Chemical Constituents of a Marine Soft Coral of the Genus *Lobophytum*

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Chemical investigations on the marine soft coral *Lobophytum* sp., collected from the Andaman and Nicobar Islands, yielded two new acyl glycerol monoalkyl ethers along with two known compounds of this series (1a—d), and a new ceramide (4a). The structures of the new and known compounds were elucidated on the basis of extensive spectroscopic studies and chemical reactions.

Key words acyl glycerol monoalkyl ether; ceramide; soft coral; *Lobophytum* sp.; Alcyoniidae

Marine soft corals are known to be rich sources of structurally and biologically intriguing natural products.[1,2] In continuation of our search for bioactive natural products from soft corals, collected from the Indian Ocean, we have reported several new compounds.[3—8] Our recent chemical studies on the ethyl acetate extract of soft coral, *Lobophytum* sp., collected from the coasts of Andaman and Nicobar Islands, have yielded a mixture of four acyl glycerol monoalkyl ethers (**1a**—**d**) including two new components (**1b**) and (**1d**), and a new ceramide **4a** along with the ubiquitous occurring mixture of sterols, chimyl alcohol, batyl alcohol[9] and two known ceramides.[10] The isolation and structure elucidation of the new compounds is reported here.

Results and Discussion

The mixture of **1a**—**d** was isolated as an amorphous colorless powder. ESI-MS gave a series of $[2M+Na]^+$ *pseudo*-molecular ion peaks at *m*/*z* 1131, 1159, 1187, 1215 which showed peak heights in the ratio of 1 : 3 : 4 : 2 suggesting a mixture of homologous compounds consistent with the molecular formulae $C_{35}H_{70}O_4$, $C_{36}H_{72}O_4$, $C_{37}H_{74}O_4$, and $C_{38}H_{76}O_4$, respectively. The mixture gave a single spot on thin-layer chromatography (TLC). Several attempts to purify the individual constituents **1a**—**d** from the mixture by column chromatography and by high-performance liquid chromatography (HPLC) including reversed-phase were not successful, therefore we recorded all spectra for the mixture.

The infrared (IR) spectrum of the mixture **1a**—**d** showed the absorptions of hydroxyl and ester functions at 3450 and 1730 cm^{-1}, respectively. The proton signals for three oxymethylene groups and one oxymethine group at δ 4.18 (2H), 4.00 (1H), and 3.45 (4H) in the ^{1}H-NMR spectrum, and an ester carbonyl carbon and four oxygenated carbon atoms at δ 173.9 (C_q), 65.4 (CH_2), 68.8 (CH), 71.4 (CH_2) and 71.7 (CH_2) in the ^{13}C-NMR spectrum were indicative for the presence of a disubstituted glyceride backbone, R–$CH_2OCH_2CH(OH)CH_2OCOCH_2$–R′ in **1a**—**d**. The proton signal at δ 3.45 (4H, m) revealed the presence of two ether linked oxymethylenes, while an ABX signal at δ 4.18 (CH_2-3) and the triplet at δ 2.34 of an α-methylene group in an acyl residue indicated the ester linkage with an acyl chain. The presence of a proton multiplet at δ 4.00 (1H) indicated the absence of substitution at C-2.[9,11—13] The ^{1}H-NMR signals at δ 1.25 (br s, $(CH_2)_n$), 0.88 (6 H, t, J=6.7 Hz), and carbon signals at δ 34.2 (CH_2), 22.7 (CH_2), and 14.1 (CH_3) showed the presence of an unbranched long aliphatic chain. Assignments of these resonances were accomplished from the multipulse attached proton test (APT), H,H correlation spectroscopy (COSY) and heteronuclear multiple quantum coherence (HMQC) spectral data.

