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A Chemoenzymatic Access to D- and L-Sphingosines Employing Hydroxynitrile Lyases

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Abstract—A chemoenzymatic access to D- or L-sphingosines is presented comprising of a total synthesis of the L-*threo*-isomer and formal syntheses of the other three isomers. Key to the development of a general synthetic strategy has been the use of enantiocomplementary hydroxynitrile lyases (Hnls) to yield an enantiomeric pair of starting materials. The (*S*)-Hnl from *Hevea brasiliensis* has been used to prepare L-*threo*-sphingosine in 14 steps and an overall 12% yield. Application of the (*R*)-Hnl from *Prunus amygdalus* formally allows synthesis of D-*threo*- and D-*erythro*-sphingosines.

Introduction

Sphingosines are long chain 2-amino-1,3 diols which exist in four stereoisomeric forms. D-*erythro*-sphingosine has been reported as an inhibitor of protein kinase-C[1] (PKC), an enzyme which mediates a number of cellular responses.[1e] L-*erythro*- and the more commonly occurring D-*erythro*-sphingosine are commercially available. Sphingosines are the basic components of a much larger class of compounds known as glycosphingolipids, which exhibit a number of biological functions including HIV binding[2] and 'biological markers' for the detection of cancer.[3] Furthermore, these compounds are of considerable biological significance in the area of cellular interactions.[4] To date numerous syntheses of sphingosines have been reported and they can be divided into those prepared from chiral pool materials (such as carbohydrates or L-serine), asymmetric reactions or by chemoenzymatic means.[5a] The chemoenzymatic methods are rarer.[5] Of all the reports, the syntheses of D-*erythro*-sphingosine by Randunz et al.,[6a] Polt et al.,[6b] Ito et al.[6c,d] and Solladié-Cavallo et al.[6e] are noteworthy for their efficiency (35–50% yield) and brevity (4–6 steps). Recently, Enders et al. have reported an elegant diastereoselective and enantioselective method to both D-*erythro*- and L-*threo*-sphingosine.[7]

The high chemo- and stereoselectivity of biocatalysts has promoted their use in the synthesis of natural products. In this respect, production of enantiomerically pure cyanohydrins by hydroxynitrile lyases (Hnls) is of significant interest in the synthesis of bioactive compounds.[8,9] These enzymes are available with complementary selectivities, i.e. the (*R*)-Hnls from *Prunus amygdalus*[10] (PaHnl) and *Linum usitatissium*[11] yield (*R*)-cyanohydrins whilst the (*S*)-Hnls from *Hevea brasiliensis*[12] (HbHnl), *Sorghum bicolor*[13] and *Manihot esculenta*[14] yield the corresponding (*S*)-cyanohydrins.

We wish to present a new method which allows access to L-*threo*- and *erythro*-sphingosine using the HbHnl, as well as the D-isomers of sphingosine using the PaHnl. In each case the stereochemistry of the secondary hydroxyl center of the sphingosine is determined by the selectivity of the Hnl (Scheme 1). To date a chemoenzymatic method which employs such a general synthetic sequence has yet to be developed.

Results and Discussion

L-Series sphingosines

Enantiopure cyanohydrin (*S*)-**2** (99% e.e., 90% yield) was prepared from (*E*)-2-octenal (Scheme 2) employing the (*S*)-selective HbHnl,[15,16] followed by protection of the free cyanohydrin. Transformation[17] of the protected cyanohydrin (*S*)-**2** gave the diastereomeric amine **3** (Scheme 2) (57% d.e. by ^{1}H and ^{13}C NMR spectra).[18]

Deprotection of **3** with tetrabutylammonium fluoride (TBAF) yielded amine **4** (57% d.e. by ^{1}H and ^{13}C NMR spectroscopy) and further treatment of the diastereomeric mixture with 1,1-carbonyldiimidazole followed by separation on silica gel column chromatography gave diastereomerically pure **5** and **6** (>99% e.e., >99% d.e.) (Scheme 3).

Keywords: hydroxynitrile lyases; sphingosines; selective coupling.

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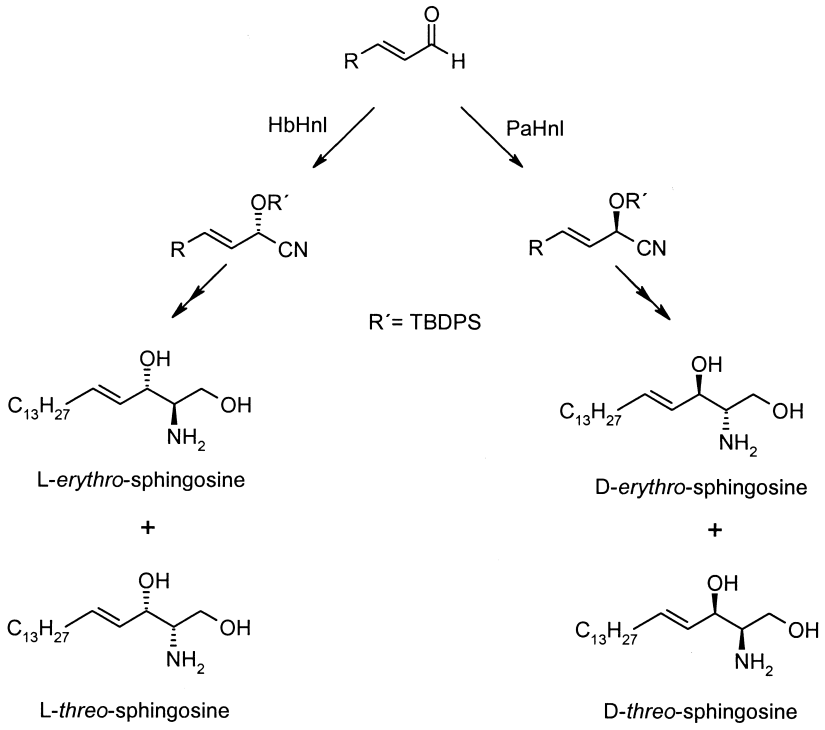
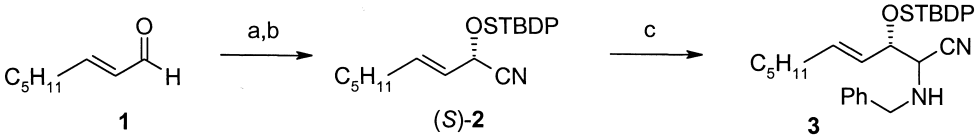


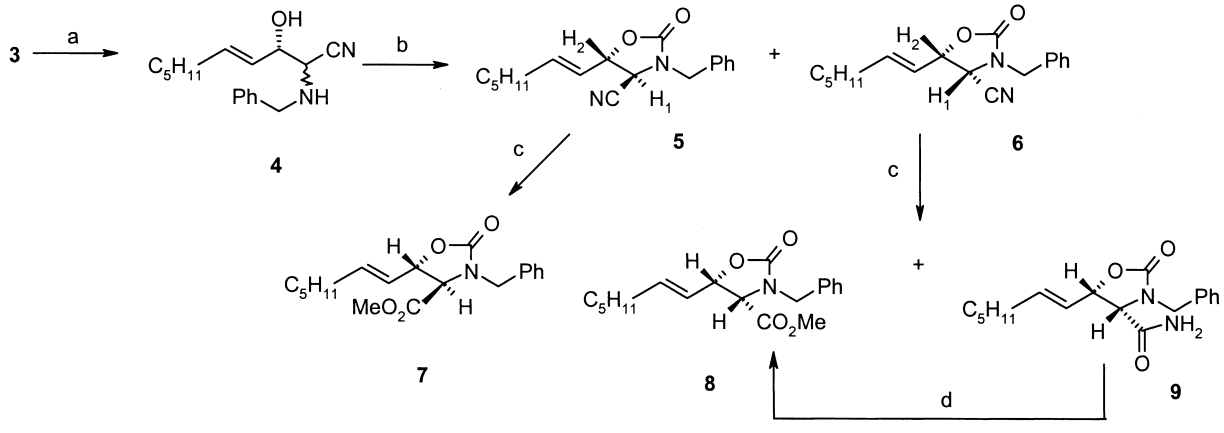
Scheme 1. Application of Hnls to the synthesis of the four isomers of C_{18} sphingosine.

