

Supporting Information

Comprehensive Molecular Motion Capture for Sphingomyelin by Site-Specific Deuterium Labeling

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1. Comparison of depth of cholesterol between DMPC and SSM bilayers

As described in the text, Figures 2 and 3 suggest that cholesterol is located deeper in the SSM bilayers than in DMPC membrane. We inspected available X-ray and neutron diffraction data in order to gain more evidence. Figures S1A and B show electron density profiles of DMPC and DMPC-cholesterol (1:1) obtained from previous X-ray diffraction experiments (Needham, D., McIntosh, T. J., and Evans, E. (1988) *Biochemistry* 27, 4668-4673). In both profiles, the high-density peaks correspond to the electron-rich phosphate groups. It is generally accepted that cholesterol in membrane increase electron density (McIntosh, T. J., (1978) *Biochim. Biophys. Acta*, 513, 43-58); therefore the shoulders seen in Figure S1B are due to the increase in electron-density because of the sterol ring system. Figure S1C shows the neutron scattering amplitude density of 2,2,3,4,4,6-*d*₆-cholesterol (thick line) in a DMPC membrane (thin line), which demonstrates that the center of mass of the six deuterons of the cholesterol molecule is located 16 Å away from the bilayer center (Léonard, A., Escrive, C., Laguerre M., Pebay-Peyroula, E., Néri, W., Pott, T., Katsaras, J., and Dufourc, E. (2001) *Langmuir* 17, 3019-2030). Reasonably, the position of deuterons of the cholesterol molecule (Figure S1C) is consistent with the shoulder of the electron density profile (Figure S1B) as indicated by the red line. In the neutron scattering results (Figure S1C), the most intense peaks correspond to the glycerol ester regions, and the outer shoulders are thought to be due to the phosphate groups. The blue lines in Figure S1 show that the position of the phosphate groups is coincident in both profiles, although the experimental method and the ratio of cholesterol are different.

Figure S2 shows the electron density profiles of SSM in the presence and absence of 50 mol% cholesterol (Maulik, P. R., and Shipley, G. G. (1996) *Biophys. J.* 70, 2256-2265). As is the case with DMPC, cholesterol induces enhancement of the electron density of SSM chains. Following the criteria in the DMPC membrane that cholesterol increases electron density to form shoulder in the profile, we put the red arrow indicating the presumed position of the A ring of cholesterol. Figure S3 compares the electron density profiles between SSM-cholesterol and DMPC-cholesterol membranes. In the figure, the highest

density peaks corresponding to the phosphate peaks are aligned as shown in the blue line. The positions of cholesterol in the membranes indicated by red arrows suggest that the location of cholesterol in the SSM bilayers can be further from the membrane head group than that in the DMPC membrane. Thus, this comparison seems supportive of our proposal that cholesterol is located deeper in SSM membrane than in DMPC bilayers.

Here we also compared the distances between the bilayer center and the Chol's A ring for SSM and DMPC membranes. Interestingly, the distances estimated based on Figures S1-3 seem mostly identical for both membranes (ca. 16 Å). This may be reasonable because the head-group separation across the bilayer of SSM-Chol membrane (46.6 Å, Maulik, P. R., and Shipley, G. G. *Biophys. J.* 1996, 70, 2256-2265) is longer than that of DMPC-Chol by ca. 5 Å than that of DMPC-Chol (42.0 Å, Needham, D., McIntosh, T. J., and Evans, E. *Biochemistry* 1988, 27, 4668-4673). Of concern is that Chol resides at the same distance from the bilayer center regardless of the membrane lipid. McIntosh (McIntosh, T. J., (1978) *Biochim. Biophys. Acta*, 513, 43-58), however, showed that the distance becomes longer with the increasing acyl chain length of PC (DLPC to DSPC). Therefore, the same distances between Chol and membrane center for SSM-Chol and DMPC-Chol membranes may be a coincidental match.

