 0·25 M HCl (25 ml), 2·5% aqueous $NaHCO_3$ (25 ml), water (25 ml), brine (25 ml), dried (Na_2SO_4) and the solvent removed to give the crude product as a yellow oil. Purification by using 97 : 3 hexane/ethyl acetate afforded (19) (120 mg, 61·5%) as a clear *oil*: R_F 0·55 (9 : 1 hexane/ethyl acetate) (Found: C, 75·9; H, 12·7. $C_{26}H_{52}O_3$ requires C, 75·7; H, 12·7%). $[\alpha]_D$ +9·5° (*c*, 9·0 in $CHCl_3$). ^{1}H n.m.r. (200 MHz) δ ($CDCl_3$) 0·82–0·88, m, 15H; 1·02–1·68, m, 24H; 1·36, s, 3H; 1·42, s, 3H; 3·37–3·56, m, 4H, H 3, H 1′; 3·72, dd, *J* 8·2, 6·4 Hz, 1H, H 1; 4·06, dd, *J* 8·2, 6·4 Hz, 1H, H 1; 4·26, dddd (apparent pentuplet), *J* 6·4 Hz, 1H, H 2. ^{13}C n.m.r. (50 MHz) δ ($CDCl_3$) 19·69, CH_3; 22·63, CH_3; 22·72, CH_3; 24·36, CH_2; 24·47, CH_2; 24·80, CH_2; 25·43, CH_3; 26·78, CH_3; 27·98, CH; 29·86, CH; 32·78, CH; 36·52, CH_2; 36·60, CH_2; 37·40, CH_2; 39·37, CH_2; 66·96, **C**H_2OC$(CH_3)_2$; 70·19, CH_2O**C**$H_2(C_{19}H_{39})$; 71·86, **C**H_2O$CH_2(C_{19}H_{39})$; 74·76, CH; 109·34, quaternary. Mass spectrum (e.i.) *m/z* 398 ($M-CH_3^+$, 51%), 133 (19), 127 (10), 123 (17), 111 (25), 101 (100).

*(2*R*)-2,3-Isopropylidene-1-[(3′*R,*7′*R,*11′*R*)-3′,7′,11′,15′-tetramethylhexadecyl]glycerol (20)*

This *compound* was prepared in an identical fashion to that used for the preparation of (19), with (*R*)-2,2-dimethyl-1,3-dioxolan-4-methanol (67·4 mg, 0·51 mmol) and (17) (211 mg, 0·58 mmol) as starting materials to afford (20) (120 mg, 57·1%) (Found: C, 75·9; H, 13·0. $C_{26}H_{52}O_3$ requires C, 75·7; H, 12·7%). $[\alpha]_D$ −6·4° (*c*, 8·8 in $CHCl_3$). R_F, ^{1}H n.m.r. and ^{13}C n.m.r. data were identical to those for the 2*S* isomer (19).

*(2*S*)-1,2-Isopropylidene-3-[(3′*S,*7′*R,*11′*R*)-3′,7′,11′,15′-tetramethylhexadecyl]glycerol (21)*

This compound was prepared in an identical fashion to that used for the preparation of (19), with (5) (85·0 mg, 0·64 mmol) and (18) (278 mg, 0·77 mmol) as starting materials to afford (21) (140 mg, 52·8%): $[\alpha]_D$ +6·4° (*c*, 12·7 in $CHCl_3$). R_F, ^{1}H n.m.r. and ^{13}C n.m.r. data were identical to those of (19).

*(2*R*)-2,3-Isopropylidene-1-[(3′*S*,7′*R*,11′*R*)-3′,7′,11′,15′-tetramethylhexadecyl]glycerol (22)*

This *compound* was prepared in an identical fashion to that used for the preparation of (19), with (*R*)-2,2-dimethyl-1,3-dioxolan-4-methanol (98·0 mg, 0·74 mmol) and (18) (315 mg, 0·87 mmol) as starting materials to afford (22) (128 mg, 41·8%) (Found: C, 75·8; H, 12·5. $C_{26}H_{52}O_3$ requires C, 75·7; H, 12·7%). $[\alpha]_D$ −8·7° (*c*, 11·2 in $CHCl_3$). R_F, ^{1}H n.m.r. and ^{13}C n.m.r. data were identical to those of (19).