The configurations of the oxazolidinones **5** and **6** were determined by ^{1}H NMR spectroscopy, the *trans*-compound **5** displayed a coupling constant of 5.86 Hz between H_1 and H_2, whilst the coupling constant for the *cis*-compound **6** was 8.02 Hz. These figures are consistent with those previously reported for this type of *cis*- and *trans*-oxazolidinones.[17,19]

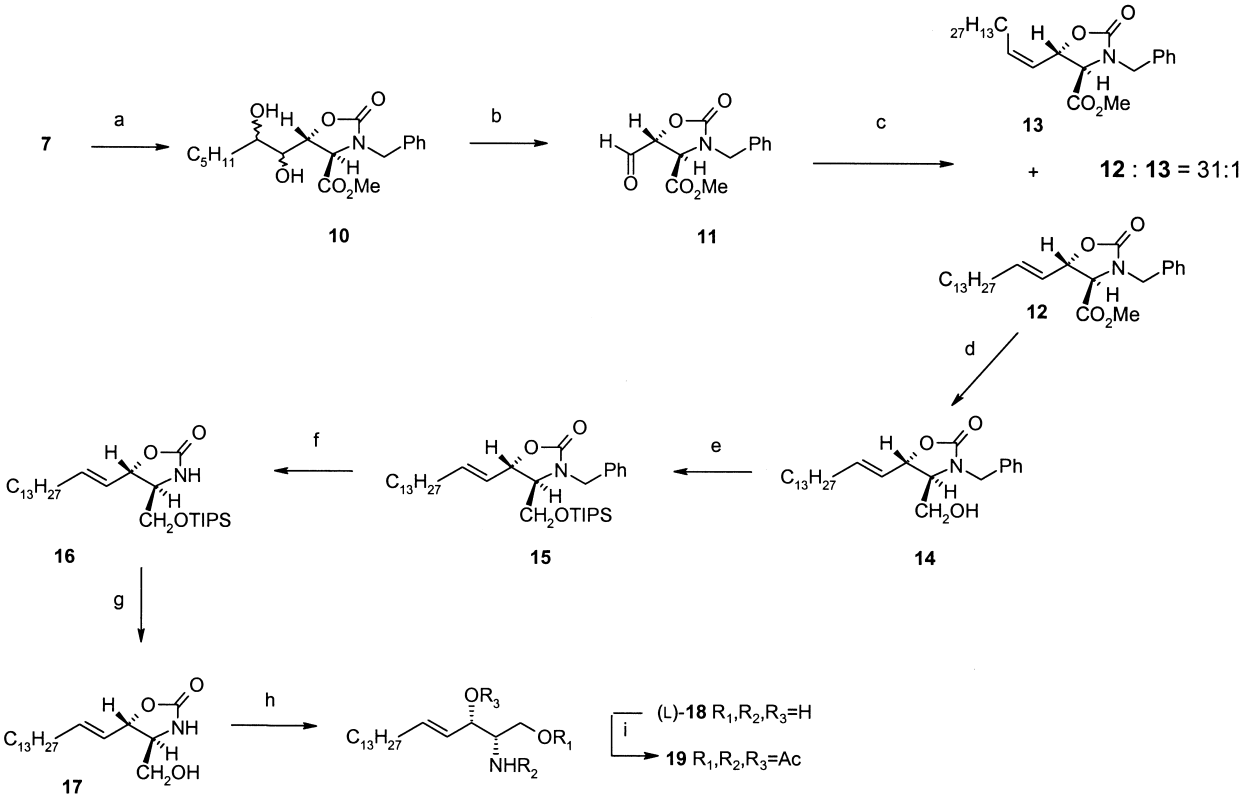
The *trans*-oxazolidinone **5** was converted[20] into the methyl ester **7** without racemization. However the *cis*-oxazolidinone **6** yielded some of the desired ester **8** (15%), but predominantly the amide **9** (73%) which was interconverted into **8** by prolonged reflux (7 days) in methanol under acidic conditions (64% yield with 30% recovered starting material, Scheme 3).



Scheme 2. Reagents: (a) Hnl from *Hevea brasiliensis*, HCN Refs. 16, 35, 100%. (b) *tert*-butyldiphenylsilyl chloride, imidazole, DMF, RT, 90%. (c) (i) DIBALH, Et_2O, 3 h, −70°C then NH_4Br/MeOH (ii) benzylamine, 1.75 h, RT (iii) NaCN/NH_4Br in MeOH, 2.25 h, −45°C, 93%.



Scheme 3. Reagents: (a) TBAF, THF, 20 min, RT, 100%. (b) 1,1-carbonyldiimidazole, CH_2Cl_2, 1 h, RT, 100%. (c) $HCl_{(g)}$, Et_2O/MeOH then H_2O, 10°C, 24 h, 88%. (d) MeOH, conc. HCl, reflux, 7 days, 64% with 30% recovered **9**.

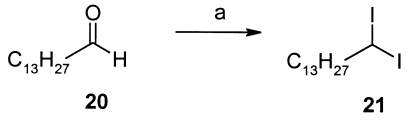


Scheme 4. Reagents: (a) OsO_4, *N*-methylmorpholino-*N*-oxide, acetone/pH 6.8 phosphate buffer, RT, 16 h, 93%. (b) $NaIO_4$, acetone/water, RT, 16 h, 96%. (c) $C_{13}H_{27}CHI_2$ (**21**), $CrCl_2$, DMF/THF, RT, 3 h, 32% (d) $LiBH_4$, THF, 30 min, RT, 94%. (e) TIPSCl, DMF, 24 h, 0°C–RT, 96%. (f) $Na/NH_{3(l)}$, 5 min, −75°C, 70% (g) TBAF, THF, 1.25 h, 0°C, 94%. (h) 2N KOH, EtOH, 2 h, 85°C, 90%. (i) Ac_2O, triethylamine, DMAP, 2 h, 0°C, 95%.

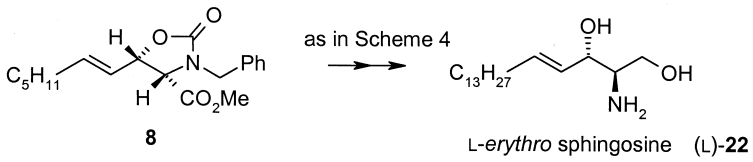
The *trans*-ester **7** was the diastereomer that possessed the correct stereochemistry for conversion to the desired L-*threo*-sphingosine. Cleavage of the heptenyl moiety of **7** was achieved in two steps by employing first, osmium tetroxide/NMO (to yield a mixture of diols **10**)[21] then sodium metaperiodate in THF/water (overall 90% for the two steps).[22] The resultant aldehyde **11** was always freshly prepared for use in the ensuing coupling reaction (Scheme 4).