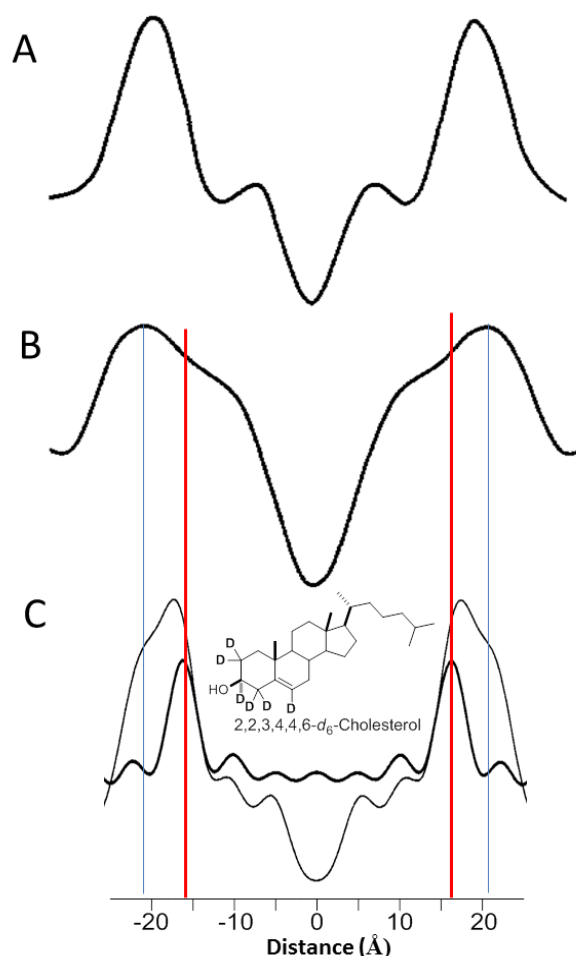


Figure S1. Electron density and neutron scattering profiles for DMPC membranes. Electron density profiles for DMPC membrane (A) and DMPC containing 50 mol% cholesterol (B) (Needham, D., McIntosh, T. J., and Evans, E. (1988) *Biochemistry* 27, 4668-4673). Both profiles were obtained by X-ray diffraction experiments. The steroid rings of cholesterol markedly increase the electron density of the hydrocarbon chain regions of the profile (B). (C): neutron scattering profile for DMPC (thin line) containing 30 mol% 2,2,3,4,4,6-*d*₆-cholesterol (thick line) (Léonard, A., Escrive, C., Laguerre M., Pebay-Peyroula, E., Néri, W., Pott, T., Katsaras, J., and Dufourc, E. (2001) *Langmuir* 17, 3019-2030). The highest density peaks in (A) and (B) correspond to the phosphate head groups, while they correspond to the glycerol ester regions in (C). The blue lines represent the position of the phosphate group in B and C, and the red lines show the position of the A ring of cholesterol.