The lengths of the acyl and alkyl ether chains in **1a**—**d** were determined by treatment with potassium hydroxide in methanol. The reaction furnished a residue, which upon separation by column chromatography gave methyl hexadecanoate (16 : 0) (M^+: *m*/*z* 270) (**2**) and a glycerol monoalkyl ether mixture (**3**). On acetylation or oxidative cleavage with periodic acid, mixture **3** furnished the corresponding diacetates and glycolaldehyde alkyl ethers, respectively, confirming the presence of a free 2,3-diol group. The electron impact mass spectrum (EI-MS) of mixture **3** showed four molecular ions at *m*/*z* 316 (10%), 330 (30%), 344 (40%) and 358 (20%), revealing the presence of chain length variation in the ether linked alkyl chain of the mixture **1a**—**d**. The ^{1}H- and ^{13}C-NMR data of the mixture **3** were identical with those of 1-*O*-hexadecyl-glycerol (chimyl alcohol) and 1-*O*-octadecyl-glycerol (batyl alcohol).[9] As the optical rotation of **3** was similar as that of (*S*)-batyl or (*S*)-chimyl alcohol, and as all other chiral glyceride-1-ethers known to date are *S* configured,[14] the configuration of **3** at C-2 was proposed to be (*S*). It should be stated that also the optical rotation of the **1a**—**d** mixture is close to that of 1-*O*-hexadecyl-3-hexadecanoyl glycerol (**1a**) described previously.[9] From the foregoing discussion, it may be proposed that **1a**—**d** is a mixture of acyl glycerol monoalkyl ethers consisting of (*R*)-1-*O*-hexadecyl-3-*O*-hexadecanoyl glycerol (**1a**), (*R*)-1-*O*-heptadecyl-3-*O*-hexadecanoyl glycerol (**1b**), (*R*)-1-*O*-octadecyl-3-*O*-hexadecanoyl glycerol (**1c**), and (*R*)-1-*O*-nonadecyl-3-*O*-hexadecanoyl glycerol (**1d**). Of these, the two components **1b** and **1d** have not been reported to date. The occurrence of components **1a** and **1c** in marine soft corals was reported for a new species of *Lobophytum*,[9] for *Sinularia fibrilla*[15] and *Sinularia simpsoni*.[16] Glycerol derivatives of the type (+)-**1** are membrane-active and known to be ichthyotoxic, while the (−)-isomers are not toxic.[9]

Compound **4a** was obtained as an amorphous powder, mp 127—129 °C and analysed for $C_{36}H_{73}NO_5$ by high resolution (HR)-ESI-MS and elemental analysis. The IR spectrum showed absorptions at 3250—3400, 1680, 1060 cm^{-1}, attributable to the hydroxyl and the carbonyl of amide functionality. Absorptions at 2924, 2868, 1350 cm^{-1} and the lack of ab-

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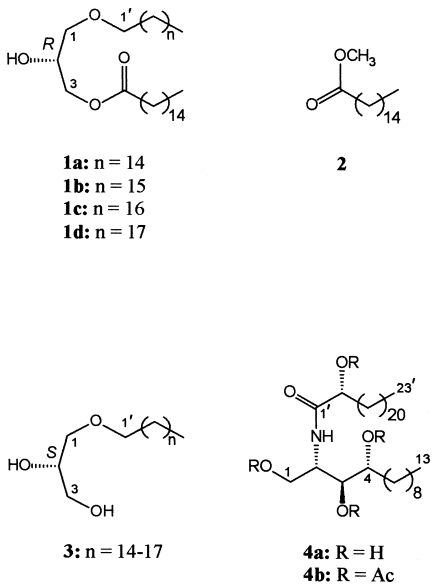

Fig. 1

sorption at 1450 cm^{-1} suggested a saturated fatty acid amide. The ultraviolet (UV) spectrum in methanol did not show characteristic absorption maxima above 210 nm. The presence of signals due to two terminal methyl groups of aliphatic chains [δ_H 0.88 (s, 13-H_3 and 23′-H_3), protons of long methylene chains [δ_H 1.25—1.68 (54H)] and an amide proton (δ_H 7.3, d, J=6.0 Hz, 1H, exchangeable with D_2O) in the ^{1}H-NMR spectrum and an NH-attached methine carbon signal at δ 51.2 (CH) and a carbonyl carbon signal at δ 173.1 in the ^{13}C-NMR spectrum indicated a sphingolipid skeleton.[17,18] Formation of a tetraacetate (**4b**) [δ_H 2.0—2.2 (12H) and δ_C 20.5—21.0] and the presence of oxygenated carbons at δ 74.4 (CH), 70.9 (CH), 71.0 (CH) and 60.3 (CH_2) suggested the presence of four hydroxyls in **4a**. On the basis of the H,H COSY spectrum of **4a**, six deshielded proton signals at δ 5.23 (m), 4.42 (m), 4.36 (m), 4.10 (m), 3.94 (m), and 3.85 (m) were assigned to 2′-H, 2-H, 3-H, 4-H, 1-H_2, respectively. The absence of a proton triplet at δ 2.22—2.50 and the appearance of a 1H multiplet at δ 5.23 in the ^{1}H-NMR spectrum suggested the presence of a –CO–CH(OH)–CH_2-moiety in **4a**.[18,19] The remaining methylene protons are buried in the range δ 1.68—1.25 region. In addition to carbon signals for several methylene groups in the range of δ 34.0—24.9, the terminal methylene carbon at δ 21.5 and the end methyls of aliphatic chains at δ 13.3 indicated the absence of branching. The positions of oxygenated carbons in **4a** were assigned on the basis of H,H COSY and HMQC spectra which led to the proposal that **4a** is a sphingolipid containing a 1,3,4-trihydroxy base and a 2-hydroxy fatty acid without unsaturation in the aliphatic chains.