Reformation of the alkene (cf. **7**) to its corresponding pentadecyl form was then required. Employing the modified Wittig conditions of Schlosser,[23] which yield high (*E*)-selectivity in alkene formation, attempts at coupling a phosphonium salt (tetradecyltriphenylphosphonium bromide)[24] to aldehyde **11** failed. Such conditions gave almost exclusive formation of an (*E*)-alkene in the synthesis of D-*erythro*-sphingosine by Schmidt and Zimmermann.[25] The aldehyde **11** proved to be too sensitive to these basic reaction conditions and only a complex mixture of products resulted which were not fully characterized. Hence, we sought an alternative method which would convert an aldehyde to an (*E*)-alkene. Recently, the use of geminal dichromium reagents, prepared from chromium dichloride and corresponding geminal diiodo compound,[26] have been reported for selective (*E*)-alkene formation[27] as well as a mild alternative to the Wittig reaction.[28] Thus, the required 1,1-diiodo compound **21** was prepared (25% yield) using commercially available tetradecanal **20**, hydrazine hydrate and iodine (Scheme 5).[26a]

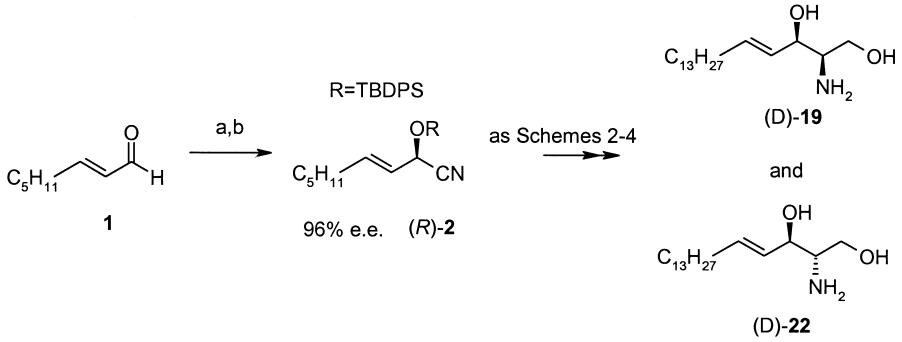
Compound **21** was coupled[27a,28] with the aldehyde **11** to yield two products in an overall yield of 32%. The selectivity proved to be good yielding a 31:1 ratio of compounds **12** and **13** in favor of the desired (*E*)-isomer (Scheme 4).[29] This is in contrast to the Wittig couplings performed by Sugawara et al.[30] or Hudlicky et al.[5a] which proceeded with a preference for (*Z*)-bond formation. The geometric isomers (**12** and **13**) were separable by silica gel column chromatography and their geometry was confirmed by analyzing the coupling constants of their ^1H NMR spectra. Chiral HPLC analysis of **12** revealed that no racemization of the aldehyde had occurred during the reaction. Conversion of the ester **12** to the alcohol **14** was achieved using lithium borohydride in THF (94%) (Scheme 6). The alcohol was then converted to the triisopropylsilyloxyether **15**, a suitable protecting group for the ensuing basic deprotection of the nitrogen.[31] Treatment of **15** with sodium in liquid ammonia at low temperature (−78°C, 5 min) cleanly removed the *N*-benzyl group. Deprotection of the triisopropylsilyl (TIPS) group in **16** proceeded smoothly with TBAF to yield the oxazolidinone **17**. Subsequent basic hydrolysis[32]



Scheme 5. Reagents: (a) (i) Hydrazine hydrate, Et_2O; (ii) triethylamine, I_2, 25%.



Scheme 6. Application to D-*erythro*-sphingosine.



Scheme 7. Reagents: (a) (*R*)-Hnl from *P. amygdalus*, HCN, 3°C; (b) *tert*-butyldiphenylsilylchloride, imidazole, DMF, RT, 30%.

of **17** yielded the desired L-*threo*-sphingosine (L-**18**) whose spectral characteristics were consistent with those previously reported.[7,33] The integrity of the structural assignment was supported by conversion of L-**18** to the known triacetate **19**.[7,33a] The preparation of the *cis*-oxazolidinone **8** (Scheme 3) followed by application of the same chemistry shown in Scheme 4 represents a formal synthesis of L-*erythro*-sphingosine (L-**22**) (Scheme 6).

D-Series sphingosines

In order to access the D-series sphingosines, a hydroxynitrile lyase with a stereochemical preference to yield (*R*)-cyanohydrins was required (Scheme 1). Two such Hnls, those from *L. usitatissium* and *P. amygdalus* (PaHnl) have found biocatalytic applications for the production of (*R*)-cyanohydrins.[9] However, the wide substrate tolerance and cheap source material (almonds) of the PaHnl makes this the preferred catalyst for (*R*)-cyanohydrin formation. This catalyst[34] transformed (*E*)-2-octenal (**1**) into its corresponding cyanohydrin, subsequent derivatization with *tert*-butyldiphenylsilyl chloride yielded (*R*)-(**2**) (30% yield, 96% e.e., Scheme 7).

The preparation of (*R*)-**2** represents a formal synthesis of the two D-series stereoisomers of sphingosine, since only the application of the same chemical steps and reagents outlined in Schemes 2–4 are required to convert (*R*)-(**2**) into the isomers D-**19** and D-**22** (Scheme 7).

Conclusions

In conclusion we have developed a novel chemoenzymatic method, which allows the total synthesis of L-*threo*-sphingosine. By exploiting two hydroxynitrile lyases with complementary enantioselectivites (i.e. those from the HbHnl and PaHnl), the same achiral precursor (i.e. (*E*)-2-octenal) and the same order of chemical steps/reagents we have also developed a general synthetic method which allows the formal synthesis of the other three stereoisomers of the sphingosine family.

Experimental

General

^{1}H and ^{13}C NMR were recorded on Varian Gemini 200 MHz and Bruker MSL 300 MHz instruments in $CDCl_3$ (unless otherwise stated). Chemical shifts are relative to TMS (d 0.00) with $CHCl_3$ as internal standard [d 7.23 (^{1}H) and d 76.90 (^{13}C)]. FT-IR (in ν/cm^{-1}) were recorded on a Bomen Michelson 100 instrument as a neat film on a NaCl disc unless otherwise stated. Optical rotations were measured using a Perkin–Elmer 341 instrument at 589 nm (Na line) in a 1 dm cuvette. Melting points were determined on a Büchi melting point apparatus and are uncorrected. Elemental analysis was made on EA 1108 CHN-analyzer. HPLC enantiomeric separations were performed using a Jasco 880-PU pump and a Jasco 875 UV/VIS detector connected to a CHIRACEL OD-H chiral HPLC column (25 cm×0.46 cm). For (*R*)- or (*S*)-**2** a mobile phase containing 99.75:0.25, heptane:isopropanol was employed and 75:25 heptane:isopropanol with 1% (v/v) triethylamine for compounds **5**–**7**, and **12**. All chromatographic runs were performed with a flow rate of 0.5 mL/min and detection at 254 nm. Tetrahydrofuran was distilled from sodium benzophenone ketyl, methanol and dichloromethane from calcium hydride, and DMF from dibutyltin dilaurate and desmopur-15 (150 mg of each/100 mL DMF). Diethyl ether was dried over calcium chloride, distilled and stored over sodium wire. $HCl_{(g)}$ was dried by passing it through a flask containing concentrated sulfuric acid. $NH_{3(g)}$ was dried by passing it through a column of solid potassium hydroxide and after condensation at −40°C was distilled at atmospheric pressure. (*E*)-2-octenal (**1**) was distilled under reduced pressure prior to use. In all preparations sodium sulfate

was used as the drying reagent. All other solvents and reagents were obtained from commercial suppliers and used as purchased. Petroleum ether refers to fraction of boiling point 60–80°C. All products were purified by silica gel column chromatography using Merck Kieselgel 60 F_{254} (230–400 mesh). TLC plates were run on silica gel Merck 60 F_{254}, compounds were visualized by spraying with Mo-reagent $(NH_4)_6Mo_7O_{24}\cdot 4H_2O$ (100 g/L), $Ce(SO_4)_2\cdot 4H_2O$ (4 g/L) in H_2SO_4 (10% v/v) or $KMnO_4$ reagent $KMnO_4$ (2.5 g/L), Na_2CO_3 (20 g/L) in H_2O. A recombinant hydroxynitrile lyase, which is homologous to the natural enzyme from *Hevea brasiliensis*, was prepared by overexpression in *Pichia pastoris* and a crude cytosolic extract[35] was used for the biotransformation of **5**. A defatted sample of PnHnl was prepared according to the literature procedure.[36]