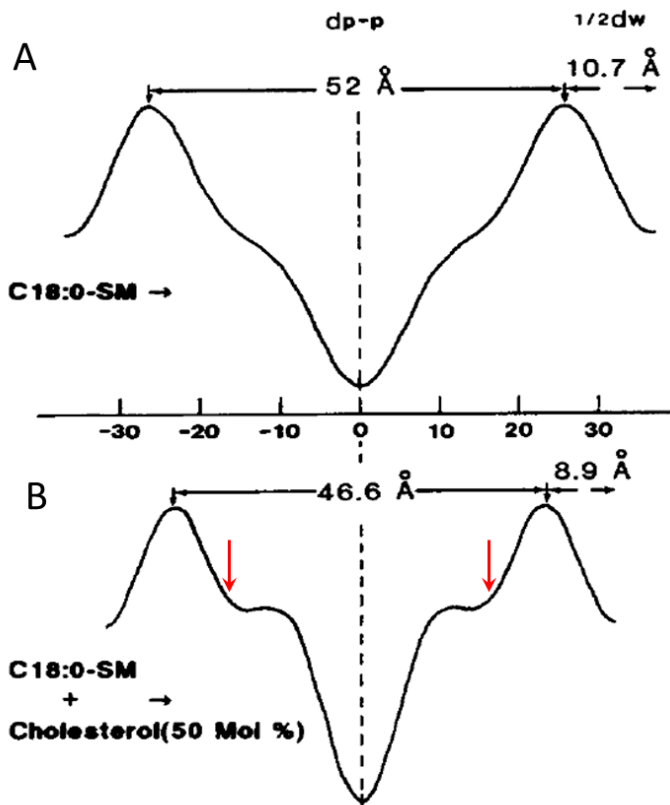


Figure S2. Electron density profiles for SSM membranes. SSM membrane (A) and SSM containing 50 mol% cholesterol (B). Both profiles were obtained by X-ray diffraction experiments (Maulik, P. R., and Shipley, G. G. (1996) *Biophys. J.* 70, 2256-2265). As in the case with DMPC membrane, the steroid rings of cholesterol markedly increase the electron density of the hydrocarbon chain regions of the profile. The red arrows indicate the presumed positions of the A ring of cholesterol, which was inferred from the DMPC profile.

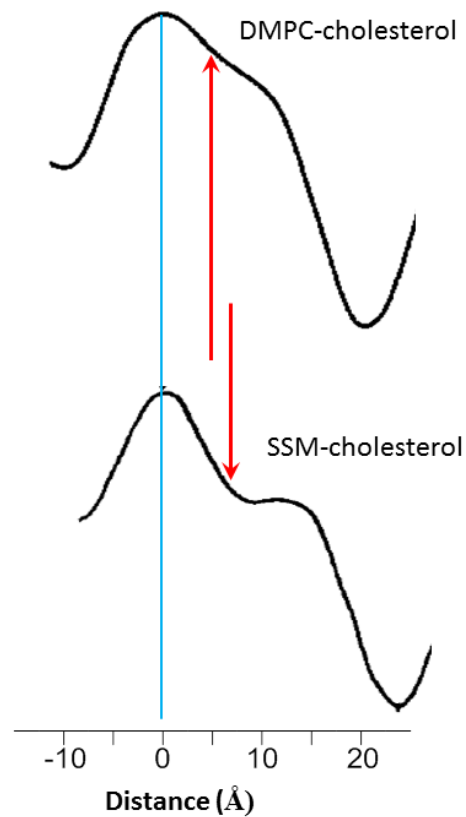


Figure S3. Comparison of electron density profiles for DMPC-cholesterol and SSM-cholesterol membranes. Both membranes contain 50 mol% cholesterol. The blue line represents the position of the phosphate groups. The red arrows represent the positions of cholesterol A ring in both membranes.