The lengths of the fatty acid and sphingosine chains in **4a** were determined by methanolysis of **4a**, followed by acetylation of sphingamines according to the method of Gaver–Sweeley[20] to afford long-chain base (LCB) and methyl ester of 2-hydroxy fatty acid (FAM). The methyl ester and tetraacetylated sphingamines thus obtained were independently analyzed by gas chromatography-mass spectrometry (GC-MS), which showed ions at m/z 384 and 415 corresponding to methyl 2-hydroxytricosanoate and 2-acetamino-1,3,4-triacetoxy-tridecane, respectively. Based on the-NMR and optical rotation values of natural and synthetic methyl esters and sphingamines,[17,21] *R* and 2*S*,3*S*,4*R* configurations were proposed to methyl ester and sphingamine moieties, respectively. The structure of **4a** is (2*S*,3*S*,4*R*)-2-[(*R*)-2′-hydroxytricosanoylamino]-1,3,4-tridecanetriol, which has not been described so far.

Table 1. ^{1}H, ^{13}C, APT and H,H COSY Spectral Data of Compound **4a**

Carbon No.	δ ^{1}H, (mult., J in Hz) (300 MHz, DMSO-d_6)	δ ^{13}C (75 MHz, DMSO-d_6)	H,H COSY
1a	3.94 (m, 1H)	60.3 (CH_2)	1_b-H, 2-H
1b	3.85 (m, 1H)	—	1_a-H, 2-H
2	4.42 (m, 1H)	51.2 (CH)	NH, 3-H
3	4.36 (m, 1H)	74.4 (CH)	2-H, 4-H
4	4.10 (m, 1H)	70.9 (CH)	3-H, 5-H_2
5	1.68 (m, 2H)	31.8 (CH_2)	4-H, 6-H_2
6	1.46 (m, 2H)	24.9 (CH_2)	5-H_2, 7-H_2
7—10		30.8—24.9 (CH_2)	6-H_2—13-H_3
11	1.25 (br s, 12H)	30.8 (CH_2)	
12		21.5 (CH_2)	
13	0.88 (t, 6.0, 3H)	13.3 (CH_3)	12-H_2
1′	—	173.1 (C_q)	—
2′	5.23 (m, 1H)	71.0 (CH)	3′-H_2
3′	2.12 (m, 2H)	34.0 (CH_2)	2′-H, 4′-H_2
4′	1.68 (m, 2H)	24.1 (CH_2)	3′-H_2, 5′-H_2
5′	1.55 (m, 2H)	28.2 (CH_2)	—
6′—20′		30.8—24.9 (CH_2)	—
21′	1.25 (br s, 32H)	30.8 (CH_2)	
22′		21.5 (CH_2)	21′-H_2, 23′-H_3
23′	0.88 (t, 6.0, 3H)	13.3 (CH_3)	22′-H_2
NH	7.30 (d, 6.0, 1H)		2-H
2OH	3.58 (m)		
2OH	3.39 (m)		