(+)-(2*S*,3*E*)-2-[(*tert*-butyldiphenylsilyl)oxy]non-3-enenitrile [(*S*)-2]. (*E*)-2-octenal (**1**) (10.0 g, 79.2 mmol) was treated with HCN (10 equiv.) and the Hnl from *Hevea brasiliensis* as catalyst using a reported method[16,35] and then derivatized with *tert*-butyldiphenylchlorosilane (24.3 g, 88.5 mmol, 1.1 equiv.) and imidazole (10.2 g, 150 mmol, 1.9 equiv.).[37] After work-up, the crude product was purified by column chromatography using a gradient mixture of petroleum ether:Et_2O (98:2–90:10) as the eluent which yielded (*S*)-**2** (27.6 g, 90%) as a colorless oil: e.e. >99%, R_f 0.42 (95:5, petroleum ether:Et_2O); $[\alpha]_D^{20}=+25.6$ (*c* 4.85, MeOH); IR (cm^{-1}) 2937, 2857, 2227 (w), 1427, 1110, 975, 702; ^1H NMR (200 MHz), d 0.9 (t, 3H, *J*=7.0 Hz), 1.11 (s, 9H), 1.19–1.38 (m, 6H), 1.99 (m, 2H), 4.77 (d, 1H, *J*=6.3 Hz), 5.50 (dd, 1H, *J*=15.2 and 6.4 Hz), 5.65 (dt, 1H, *J*=15.2 and 6.6 Hz), 7.37–7.52 (m, 6H), 7.65–7.77 (m, 4H); ^{13}C NMR (50 MHz) d 14.04, 19.29, 22.49, 26.69, 28.21, 31.27, 31.80, 63.58, 118.72, 124.69, 127.85, 128.00, 130.28, 130.41, 131.75, 132.17, 135.81, 135.86, 136.89; EIMS *m*/*z* (relative intensity) 391 (3), 334 (100), 307 (11), 264 (19), 239 (11), 209 (21), 199 (52), 181 (10), 109 (15), 78 (11), 57 (9); HRMS calcd for $C_{25}H_{33}NSiO$: 391.2331, found: 391.2340.

(−)-(2*R*,3*E*)-2-[(*tert*-butyldiphenylsilyl)oxy]non-3-enenitrile [(*R*)-2]. To a cooled solution (5°C) of (*E*)-2-octenal (**1**) (504 mg, 4 mmol) in *tert*-butylmethyl ether (MTBE) was added rehydrated (*R*)-Hnl from *Prunus amygdalus*[36] (2 g) then HCN (0.54 g, 0.78 mL, 20 mmol) and the mixture was vigorously stirred using an overhead mechanical stirrer. After 26 h, a further portion of (*R*)-Hnl was added (2 g). After 42 h the enzyme was removed by filtration and extracted with MTBE (2×40 mL). The conversion was estimated to be 30% (by TLC). The organic layers were combined, dried, filtered and concentrated in vacuo. The crude product was then treated with *tert*-butyldiphenylchlorosilane (550 mg, 2.00 mmol, 0.5 equiv.) and imidazole (163 mg, 2.40 mmol, 0.6 equiv.) as previously described.[37] After work-up, the silyl ether was purified by column chromatography eluting with a gradient mixture of petroleum ether:Et_2O (98:2–90:10) to yield (*R*)-**2** (430 mg, 30%) as a colorless oil: e.e. 96%, R_f 0.42 (95:5, petroleum ether:Et_2O); $[\alpha]_D^{20}=-27.2$ (*c* 3.90, MeOH); ^1H NMR (200 MHz) d 0.9 (t, 3H, *J*=7.0 Hz), 1.11 (s, 9H), 1.21–1.37 (m, 6H), 1.96–2.06 (m, 2H), 4.78 (d, 1H, *J*=7.05 Hz), 5.50 (dd, 1H, *J*=15.3 and 6.3 Hz), 5.72 (dt, 1H, *J*=15.3 and 6.4 Hz), 7.35–7.51 (m, 6H), 7.63–7.75 (m, 4H); ^{13}C NMR (50 MHz) d 14.05, 19.31, 22.50, 26.71, 28.23, 31.28, 31.81, 63.59, 118.72, 124.71, 127.89, 128.02, 130.30, 130.43, 131.67, 132.15, 132.17, 135.82, 135.87, 136.90.

(+)-(2*R*/*S*,3*S*,4*E*)-2-benzylamino-3-[(*tert*-butyldiphenylsilyl)oxy]dec-4-enenitrile (3). To a solution of (*S*)-**2** (11.7 g, 30 mmol) in Et_2O (240 mL) under an argon atmosphere at −70°C was added DIBALH (1.0 M in hexane, 75 mL, 75 mmol, 2.5 equiv.) over a 45 min period. Stirring at this temperature was continued for a further 3 h. Then, ammonium bromide (7.31 g, 57 mmol, 2.5 equiv.) in methanol (120 mL) was added slowly and the mixture was allowed to warm to room temperature (RT). Subsequently, benzylamine (16.4 mL, 150 mmol, 5 equiv.) was added and stirring continued for a further 100 min after which the solution was cooled to −45°C and a mixture of sodium cyanide (4.41 g, 90 mmol, 3 equiv.) and ammonium bromide (8.78 g, 90 mmol, 3 equiv.) in methanol (180 mL) was added dropwise. The mixture was stirred for 100 min at −45°C then 1 h at RT, poured into a flask containing water (500 mL) and Et_2O (1000 mL), and stirred for 15 min. The biphasic mixture was separated and the aqueous layer was extracted with Et_2O (2×250 mL). The organic extracts were combined, dried, filtered and evaporated under reduced pressure. Purification using flash chromatography [98:2, petroleum ether:EtOAc with 1% (v/v) triethylamine] yielded the amine **3** (14.00 g, 93%) as a colorless oil: d.e. 57%, R_f 0.27 (95:5, petroleum ether:Et_2O); $[\alpha]_D^{20}=+24.2$ (*c* 4.35, MeOH); IR (cm^{-1}) 2938, 2857, 2227 (w), 1427, 1110, 975, 739, 703; ^1H NMR (200 MHz) 0.85 (t, 3H, *J*=6.3 Hz), 0.97–1.39 (m, 6H), 1.15 (s, 9H), 1.63 (bs, 1H), 1.90 (m, 2H), 3.40 (d, 0.2H, *J*=3.34 Hz), 3.49 (d, 0.8 H, 4.52 Hz), 3.98 (2×d, 1H, *J*=13.2 and 13.0 Hz), 4.02 (2×d, 1H, *J*=13.2 and 13.0 Hz), 4.22 (dd, 0.8H, *J*=8.05 and 4.57 Hz), 4.36 (dd, 0.2H, *J*=7.5 and 3.3 Hz), 5.29–5.68 (m, 2H), 7.05–7.49 (m, 11H), 7.57–7.77 (m, 4H); ^{13}C NMR (50 MHz) 14.05, 19.31, 22.50, 26.73, 26.94, 27.04, 28.20, 28.39, 31.32, 31.96, 32.11, 51.38, 55.93, 56.87, 74.53, 75.19, 118.37, 118.86, 126.75, 127.53, 127.79, 128.08, 128.21, 128.29, 128.35, 128.43, 128.59, 129.79, 129.96, 130.01, 133.20, 133.25, 133.30, 135.83, 135.99, 136.15, 136.39, 137.55, 138.27; EIMS *m*/*z* (relative intensity) 483 (12), 453 (29), 426 (92), 365 (68), 336 (10), 309 (20), 199 (100), 183 (17), 135 (40), 91 (84), 78 (19), 57 (8); HRMS calcd for $C_{33}H_{42}N_2SiO$: 510.3067, found: 510.3058.