2. Supplementary Figures

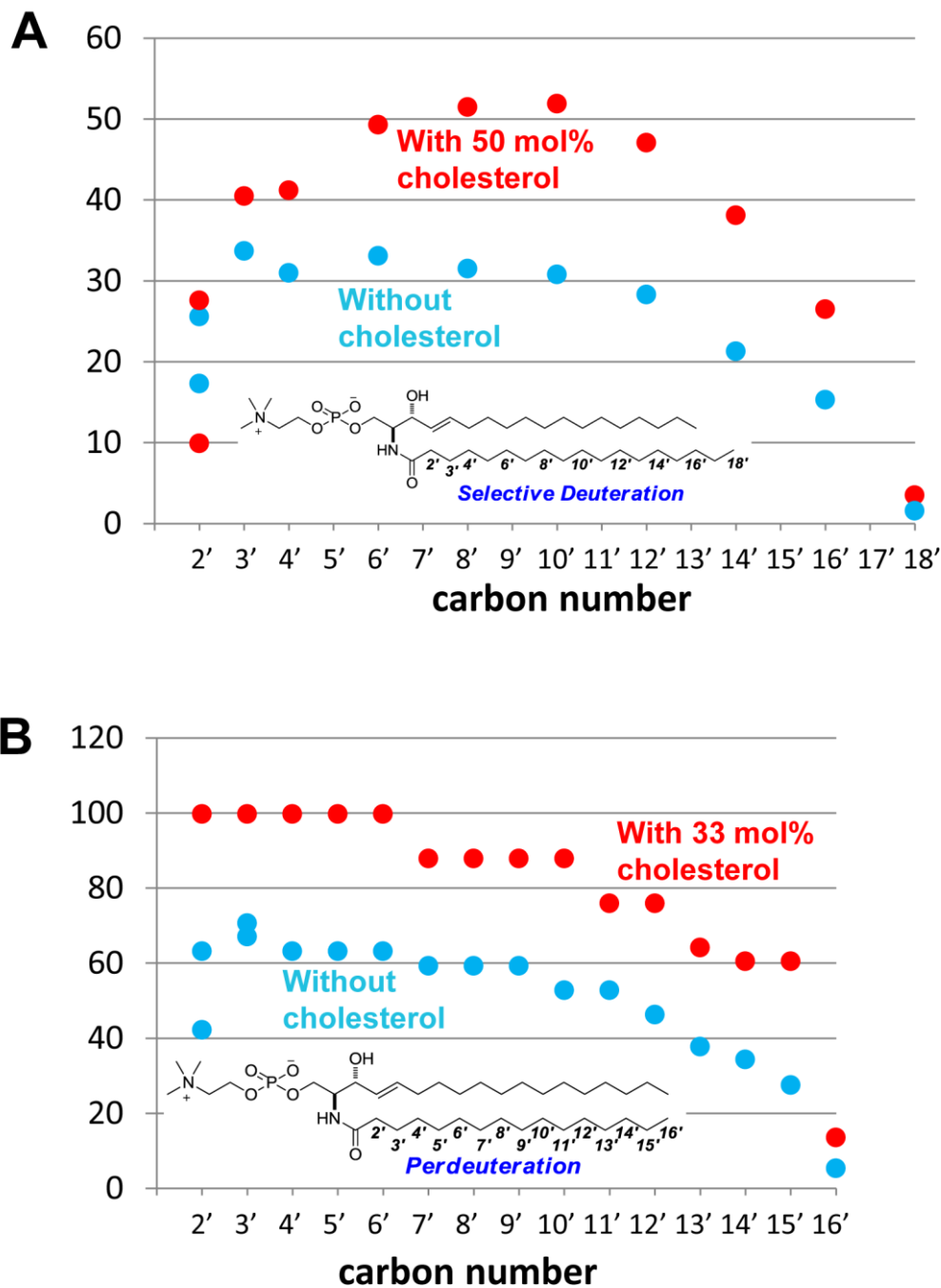


Figure S4. Comparison of quadrupole coupling profiles of SM acyl chain between currently obtained data (A) and reported one (B) (Bartels, T., et al (2008) Raftlike mixtures of sphingomyelin and cholesterol investigated by solid-state ^2H NMR spectroscopy. *J. Am. Chem. Soc.* 130, 14521–14532). Both data were obtained at 45 °C. Because Bartels et al used planar membrane, the quadrupolar couplings in (B) are twice the size of (A). As is clear from this comparison, the previous report has possibility of misassignment because they used sphingomyelin whose acyl chain is perdeuterated.