*(2*R*)-3-[(3′*R*,7′*R*,11′*R*)-3′,7′,11′,15′-Tetramethylhexadecyl]glycerol (23)*

Water (1 ml) and conc. HCl (0·1 ml) were added to a solution of (19) (102 mg, 0·25 mmol) in 1,4-dioxan (10 ml). The mixture was heated at reflux under nitrogen for 4 h after which time the solvent was removed under reduced pressure. The residue was diluted with ethyl acetate (25 ml), washed with water (25 ml), 5% aqueous $NaHCO_3$ (25 ml), brine (25 ml), dried (Na_2SO_4) and the solvent removed in vacuum to give the crude material as a colourless viscous oil. Purification with diethyl ether as eluent gave (23) (88 mg, 96%) as a colourless viscous oil: R_F 0·39 (diethyl ether). $[\alpha]_D$ +0·5° (*c*, 4·1 in $CHCl_3$) (lit.[11] $[\alpha]_D$ −0·94°). ν_{max} 3384, 2953, 2925, 2868, 1463, 1377, 1119, 1047 cm^{-1}. ^{1}H n.m.r. (200 MHz) δ ($CDCl_3$) 0·81–0·88, m, 15H; 0·98–1·65, m, 24H; 2·66, br s, 2H, OH; 3·45–3·88, m, 7H. ^{13}C n.m.r. (50 MHz) δ ($CDCl_3$) 19·61, CH_3; 19·67, CH_3; 19·73, CH_3; 22·61, CH_3; 22·70, CH_3; 24·34, CH_2; 24·45, CH_2; 24·78, CH_2; 27·95, CH; 29·87, CH; 32·75, CH; 36·50, CH_2; 36·58, CH_2; 37·36, CH_2; 37·42, CH_2; 39·33, CH_2; 64·20, $CH_2O\mathbf{C}H_2(C_{19}H_{39})$; 70·13, CH_2OH; 70·48, CHOH; 72·42, $\mathbf{C}H_2OCH_2(C_{19}H_{39})$. Mass spectrum (c.i.) *m/z* 373 ($M+H^+$, 100%).

*(2*S*)-1-[(3′*R*,7′*R*,11′*R*)-3′,7′,11′,15′-Tetramethylhexadecyl]glycerol (24)*

This *compound* was prepared in an identical fashion to that used for the preparation of (23), with (20) (97 mg, 0·24 mmol) as starting material to afford (24) (78 mg, 89%) (Found: C, 73·5; H, 13·0. $C_{23}H_{48}O_3.\frac{1}{6}H_2O$ requires C, 73·5; H, 13·0%). $[\alpha]_D$ +4·5° (*c*, 3·9 in $CHCl_3$). R_F, ^{1}H n.m.r. and ^{13}C n.m.r. data were identical to those of (23).

*(2*R*)-3-[(3′*S*,7′*R*,11′*R*)-3′,7′,11′,15′-Tetramethylhexadecyl]glycerol (25)*

This *compound* was prepared in an identical fashion to that used for the preparation of (23), with (21) (125 mg, 0·30 mmol) as starting material to afford (25) (109 mg, 96%) (Found: C, 74·1; H, 13·2. $C_{23}H_{48}O_3$ requires C, 74·1; H, 13·0%). $[\alpha]_D$ −2·0° (*c*, 10·2 in $CHCl_3$). R_F, ^{1}H n.m.r. and ^{13}C n.m.r. data were identical to those of (23).

*(2*S*)-1-[(3′*S*,7′*R*,11′*R*)-3′,7′,11′,15′-Tetramethylhexadecyl]glycerol (26)*

This *compound* was prepared in an identical fashion to that used for the preparation of (23), with (22) (112 mg, 0·27 mmol) as starting material to afford (26) (87 mg, 86%) (Found: C, 73·9; H, 13·1. $C_{23}H_{48}O_3$ requires C, 74·1; H, 13·0%). $[\alpha]_D$ −1·3° (*c*, 8·2 in $CHCl_3$). R_F, ^{1}H n.m.r. and ^{13}C n.m.r. data were identical to those of (23).

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