(+)-(2*R*/*S*,3*S*,4*E*)-2-benzylamino-3-hydroxydec-4-enenitrile (4). Tetrabutylammonium fluoride (1.0 M in THF, 21.9 mL, 21.9 mmol, 1.02 equiv.) was added dropwise to a cooled (5°C) solution of **3** (10.6 g, 21.5 mmol) in THF (445 mL). The mixture was allowed to warm to RT and stirred for a further 1.5 h after which it was poured into a mixture of EtOAc:saturated sodium bicarbonate solution (550 mL:190 mL) and stirred for further 15 min. The aqueous layer was saturated with sodium chloride, then separated and extracted with EtOAc (2×200 mL). The organic extracts were combined, dried, filtered and evaporated under reduced pressure. Purification using flash chromatography and a gradient eluent mixture [80:20, petroleum ether:EtOAc with 1% (v/v) triethylamine to 50:50 petroleum ether:EtOAc with 1% (v/v) triethylamine] yielded

the amine **4** (5.86 g, 100%) as a colorless oil: d.e. 58%, R_f 0.24 (80:20, petroleum ether:EtOAc); $[\alpha]_D^{20}=+34.5$ (*c* 5.19, MeOH); IR (cm^{-1}) 3436, 2937, 2247 (w), 1697, 1459, 1257, 1121, 980, 739, 699; ^1H NMR (200 MHz, d_4-MeOH), 0.92 (t, 3H, *J*=6.27 Hz), 1.22–1.53 (m, 6H), 2.03–2.19 (m, 2H), 3.33 (bs, 0.5H), 3.05 (2×d, 1H), 3.92 (q, 2H, *J*=13.1 Hz), 4.13–4.30 (m, 1H), 5.58 (dd, 1H, *J*=15.21 and 7.29 Hz), 5.75–5.95 (m, 1H), 7.21–7.47 (m, 5H); ^{13}C NMR (50 MHz, d_4-MeOH) 14.48, 23.62, 29.83, 32.52, 33.31, 33.35, 52.40, 56.65, 57.30, 73.14, 73.59, 119.52, 119.84, 128.56, 128.81, 129.57, 129.63, 135.80, 137.38, 139.88; EIMS *m/z* (relative intensity) 245 (6), 199 (79), 181 (5), 146 (100), 127 (7), 120 (20), 91 (62), 78 (5), 57 (10); HRMS calcd for $C_{17}H_{24}N_2O$: 272.1889, found: 272.1878.

(−)-(*E*,4*S*,5*S*)-3-benzyl-4-cyano-5-(1-heptenyl)oxazolidin-2-one (5) and (−)-(*E*,4*R*,5*S*)-3-benzyl-4-cyano-5-(hept-1-enyl)oxazolidin-2-one (6). To a stirred solution of the diastereomeric mixture **4** (5.66 g, 20.80 mmol) in CH_2Cl_2 (60 mL) under an argon atmosphere was added a solution of 1,1-carbonyldiimidazole (6.74 g, 41.6 mmol, 2 equiv.) in CH_2Cl_2 (60 mL) and stirring was continued for 1 h. To this was added a mixture of CH_2Cl_2:water (500 mL:200 mL), the biphasic mixture separated and the aqueous layer extracted with CH_2Cl_2 (2×200 mL). The organic extracts were combined, dried, filtered and evaporated under reduced pressure. Purification by column chromatography (90:10, cyclohexane:EtOAc to 80:20 cyclohexane:EtOAc) yielded, in order of elution, pure **5** (5.22 g, 84%) as a colorless oil and pure **6** (963 mg, 16%) as a pale yellow oil.

(−)-(*E*,4*S*,5*S*)-3-benzyl-4-cyano-5-(1-heptenyl)oxazolidin-2-one (5). d.e.>99%, e.e. >99%, R_f 0.42 (80:20, petroleum ether:EtOAc); $[\alpha]_D^{20}=-73.4$ (*c* 3.05, MeOH); IR (cm^{-1}) 2935, 2860, 2247 (w), 1768, 1446, 1398, 1197, 970, 703; ^1H NMR (200 MHz) 0.88 (t, 3H, *J*=6.23 Hz), 1.15–1.50 (m, 6H), 1.98–2.18 (m, 2H), 3.93 (d, 1H, *J*=5.86 Hz), 4.15 (d, 1H, *J*=15.0 Hz), 4.98 (2×d, 2H, *J*=15.0 and 13.4 Hz), 5.35 (dd, 1H, *J*=15.3 and 7.65 Hz), 5.97 (dt, 1H, *J*=15.2 and 6.82 Hz), 7.22–7.53 (m, 5H); ^{13}C NMR (50 MHz, $CDCl_3$) 13.99, 22.44, 28.09, 31.25, 32.04, 47.26, 51.21, 76.94, 114.82, 123.07, 128.59, 128.83, 129.30, 133.77, 140.26, 155.68; EIMS *m/z* (relative intensity) 254 (3), 150 (100), 106 (31), 91 (89), 65 (6); HRMS calcd for $C_{18}H_{22}N_2O_2$: 298.1681, found: 298.1690.

(−)-(*E*,4*R*,5*S*)-3-benzyl-4-cyano-5-(1-heptenyl)oxazolidin-2-one (6). d.e.>99%, e.e. >99%, R_f 0.30 (80:20, petroleum ether:EtOAc); $[\alpha]_D^{20}=-67.7$ (*c* 5.49, MeOH); IR (cm^{-1}) 2919, 2243 (w), 1756, 1453, 1411, 1204, 972, 702; ^1H NMR (200 MHz) 0.88 (t, 3H, *J*=6.59 Hz), 1.13–1.50 (m, 6H), 2.07–2.21 (m, 2H), 4.10 (d, 1H, *J*=14.8 Hz), 4.35 (d, 1H, *J*=8.02 Hz), 4.90 (d, 1H, *J*=8.06 Hz), 5.00 (d, 1H, *J*=14.8 Hz), 5.68 (dd, 1H, *J*=15.2 and 8.06 Hz), 6.05 (dt, 1H, *J*=15.2 and 6.77 Hz), 7.28–7.50 (m, 5H); ^{13}C NMR (50 MHz) 14.01, 22.45, 28.11, 31.23, 32.19, 47.45, 51.52, 74.86, 113.33, 121.64 128.66, 128.80, 129.29, 134.05, 141.90, 155.83; EIMS *m/z* (relative intensity) 298 (3), 254 (17), 150 (100), 197 (10), 106 (39), 91 (81), 65 (6); HRMS calcd for $C_{18}H_{22}N_2O_2$: 298.1681, found: 298.1687.

(−)-(*E*,4*R*,5*S*)-3-benzyl-5-(1-heptenyl)-4-methoxycarbonyl-oxazolidin-2-one (7). Through a cooled (3°C) solution of **5** (3.00 g, 10.06 mmol) in methanol:Et_2O (15 mL:225 mL) was bubbled dry $HCl_{(g)}$ for 10 min. The mixture was allowed to stand at 10°C for 24 h after which it was further cooled to 4°C. Water (5 mL) was added and stirring continued for 20 min. The aqueous layer was separated, extracted with Et_2O (3×60 mL). The organic layers were combined, washed with 5% sodium bicarbonate solution, dried, filtered and concentrated under reduced pressure. Purification using flash chromatography (80:20, petroleum ether:EtOAc) yielded **7** (2.93 g, 88%) as a colorless oil: e.e. >99%, R_f 0.34 (80:20, petroleum ether:EtOAc); $[\alpha]_D^{20}=-70.92$ (*c* 5.35, MeOH); IR (cm^{-1}) 2936, 2864, 1757, 1407, 1214, 989, 704; ^1H NMR (200 MHz) 0.88 (t, 3H, *J*=6.27 Hz), 1.16–1.45 (m, 6H), 1.97–2.13 (m, 2H), 3.72 (s, 3H), 3.75 (d, 1H, *J*=5.13 Hz), 4.22 (d, 1H, *J*=15.0 Hz), 4.87 (2×d, 2H, *J*=15.2 and 11.9 Hz), 5.40 (dd, 1H, *J*=15.2 and 6.96 Hz), 5.85 (dt, 1H, *J*=15.3 and 6.78 Hz), 7.18–7.41 (m, 5H); ^{13}C NMR (50 MHz) 14.03, 22.48, 28.29, 31.26, 32.01, 47.30, 52.84, 61.80, 76.64, 125.27, 128.20, 128.43, 128.91, 134.59, 137.38, 156.97, 169.47; EIMS *m/z* (relative intensity) 331 (5), 287 (6), 272 (56), 228 (61), 182 (10), 106 (11), 91 (100), 65 (5); HRMS calcd for $C_{19}H_{25}NO_4$: 331.1784, found: 331.1782.