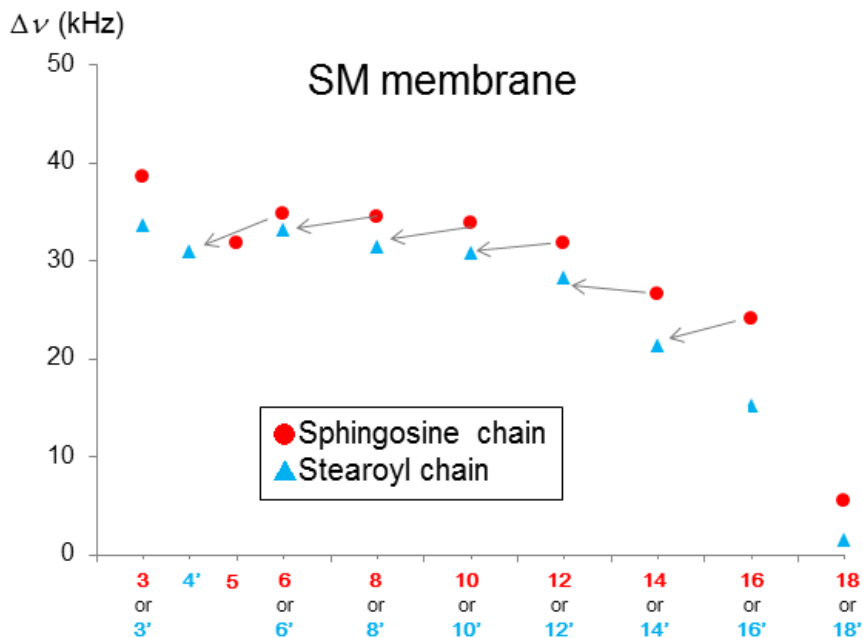


Figure S5. Comparison of quadrupole coupling profiles between sphingosine and stearoyl chains for SSM membrane.

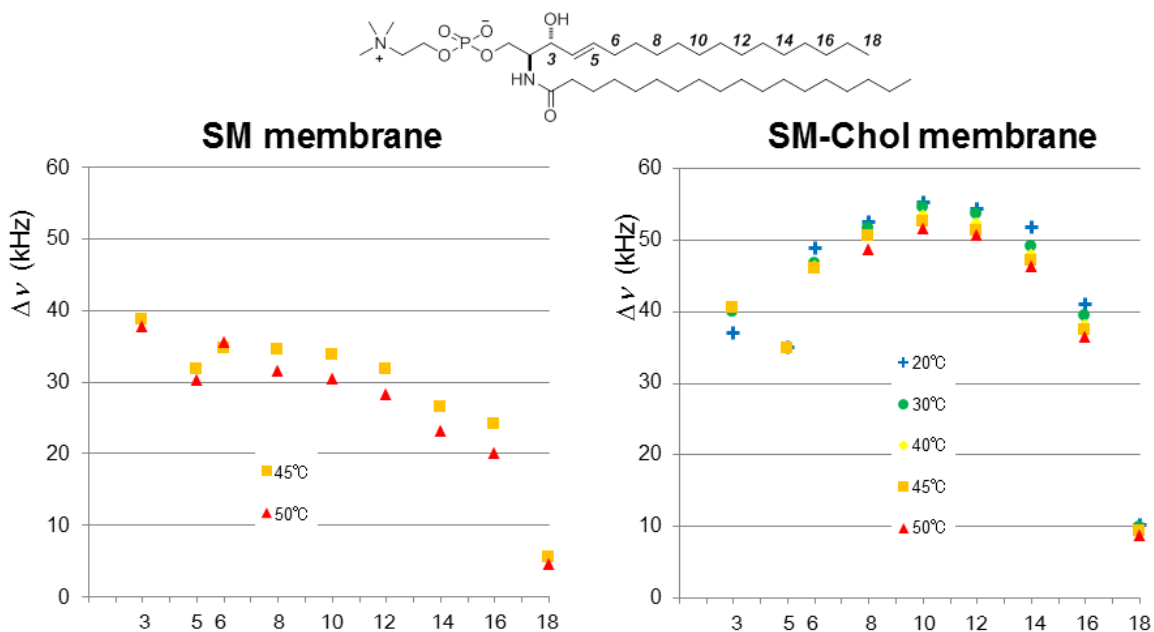


Figure S6. Temperature dependency of quadrupole couplings of the sphingosine chain of SSM. The upper region of the chain generally shows less temperature dependency than lower regions in both the presence and absence of Chol.

3. Supplementary Tables

Table S1 Quadrupolar couplings (kHz) of 3-*d*-SSM membranes

	SM	SM/Chol
20 °C	-	37.1
30 °C	-	39.9
40 °C	-	40.6
45 °C	38.6	40.5
50 °C	37.8	n.d.