Experimental

General Melting points were determined on a Kofler hot stage and are uncorrected. UV and IR spectra were measured with an HP 8451 A diode array spectrophotometer and a Perkin-Elmer 1600 series FTIR spectrophotometer. Optical rotations were determined on a Perkin-Elmer Model 241 Polarimeter. ESI-MS was recorded on a Quadro Triple Quadrupole Mass Spectrometer, Finnigan TSQ 7000 with nano-ESI-API ion source. EI-mass spectra were recorded on a Varian MAT 731 (70 eV). Perfluorokerosene was used as reference substance in EI-HR-MS. ^{1}H- and ^{13}C-NMR spectra were measured on Varian Unity 300 (300.145 MHz), Varian Unity 300 (75.479 MHz) and AMX 300 (300.135 MHz) spectrometers, using $CDCl_3$ and DMSO-d_6 as the solvents and TMS as internal standard. Chemical shifts were reported in δ values, and coupling constants (J) were expressed in Hertz. Column chromatography was carried out using silica gel (100—200 mesh, Acme), *semi*-preparative reversed-phase HPLC (Akzo Nobel RP_{18} column, 7.5×250 mm, MeOH/H_2O 95 : 5) was performed with a L-6200 A pump (Merck-Hitachi) equipped with a UV/vis detector (λ=220 nm) L-4250 C (Merck-Hitachi) and a Chromato-integrator D-2500 (Merck-Hitachi). Fractions were monitored by TLC on Polygram SIL G/UV_{254} (Macherey-Nagel & Co.) with visualization under UV (254, 366 nm) and Dragendorff's spray reagent. R_F values were measured on Polygram SILG/UV_{254} (Macherey-Nagel & Co.), gel filtration was carried out using Sephadex LH 20 (Pharmacia Biotech), GC-MS experiments were carried out on Shimadzu's GC-MS-QP5050 A system. All solvents were distilled prior to their use. Elemental analysis was carried out on a Carlo Erba 1108 analyser.

Animal Material The soft coral (1 kg dry weight) was collected by hand-picking in inter-tidal rocky region during March 1993 off the coasts of the Andaman and Nicobar Islands, India (Diglipur Island 13°20′N, 93°02′E) at a depth of 1 m and it was cut into small pieces and preserved in ethanol at room temperature until extraction. A voucher specimen was deposited in the Department of Organic Chemistry, Andhra University, Visakhapatnam, India (Voucher No. MF-VA/35).

Extraction and Isolation The soft coral was extracted with ethanol by

percolation every 4 d. The process was repeated several times. The solvent was evaporated by distillation under reduced pressure, and the resulting crude extract was partitioned between ethyl acetate and water. Concentration of the organic layer resulted in a brownish gummy residue (30 g), which was passed over anhydrous $MgSO_4$. The extract was subjected to silica gel column chromatography (500 g, 100—200 mesh, Acme) eluting with hexane through hexane/ethyl acetate to ethyl acetate and methanol. Petroleum ether/ethyl acetate (9 : 1) eluted a mixture of inseparable sterols (1.0 g). Acetylation of the sterol mixture (60 mg) gave the corresponding sterol acetates. The GC-MS and NMR data of the sterol acetates suggested the presence of three commonly occurring sterols, namely 24-methylenecholest-7-en-3β-ol (20%), 24-methylcholest-5,22-diene-3β-ol (43.5%) and 24-methylcholest-5,25-diene-3β-ol (36.5%). Petroleum ether/ethyl acetate (4 : 1) yielded 600 mg of a mixture of 1-*O*-hexadecylglycerol (chimyl alcohol) and 1-*O*-octadecylglycerol (batyl alcohol). Elution with petroleum ether/ethyl acetate (3 : 2) yielded 200 mg of a mixture of **1a**—**d**, which was inseparable by reverse phase HPLC on C_{18} column. Elution with petroleum ether/ethyl acetate (1 : 1) furnished 40 mg of compound **4a**. Further elution with petroleum ether/ethyl acetate (1 : 4) successively yielded 50 mg each of *N*-palmitoyl-D-*erythro*-(2*S*,3*R*)-octadecasphinga-4(*E*),8(*E*),10(*E*)-triene[10] and *N*-palmitoyl-D-*erythro*-(2*S*,3*R*)-octadecasphinga-4(*E*),8(*E*)-diene.[10] Separation of the chemical components was monitored by TLC analysis. Successive column chromatography and recrystallisation from methanol was carried out for purification of each compound. The known compounds were identified by comparison of their physical and spectral data with authentic samples.[22,23]

Mixture **1a**—**d**: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ: 0.88 (6H, t, J=6.7 Hz, 2CH_3), 1.25 (s, $(CH_2)_n$), 1.60 (4H, m, 2′-,3″-H_2), 2.34 (2H, t, J=7.5 Hz, 2″-H), 2.58 (1H, br s, 2-OH), 3.45 (4H, m, 1,1′-H_2), 4.00 (1H, m, 2-H), 4.18 (2 H, ABX, J_{AB}=9.0 Hz, 3-H_2). ^{13}C-NMR ($CDCl_3$, 75 MHz) δ: 14.1 (2 CH_3), 22.7 (2 CH_2), 25.0 (CH_2-3″), 26.1 (CH_2-3′), 29.1—29.7 (nCH_2), 31.9 (CH_2-14″,-16′), 34.2 (CH_2-2″), 65.4 (CH_2-3), 68.8 (CH-2), 71.4 (CH_2-1*), 71.7 (CH_2-1′*), 173.9 (C_q, CO-1″). IR (KBr) cm^{-1}: 3450, 1730. ESI-MS *m*/*z* (%): 1215 ([2M+Na]$^+$, 35), 1187 ([2M′+Na]$^+$, 100), 1159 ([2M″+Na]$^+$, 75), 1131 ([2M‴+Na]$^+$, 16) corresponding to $C_{38}H_{76}O_4$, $C_{37}H_{74}O_4$, $C_{36}H_{72}O_4$, and $C_{35}H_{70}O_4$. $[\alpha]_D^{25}$ +6.8° (c=0.3, CH_3OH). * Assignment may be exchanged.