(−)-(*E*,4*S*,5*S*)-3-benzyl-5-(1-heptenyl)-4-methoxycarbonyl-oxazolidin-2-one (8). Through a cooled (3°C) solution of **6** (914 mg, 3.06 mmol) in methanol:Et_2O (0.25 mL:3.8 mL) was bubbled dry $HCl_{(g)}$ for 10 min. The mixture was allowed to stand at 10°C for 24 h after which a further portion of methanol (2 mL) was added and the mixture stirred for further 3 h at 5°C. Water (5 mL) was added and stirring continued for 45 min. The aqueous layer was separated and extracted with EtOAc (3×70 mL). The organic layers were combined, dried, filtered and concentrated under reduced pressure. Recrystallization from acetone yielded **9** (780 mg, 73%) as a white solid. R_f 0.40 (50:50, petroleum ether: EtOAc); Mp 185–186°C, $[\alpha]_D^{20}=-36.78$ (*c*=3.00, DMSO); IR (cm^{-1}) 3381, 3215, 2924, 2856, 1732, 1677, 1427, 1228, 968, 704; ^1H NMR (300 MHz, d_6-DMSO) 0.89 (t, 3H, *J*=6.59 Hz), 1.21–1.42 (m, 6H), 2.01–2.08 (m, 2H), 3.35 (s, 0.75H), 3.83 (d, 1H, *J*=15.44 Hz), 4.11 (d, 1H, *J*=8.51 Hz), 4.76 (d, 2H, *J*=15.4 Hz), 5.12 (t, 1H, *J*=8.48 Hz), 5.42 (dd, 1H, *J*=15.3 and 8.48 Hz), 5.92 (dt, 1H, *J*=15.3 and 6.75 Hz), 7.29–7.44 (m, 5H), 7.61 (bs, 0.5H); ^{13}C NMR (50 MHz, $CDCl_3$) 13.79, 21.82, 27.71, 30.54, 31.43, 46.34, 60.43, 75.78, 123.12, 127.52, 127.68, 128.60, 136.08, 138.28, 157.51, 168.05, 168.11; Anal. calcd for $C_{18}H_{24}N_2O_3$: C, 68.33; H, 7.65; N, 8.85; Found: C, 68.35; H, 7.60; N, 8.77.

The mother liquor was concentrated under reduced pressure and purified using column chromatography to yield **8** (150 mg, 15%) as a pale yellow oil R_f 0.36 (80:20, petroleum ether:EtOAc); $[\alpha]_D^{20}=-45.29$ (*c* 1.10, CH_2Cl_2); IR (cm^{-1}) 2926, 2856, 1759, 1439, 1214, 981, 704; ^1H NMR (200 MHz) 0.86 (t, 3H, *J*=6.60 Hz), 1.20–1.36 (m, 6H), 1.97–2.08 (m, 2H), 3.67 (s, 3H), 4.00 (d, 1H, *J*=14.89 Hz), 4.12 (d, 1H, *J*=8.98 Hz), 4.91 (d, 1H, *J*=14.89 Hz), 4.96 (t, 1H, 8.00 Hz), 5.30 (dd, 1H, *J*=15.3 and 7.81 Hz), 5.89 (dt, 1H, *J*=15.3 and 6.64 Hz), 7.20–7.34 (m, 5H); ^{13}C NMR (50 MHz) 14.00, 22.47, 28.33, 31.20, 32.14, 47.44, 52.30, 61.12, 75.70, 121.78, 128.25, 128.43, 128.61, 128.95, 135.08, 139.37, 157.48, 168.28; EIMS *m/z*

(relative intensity) 287 (4), 272 (22), 228 (30), 182 (9), 106 (11), 91 (100), 65 (5); HRMS calcd for $C_{19}H_{25}NO_4$: 331.1784, found: 331.1785.

Compound **9** was converted into the desired ester **8** by the following procedure: A solution of **9** (650 mg, 1.85 mmol) in methanol (270 mL) and concentrated hydrochloric acid (3 mL) was heated at reflux for 7 days. The mixture was cooled to RT, concentrated to approximately one quarter of its original volume, diluted with EtOAc (250 mL) and washed with water (2×30 mL) and then brine (30 mL). Silica gel column chromatography using a gradient elution (petroleum ether:EtOAc, 5:1 to neat EtOAc) yielded in order of elution **8** (400 mg, 64%) and **9** (200 mg, 30%).

(−)-(4*R*,5*R*)-3-benzyl-5-(1,2-dihydroxyheptyl)-4-methoxycarbonyloxazolidin-2-one (10). Ester **7** (1.63 g, 4.92 mmol) was suspended in phosphate buffer (0.1 M, pH 6.8, 8.1 mL) to which was added acetone (82 mL), then *N*-methylmorpholino-*N*-oxide (1.15 g, 9.84 mmol, 2 equiv.) followed by two crystals of osmium tetroxide and stirred for 16 h at RT. Sodium sulfite (3.70 g, 29.3 mmol, 5.9 equiv.) was added and stirring was continued for 30 min. The mixture was concentrated to one half of its original volume, diluted with EtOAc (120 mL) and the organic phase washed with water (2×10 mL), followed by brine (15 mL). The combined organic extracts were dried, filtered and concentrated under reduced pressure. The crude residue was purified using flash chromatography (99:1–99:2, CH_2Cl_2:methanol) to yield **10** (1.66 g, 93%) as a white solid. Mp 115–116°C; R_f 0.37 (50:50 petroleum ether: EtOAc); $[\alpha]_D^{20}=-42.44$ (*c* 1.43, MeOH); IR (cm^{-1}) 3403 (w), 2937 (w), 1758, 1743, 1415, 1227, 705; ^{1}H NMR (200 MHz, d_6-acetone) 0.88 (t, 3H, *J*=6.35 Hz), 1.20–1.57 (m, 8H), 3.45–3.57 (m, 1H), 3.67 (2×s, 4H), 4.22–4.38 (m, 3H), 4.61–4.74 (m, 2H), 7.28–7.34 (m, 5H); ^{13}C NMR (50 MHz, d_6-acetone) 14.42, 23.39, 26.14, 26.23, 32.67, 33.79, 34.45, 47.71, 47.82, 52.92, 59.29, 70.73, 71.78, 73.95, 75.53, 77.01, 77.46, 128.53, 128.60, 129.00, 129.57, 129.52, 136.75, 137.15, 171.19, 183.49; EIMS *m/z* (relative intensity) 365 (15), 306 (8), 262 (5), 234 (12), 205 (6), 220 (12), 176 (55), 130 (10), 106 (9), 91 (100), 65 (4); HRMS calcd for $C_{19}H_{27}NO_6$: 365.1838, found: 365.1838; Anal. calcd for $C_{19}H_{27}NO_6$: C, 62.58; H, 7.60; N, 3.77; Found: C, 62.63; H, 7.67; N, 3.78.

(−)-(4*R*,5*R*)-3-benzyl-5-formyl-4-methoxycarbonyloxazolidin-2-one (11). The diol **10** (200 mg, 0.547 mmol) was suspended in water (4 mL) to which was added acetone (10 mL) followed by $NaIO_4$ (195 mg, 0.915 mmol) portion-wise. The mixture was stirred for 16 h at RT then concentrated to approximately one quarter of its original volume under reduced pressure, then solid NaCl was added to the point of saturation and extracted with EtOAc (3×30 mL). The combined organic extracts were dried filtered and concentrated under reduced pressure. The crude residue was purified using flash chromatography (70:30, EtOAc:petroleum ether) to yield **11** (144 mg, 96%) as a colorless oil: R_f 0.22 (30:70, petroleum ether: EtOAc); $[\alpha]_D^{20}=-1.0$ (*c* 0.8, $CHCl_3$); ^{1}H NMR (200 MHz, d_6-acetone) 1.07 (m, 2H), 3.84–3.81 (m, 3H), 4.19–4.31 (m, 2H), 4.42–4.49 (m, 1H), 4.69–4.85 (m, 1.5 H), 5.66–5.81 (m, 0.5H), ^{13}C NMR (50 MHz, d_6-acetone)14.41, 15.26, 15.35, 20.73, 47.42, 47.54, 47.77, 52.84, 54.88, 57.37, 57.50, 57.66, 57.76, 60.45, 63.29, 63.68, 77.09, 77.33, 77.47, 78.40, 89.99, 95.19, 95.42, 96.62, 96.72, 128.43, 128.70, 128.90, 129.30, 157.73, 170.98, 171.26, 197.09.