Table S2 Quadrupolar couplings (kHz) of 5-*d*-SSM membranes

	SM	SM/Chol
20 °C	-	35.1
30 °C	-	34.9
40 °C	-	35.1
45 °C	31.7	34.9
50 °C	30.2	n.d.

Table S3 Quadrupolar couplings (kHz) of 6-*d*₂-SSM membranes

	SM	SM/Chol
20 °C	-	48.9
30 °C	-	46.7
40 °C	-	46.4
45 °C	34.7	46.0
50 °C	35.5	n.d.

Table S4 Quadrupolar couplings (kHz) of 8-*d*₂-SSM membranes

	SM	SM/Chol
20 °C	-	52.6
30 °C	-	51.7
40 °C	-	50.7
45 °C	34.5	50.5
50 °C	31.6	48.8

Table S5 Quadrupolar couplings (kHz) of 10- d_2 -SSM membranes

	SM	SM/Chol
20 °C	-	55.3
30 °C	-	54.6
40 °C	-	53.4
45 °C	33.8	52.6
50 °C	30.5	51.7

Table S6 Quadrupolar couplings (kHz) of 12- d_2 -SSM membranes

	SM	SM/Chol
20 °C	-	54.4
30 °C	-	53.6
40 °C	-	52.3
45 °C	31.8	51.3
50 °C	28.2	50.7

Table S7 Quadrupolar couplings (kHz) of 14- d_2 -SSM membranes

	SM	SM/Chol
20 °C	-	51.9
30 °C	-	49.1
40 °C	-	47.9
45 °C	26.5	47.1
50 °C	23.2	46.3

Table S8 Quadrupolar couplings (kHz) of 16- d_2 -SSM membranes

	SM	SM/Chol
20 °C	-	41.1
30 °C	-	39.4
40 °C	-	38.2
45 °C	24.1	37.4
50 °C	20.1	36.5

Table S9 Quadrupolar couplings (kHz) of 18- d_3 -SSM membranes

	SM	SM/Chol
20 °C	18.9	10.2
30 °C	18.5	9.8
40 °C	14.1	9.4
45 °C	5.5	9.4
50 °C	4.6	8.8

Table S10 Quadrupolar couplings (kHz) of 2'- d_2 -SSM membranes

	SM	SM/Chol
20 °C	-	34.1, 7.8
30 °C	-	29.1, 8.6
40 °C	-	29.6, 9.8
45 °C	25.6, 17.3	27.6, 9.9
50 °C	25.8, 17.6	27.3, 10.4

Table S11 Quadrupolar couplings (kHz) of 3'- d_2 -SSM membranes

	SM	SM/Chol
20 °C	-	42.1
30 °C	-	41.8
40 °C	-	41.6
45 °C	33.7	40.5
50 °C	32.7	40.2

Table S12 Quadrupolar couplings (kHz) of 4'- d_2 -SSM membranes

	SM	SM/Chol
20 °C	-	43.6
30 °C	-	43.2
40 °C	-	42.2
45 °C	31.0	41.2
50 °C	29.5	41.0

Table S13 Quadrupolar couplings (kHz) of 6'- d_2 -SSM membranes

	SM	SM/Chol
20 °C	-	51.7
30 °C	-	50.1
40 °C	-	49.3
45 °C	33.1	49.3
50 °C	30.1	48.6

Table S14 Quadrupolar couplings (kHz) of 8'- d_2 -SSM membranes

	SM	SM/Chol
20 °C	-	54.9
30 °C	-	54.3
40 °C	-	53.0
45 °C	31.5	51.5
50 °C	28.5	50.7

Table S15 Quadrupolar couplings (kHz) of 10'- d_2 -SSM membranes

	SM	SM/Chol
20 °C	-	54.8
30 °C	-	54.3
40 °C	-	52.9
45 °C	30.8	51.9
50 °C	27.4	50.8