Hydrolysis of Mixture 1a—d: Fifty milligrams of **1a**—**d** in 5 ml of 10% methanolic KOH was refluxed for 6 h. Column chromatography of the residue so obtained using hexane as eluant yielded 10 mg of methyl hexadecanoate (**2**), evidenced by GC-MS analysis (*m*/*z* 270, M$^+$), and 25 mg of a mixture of glycerol monoalkyl ethers (**3**).

3: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ: 0.88 (3H, t, J=6.7 Hz, CH_3), 1.26 (26H, br s, CH_2), 1.58 (m, 2H, 2′-H_2), 2.60, 2.90 (2H, 2 br s, 2OH), 3.45 (4H, m, 1′-,3-H_2), 3.72, 3.64 (2H, ABX, J_{AB}=11.5, J_{AX}=5.0, J_{BX}=3.5 Hz, 1-H_2), 3.87 (1H, m, H-2). ^{13}C-NMR ($CDCl_3$, 75 MHz) δ: 14.1 (CH_3), 22.7 (CH_2-14′), 26.1 (CH_2-3′), 29.7—29.3 (nCH_2), 31.9 (CH_2-13′), 64.2 (CH_2-3), 70.5 (CH-2), 71.8 (CH_2-1), 72.4 (CH_2-1′). IR (KBr) cm^{-1}: 3500—3350, 2920, 2810, 1140. EI-MS *m*/*z*: 316, 330, 344, 358 (4×[M]$^+$). $[\alpha]_D^{25}$ +7.2° (c=0.3, $CHCl_3$).

Acetylation An aliquot of the glycerol monoalkyl ether mixture (**3**) (10 mg) was treated with acetic anhydride (0.2 ml) and pyridine (0.1 ml) at room temperature and left aside overnight. The excess reactants were removed on a rotary evaporator under reduced pressure. The residue was rechromatographed on a column of silica gel to yield the diacetate (5 mg) as a pale yellow oil.

^{1}H-NMR ($CDCl_3$, 300 MHz) δ: 0.89 (3H, t, J=6.9 Hz, CH_3), 1.26 (s, –$(CH_2)_n$–), 1.56 (1H, quint, H-2′), 2.08 (3H, s, OAc), 2.10 (3H, s, OAc), 3.44 (2H, m, H_2-1′), 3.56 (2H, d, J=5.3 Hz, H_2-1), 4.16 (1H, dd, J=11.5, 5.8 Hz, H-3b), 4.34 (1H, dd, J=11.5, 4.0 Hz, H-3a), 5.28 (1H, m, H-2). ^{13}C-NMR ($CDCl_3$, 75 MHz) δ: 14.1 (CH_3), 20.8 (C_q), 21.0 (C_q), 22.7 (CH_2), 26.0 (CH_2), 29.4 (CH_2), 29.5 (CH_2), 29.6 (CH_2), 29.7 (CH_2), 31.9 (CH_2), 34.8 (CH_2), 63.5 (CH_2-3), 66.6 (CH-2), 69.8 (CH_2-1′), 70.1 (CH_2-1), 170.1 (C_q), 171.2 (C_q).

Periodate Oxidation A solution of the glycerol monoalkyl ether mixture (**3**) (10 mg) in diethyl ether was treated with 38% aqueous periodic acid[24] (10 ml) at room temperature overnight, and the reaction mixture was purified by column chromatography to yield the respective glycolaldehyde ethers (5 mg).

^{1}H-NMR ($CDCl_3$, 300 MHz) δ: 0.94 (3H, t, J=7.0 Hz, CH_3), 1.25 (s, CH_2, aliphatic chain), 1.57 (2H, m, H-2′), 3.53 (2H, t, J=6.5 Hz, H-1′), 4.05 (2H, d, J=1.0 Hz, H-1), 9.84 (1H, t, J=1.0 Hz, H-2).