(−)-(*E*,4*R*,5*S*)-3-benzyl-4-methoxycarbonyl-5-(1-pentadecenyl)oxazolidin-2-one (12). All apparatus was covered in aluminum foil to exclude light. To a stirred suspension of $CrCl_2$ (1.83 g, 14.9 mmol, 9.3 equiv.) under an argon atmosphere at RT in THF (36 mL) was added DMF (1.14 mL, 14.9 mmol, 9.3 equiv.). After 30 min a solution containing iodoalkane **21** (1.68 g, 3.73 mmol, 2.3 equiv.) and aldehyde **11** (441 mg, 1.59 mmol) in THF (8 mL) was added and stirring continued for 1.5 h. The mixture was then diluted with EtOAc (30 mL) and poured into water (80 mL). Solid NaCl was added to the point of saturation and the aqueous layer extracted with EtOAc (3×40 mL). The combined extracts were dried, filtered and concentrated under reduced pressure. Purification using flash chromatography (95:5–90:10–85:15, petroleum ether:EtOAc) yielded in order of elution pure **12** (220 mg, 31%) as a colorless oil which solidified upon storage in a refrigerator and the corresponding pure *cis*-isomer **13** (7 mg, 1%).

(−)-(*E*,4*R*,5*S*)-methyl-3-benzyl-4-oxycarbonyl-5-(1-pentadecenyl)oxazolidin-2-one (12). e.e. >99%, R_f 0.15 (85:15, petroleum ether: EtOAc); $[\alpha]_D^{20}=-53.09$ (*c* 1.34, MeOH); Mp. 31–32°C; IR (cm^{-1}) 2923, 2852, 1764, 1753, 1445, 1407, 1203, 988, 704; ^{1}H NMR (200 MHz) 0.88 (t, 3H, *J*=6.78 Hz), 1.25 (bs, 22H), 1.98–2.10 (m, 2H), 3.72 (s, 3H), 3.57 (d, 1H, *J*=5.13 Hz), 4.21 (d, 1H, *J*=15.0 Hz), 4.86 (2×d, 2H, *J*=15.2 and 11.7 Hz), 5.39 (ddt, 1H, *J*=15.2, 6.98 and 1.3 Hz), 5.85 (dt, 1H, *J*=15.2 and 8.43 Hz), 7.18–7.40 (m, 5H); ^{13}C NMR (50 MHz) 14.16, 22.73, 28.51, 28.64, 28.94, 29.00, 29.11, 29.40, 29.47, 29.60, 29.70, 30.11, 31.96, 32.06, 47.29, 52.83, 61.80, 76.65,125.23, 128.18, 128.28, 128.42, 128.90, 135.10, 137.40, 157.18, 169.64; EIMS (EI) *m/z* (relative intensity) 384 (15), 340 (19), 228 (11), 106 (18), 91 (100), 44 (11); HRMS calcd for $C_{27}H_{41}NO_4$: 443.3036, found: 443.3043; Anal. calcd for $C_{27}H_{41}NO_4.1H_2O$: C, 72.11; H, 9.77; N, 2.93; Found: C, 72.04; H, 9.74; N, 2.96.

(−)-(*Z*,4*R*,5*S*)-3-benzyl-4-methoxycarbonyl-5-(1-pentadecenyl)oxazolidin-2-one (13). R_f 0.21 (80:20, petroleum ether:EtOAc); $[\alpha]_D^{20}=-3.70$ (*c* 0.60, $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$) 0.90 (t, 3H, *J*=6.54 Hz), 1.24 (bs, 22H), 1.98–2.10 (m, 2H), 3.72 (s, 3H), 3.57 (d, 1H, *J*=5.13 Hz), 4.22 (d, 1H, *J*=15.0 Hz), 4.90 (d, 1H, *J*=14.9 Hz), 5.21 (dd, 1H, *J*=8.84 and 5.18 Hz), 5.36 (ddt, 1H, *J*=10.3 and 8.92 Hz), 5.73 (dt, 1H, *J*=10.5 and 7.32 Hz), 7.24–7.32 (m, 5H); ^{13}C NMR (50 MHz) 14.17, 22.74, 27.83, 29.21, 29.30, 29.40, 49.11, 29.59, 29.60, 29.69, 29.70, 30.11, 31.96, 32.06, 47.39, 52.83, 62.25, 76.06, 125.11, 128.21, 128.43, 128.46, 128.94, 137.96, 137.40, 157.18, 169.64.

(−)-(*E*,4*S*,5*S*)-3-benzyl-4-(hydroxymethyl)-5-(1-pentadecenyl)oxazolidin-2-one (14). To a stirred solution of ester **12** (205 mg, 0.463 mmol) in THF (1.5 mL) at RT was added $LiBH_4$ (20 mg, 0.926 mmol, 2 equiv.). After 20 min the mixture was cooled on an ice bath to 5°C and

methanol (1.5 mL) was added. After stirring for a further 15 min, Amberlite IR-120-H^+ resin (which had been washed with MeOH) was added portion-wise until the basic solution became neutral. The mixture was filtered and the solvent removed under reduced pressure. The residue was re-suspended in methanol (10 mL) which was then removed under reduced pressure, a procedure which was repeated a further two times in order to remove boric acid as its methyl ester. Purification using flash chromatography (70:30, petroleum ether:EtOAc) yielded **14** (180 mg, 94%) as a colorless oil: R_f 0.36 (60:40, petroleum ether:EtOAc); $[\alpha]_D^{20}=-71.27$ (*c* 1.53, MeOH); IR (cm^{-1}) 3494, 2920, 2850, 1730, 1712, 1430, 1128, 972, 704; ^1H NMR (200 MHz) 0.88 (t, 3H, *J*=6.85 Hz), 1.25 (bs, 22H), 1.98–2.05 (m, 2H), 2.60 (bs, 1H), 3.32 (dt, 1H, *J*=6.67 and 3.41 Hz), 3.50 (dd, 1H, *J*=12.48 and 2.90 Hz), 3.75 (dd, 1H, *J*=12.35 and 3.17 Hz), 4.32 (d, 1H, *J*=15.28 Hz), 4.69 (d, 1H, *J*=15.30 Hz), 4.79 (t, 1H, *J*=7.12 Hz), 5.40 (dd, 1H, *J*=15.32 and 7.83 Hz), 5.83 (dt, 1H, *J*=15.27 and 6.74 Hz), 7.27–7.38 (m, 5H); ^{13}C NMR (50 MHz,) 14.23, 22.81, 28.84, 29.25, 29.48, 29.57, 29.69, 29.79, 32.05, 32.25, 46.68, 59.76, 62.34, 76.85, 126.01, 128.14, 129.08, 136.38, 137.57, 157.78; EIMS *m/z* (relative intensity) 415 (3), 384 (61), 340 (46), 280 (5), 150 (9), 106 (11), 91 (100), 44 (12); HRMS calcd for $C_{26}H_{41}NO_3$: 415.3087, found: 415.3082.

(−)-(*E*,4*R*,5*S*)-3-benzyl-5-(1-pentadecenyl)-4-(tri-iso-propylsilyloxymethyl)oxazolidin-2-one (15). To a solution of imidazole (45 mg, 0.662 mmol, 2 equiv.) in DMF (50 mL) at 0°C was added tri-isopropylchlorosilane (95 mg, 0.496 mmol, 1.5 equiv.). After stirring for 20 min, **14** (137 mg, 0.331 mmol) in DMF (200 mL) was added dropwise and the mixture was allowed to warm to RT and stirring continued for 24 h. The mixture was diluted with water (5 mL), extracted with CH_2Cl_2 (3×15 mL) and the organic extracts were combined, washed with brine solution (5 mL), dried, filtered and concentrated under reduced pressure. Purification using flash chromatography (95:5–90:10, petroleum ether:EtOAc) yielded **15** (184 mg, 96%) as a colorless oil: R_f 0.31 (90:10, petroleum ether:EtOAc); $[\alpha]_D^{20}=-36.30$ (*c* 2.03, MeOH); IR (cm^{-1}) 2926, 2858, 1754, 1115, 970 (w); ^1H NMR (200 MHz) 0.88 (t, 3H, *J*=6.85 Hz), 1.05 (bs, 21H), 1.25 (bs, 22H), 1.97–2.06 (m, 2H), 3.26–3.33 (m, 1H), 3.69 (dq, 2H, *J*=10.74 and 4.64 Hz), 4.15 (d, 1H, *J*=15.14 Hz), 4.72 (t, 1H, *J*=6.35 Hz), 4.84 (d, 1H, *J*=15.14 Hz), 5.40 (dd, 1H, *J*=15.35 and 7.40 Hz), 5.79 (dt, 1H, *J*=15.14 and 6.65 Hz), 7.27–7.38 (m, 5H); ^{13}C NMR (50 MHz) 11.90, 14.17, 17.98, 22.74, 28.80, 29.14, 29.41, 29.51, 29.62, 29.72, 31.98, 32.14, 46.45, 61.65, 61.84, 77.08, 126.43, 127.85, 128.05, 128.79, 136.21, 136.52, 158.14; EIMS *m/z* (relative intensity) 528 (60), 484 (20), 379 (25), 340 (20), 307 (5), 262 (16), 174 (7), 131 (62), 91 (100), 75 (35), 65 (20), 44 (40); HRMS calcd for $C_{35}H_{61}NO_3Si$: 571.4421, found: 571.4428.