Table S16 Quadrupolar couplings (kHz) of 12'- d_2 -SSM membranes

	SM	SM/Chol
20 °C	-	51.1
30 °C	-	50.7
40 °C	-	47.8
45 °C	28.3	47.1
50 °C	24.3	46.2

Table S17 Quadrupolar couplings (kHz) of 14'- d_2 -SSM membranes

	SM	SM/Chol
20 °C	-	42.6
30 °C	-	41.0
40 °C	-	39.5
45 °C	21.3	38.1
50 °C	18.5	37.7

Table S18 Quadrupolar couplings (kHz) of 16'- d_2 -SSM membranes

	SM	SM/Chol
20 °C	-	30.4
30 °C	-	29.3
40 °C	-	27.7
45 °C	15.3	26.5
50 °C	13.0	25.4

Table S19 Quadrupolar couplings (kHz) of 18'- d_3 -SSM membranes

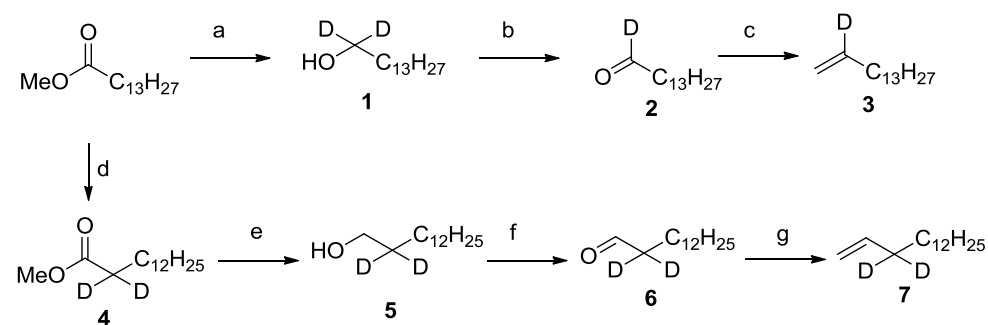
	SM	PSPC/Chol
20 °C	-	4.3
30 °C	-	4.0
40 °C	-	3.7
45 °C	1.6	3.5
50 °C	0	3.2

4. Synthesis of site-specifically-deuterated SSMs.

In this study, we selected *N*-stearoyl SM (SSM) because it is known to be the most abundant SM constituent in bovine brain (Jungalwala, F. B., Hayssen, V., Pasquini, J. M., and McCluer, R. H. (1979) *J. Lipid Res.* 20, 579–587). A series of site-specifically deuterated SSMs were synthesized as shown in Schemes S1 to S8 based on the previous report by Katsumura's group (Yamamoto, T., Hasegawa, H., Hakogi, T., and Katsumura, S. (2006) *Org. Lett.* 8, 5569–5572). 3-*d*-SSM and 10'-*d*₂-SSM were prepared as previously reported (Yamaguchi, T., Suzuki, T., Yasuda, T., Oishi, T., Matsumori, N., and Murata, M. (2012) *Bioorg. Med. Chem.* 20, 270-278).

4-1. Synthesis of 5-*d*- and 6,6-*d*₂-SSMs. For synthesis of 5-*d*- and 6,6-*d*₂-SSMs, we first prepared 2-*d*- and 3-*d*₂-pentadecenes, **3** and **7** (Scheme S1), both of which commenced with methyl myristate. Reduction of methyl myristate with LiAlD₄ to 2-*d*-myristic alcohol **1**, followed by oxidation to aldehyde **2** and Wittig reaction, provided 2-*d*-pentadecene **3**. For 5-*d*₂-pentadecene **7**, α -methylene hydrogens of methyl myristate were replaced by deuterons with excess MeOD in the presence of MeONa to afford **4**, which was subjected to the same reactions as above to give 3-*d*₂-pentadecene **7**.