Compound **4a**: IR (KBr) cm^{-1}: 3250—3400, 2924, 2868, 1680, 1350, 1060. (+)-ESI-MS *m*/*z* (%): 1221.8 ([2M+Na]$^+$, 100), 622.9 ([M+Na]$^+$, 4); (−)-ESI-MS/MS *m*/*z* (%): 598.6 ([M−H]$^-$, 30), 354.5 (15), 342.6 (10), 299.6 (95). $[\alpha]_D^{25}$ +8.2° (c=0.5, CH_3OH). *Anal.* Calcd for $C_{36}H_{73}NO_5$: C, 82.72; H, 12.50; N, 2.41. Found: C, 82.68; H, 12.52; N, 2.40. ^{1}H-, ^{13}C-NMR, APT (DMSO-d_6), H,H COSY, see Table 1.

Acetylation Dry pyridine (0.5 ml) and Ac_2O (1.0 ml) were added to compound **4a** (25 mg) and the mixture left overnight. Usual workup and crystallization yielded **4b**.

Compound **4b**: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ: 0.85 (6H, t, J=6.9 Hz, terminal Me), 1.25 (48H, br s, 24 CH_2), 1.54 (4H, m, 2CH_2), 1.80 (2H, m, CH_2), 1.91 (2H, m, CH_2), 2.08 (3H, s, OAc), 2.12 (6H, s, 2OAc), 2.16 (3H, s, OAc), 2.20 (2H, t, J=6.0 Hz, 2-H_2), 4.12, 4.25 (2H, *AB*X, J_{AB}=10.6, J_{AX}=4.0, J_{BX}=3.5 Hz, 1-H_2), 4.60 (1H, m), 4.95 (1H, dd, J=9.5, 3.5 Hz), 5.10 (1H, t, J=7.0 Hz), 5.30 (1H, dt, J=10, 5 Hz), 6.50 (1H, d, J=9 Hz, NH). ^{13}C-NMR ($CDCl_3$, 75 MHz) δ: 14.1 (2 CH_3), 20.5 (OAc), 20.7 (2 OAc), 21.0 (OAc), 22.6 (2 CH_2), 25.5 (CH_2), 25.6 (CH_2), 28.0 (CH_2), 29.2—29.7 (21 CH_2), 31.9 (5-CH_2), 36.7 (CH_2), 47.3 (2-CH), 62.8 (1-CH_2), 71.8 (4-CH), 72.9 (3-CH), 170.0 (4-CO), 170.8, 171.1, 172.8. $[\alpha]_D^{25}$ +9.2° (c=0.5, CH_3OH).

Methanolysis Compound **4a** (10 mg) was treated with 3 ml of 1 N HCl in methanol at 90 °C for 15 h with magnetic stirring. The fatty acid methyl ester so obtained was extracted with *n*-hexane and analyzed by GC-MS. Fatty acid methyl ester (3.0 mg) from compound **4a**: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ: 0.83 (3H, t, J=6.7 Hz, CH_3), 1.25—1.45 (36H, br s, CH_2), 1.75 (2H, m, H-3), 3.75 (3H, s, OCH_3), 4.15 (1H, dd, J=7.5, 4.2 Hz, H-2). GC-MS *m*/*z*: 384 ([M]$^+$). $[\alpha]_D^{25}$ −2.4° (c=0.5, CH_3OH). The MeOH/H_2O phase was evaporated and the residue was acetylated. Purification by filtration over a Sephadex LH 20 column (CH_2Cl_2/MeOH, 1 : 1) yielded the acetylated sphingamine. The sphingamine was subjected to GC-MS. Acetylated sphingamine (4.0 mg) from compound **4a**: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ: 0.88 (3H, t, J=7.0 Hz, CH_3), 1.25 (14H, br s, –CH_2–), 1.72 (2H, m, H-5), 2.02—2.08 (12H, s, 4 $COCH_3$), 4.00 (1H, dd, J=11.5, 3.0 Hz, H-1b), 4.28 (1H, dd, J=11.0, 4.2 Hz, H-1a), 4.43 (1H, m, H-2), 4.89 (1H, dt, J=9.5, 3.0 Hz, H-4), 5.10 (1H, dd, J=8.5, 3.2 Hz, H-3), 6.00 (1H, d, J=9.0 Hz, NH). GC-MS *m*/*z*: 415 [M$^+$]. $[\alpha]_D^{25}$ +8.0° (c=0.5, CH_3OH).

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