(−)-(*E*,4*R*,5*S*)-5-(1-pentadecenyl)-4-(tri-iso-propylsilyloxymethyl)oxazolidin-2-one (16). To a flask containing freshly distilled liquid ammonia under an argon atmosphere at −65°C was added sodium (132 mg, 5.74 mmol, 23 equiv.) and stirring continued for 30 min. The blue–bronze colored solution was then further cooled to −75°C and a solution of **15** (144 mg, 0.25 mmol) in dry Et_2O (1.35 mL) added. After 5 min phosphate buffer (0.5 M, pH 6.8, 2.8 mL) was added quickly and stirring continued for another 10 min until the blue color had disappeared. The mixture was allowed to warm to RT. After 1 h, water (15 mL) was added and the reaction mixture was extracted with EtOAc (3×20 mL), the organic extracts were combined, dried, filtered and concentrated under reduced pressure. Purification by flash chromatography using a gradient mixture of petroleum ether:EtOAc (90:10–80:20) containing 1% (v/v) triethylamine yielded **16** (84 mg, 70%) as a colorless oil: R_f 0.42 (70:30, petroleum ether:EtOAc); $[\alpha]_D^{20}=-48.9$ (*c* 2.05, MeOH); IR (cm^{-1}) 3264 (w), 2926, 2859, 1756, 1121, 991 (w); ^1H NMR (200 MHz) 0.88 (t, 3H, *J*=6.85 Hz), 1.05 (bs, 21H), 1.25 (bs, 22H), 2.01–2.11 (m, 2H), 3.59–3.36 (m, 1H), 3.69–3.72 (m, 2H), 4.71 (dd, 1H, *J*=5.38 and 1.95 Hz), 5.54 (dd, 1H, *J*=15.32 and 7.51 Hz), 5.82 (dt, 1H, *J*=15.4 and 6.60 Hz); 5.96 (bs, 1H); ^{13}C NMR (50 MHz) 11.88, 14.15, 17.95, 22.73, 28.74, 29.15, 29.39, 29.50, 29.62, 29.71, 31.96, 32.12, 59.97, 64.78, 79.80, 126.37, 136.70, 159.11; EIMS *m/z* (relative intensity) 438 (30), 408 (33), 394 (100), 250 (25), 216 (7), 172 (12), 131 (42), 103 (25), 75 (13), 43 (30); HRMS calcd for $C_{28}H_{55}NO_3Si$: 481.3951, found: 481.3951.

(−)-(*E*,4*S*,5*S*)-4-hydroxymethyl-5-(1-pentadecenyl)oxazolidin-2-one (17). To a solution of **16** (70 mg, 0.145 mmol) in THF (200 mL) at 0°C under an argon atmosphere was added TBAF (1.0 M in THF, 152 mL, 1.05 equiv.) dropwise over 5 min. Stirring was continued for 75 min. The solution was neutralized by the portion-wise addition of Amberlite IR-120-H^+ resin. The resin was removed by filtration and washed with EtOAc (3×2 mL) and the solvent removed by evaporation under reduced pressure. Purification using flash chromatography [CH_2Cl_2:methanol, 95:5+1% (v/v) triethylamine] yielded **17** (44 mg, 94%) as a white solid whose spectral characteristics were consistent with those previously reported:[30] R_f 0.23 (CH_2Cl_2:methanol, 95:5); Mp 101–102°C, (Lit. 104–105°C),[30] $[\alpha]_D^{20}=-63.9$ (*c* 2.05, $CHCl_3$), [Lit.[34] −32.0 (*c* 0.5, $CHCl_3$)]; Anal. calcd for $C_{19}H_{35}NO_3\cdot 1H_2O$: C, 70.51; H, 11.46; N, 4.01; Found: C, 70.38; H, 11.41; N, 4.02.

L-*threo*-sphingosine: (−)-(*E*,2*S*,3*S*)-2-aminooctadec-4-ene-1,3-diol [(L)-18]. A mixture of **17** (20 mg, 61.5 mmol) in ethanol (0.48 mL) and 2N NaOH (0.48 mL) was heated at 85°C for 2.5 h. The mixture was then cooled to RT, diluted with Et_2O (10 mL) and washed with 2N NaOH (3×2 mL) and then brine solution (2 mL). The organic layer was dried, filtered and concentrated in vacuo. The crude residue was purified by flash chromatography [CH_2Cl_2:methanol, 90:10+1% (v/v) NH_4OH (25% in H_2O)] to yield (L)-**18** (16 mg, 90%) as a white solid whose spectral characteristics were identical with those previously reported.[7,33a,b] Mp 84–85°C, (Lit.[7]: 86–87°C); $[\alpha]_D^{20}=-2.40$ (*c* 0.8, $CHCl_3$), [Lit.[7] −2.83 (*c* 1.0, $CHCl_3$)].

L-*threo*-sphingosine-*N*,*O*,*O*-triacetate (19). The compound was prepared according to the method reported by Enders et al.[7] which afforded a colorless solid (16 mg, 95%) whose spectral characteristics were identical to those previously reported.[7,33a] Mp 42–43°C, (Lit.[7]: 41–42°C); $[\alpha]_D^{20}=+8.35$ (*c* 0.8, $CHCl_3$), [Lit.[7]: +8.78 (*c* 1.2, $CHCl_3$)].

1,1-Di-iodotetradecane (21). This compound is light sensitive and all apparatus should be covered in aluminum foil to exclude light. Compound **21** was prepared using the procedure of Pross et al.[26a] employing tetradecanal (10 g, 47.1 mmol), hydrazine hydrate (10.61 g, 212 mmol, 4.5 equiv.) and triethylamine (126 mL, 904 mmol, 19 equiv.). Purification using column chromatography and neat hexane as the eluant yielded **21** (4.12 g, 22%) as a pale red oil which solidified upon storage at −20°C: IR (cm^{-1}) 2922, 2852, 1460, 1121, 1088, 721, 620, 545 (w); ^{1}H NMR (200 MHz) 0.88 (t, 3H, J=6.23 Hz), 1.16–1.55 (m, 22H), 2.33 (m, 2H), 5.11 (t, 1H, J=6.59 Hz, CHI_2); ^{13}C NMR (50 MHz) 14.19, 22.75, 27.68, 29.41, 29.54, 29.63, 29.70, 29.73, 31.88, 31.98, 48.40.

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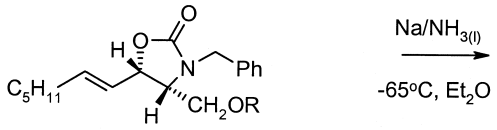

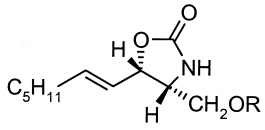

30	Reaction time (min)	Yield(%)[a]
a R=H	5	10
b R=TBDMS	15	30
c R=TIPS	10	76
c R=TIPS	5	91

[a] In all experiments the starting material was fully consumed.

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