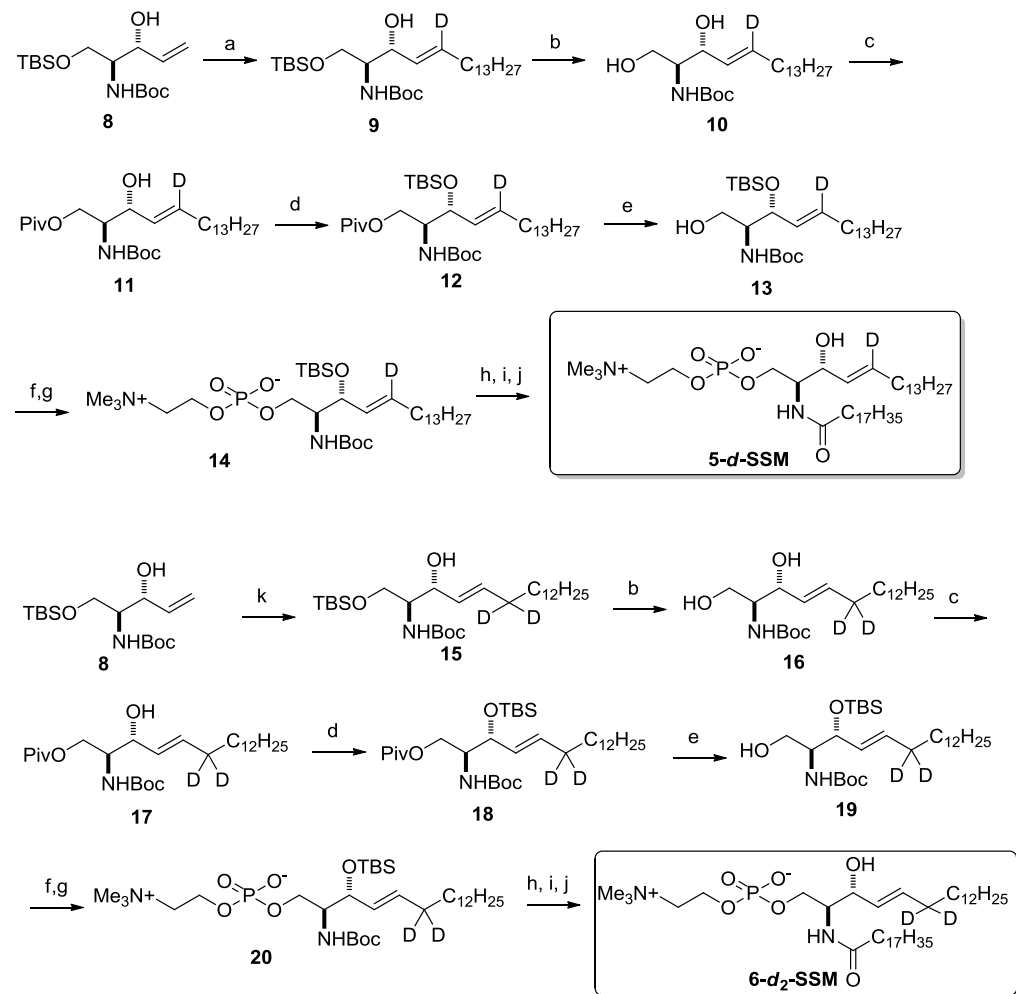
Scheme S1. Synthesis of 2-*d*- and 3-*d*₂-pentadecenes, **3** and **7**



Reagents and conditions: (a) LiAlD₄, THF, rt, 30 min, quant; (b) SO₃Py, Et₃N, DMSO, CH₂Cl₂, rt, 1.5 h; (c) CH₃Ph₃PBr, *t*-BuOK, THF, rt, 1 h, 72% (2 steps) ; (d) MeONa, MeOD, reflux 13 h, 63%, deuterium rate 80%; (e) LiAlH₄, THF, 0 °C, quant; (f) TEMPO, PhI(OAc)₂, CH₂Cl₂, rt, 4 h; (g) CH₃Ph₃PBr, *t*-BuOK, THF, rt, 1 h 2 steps, 52%.

Then deuterated spingosines (**9** and **15**) were synthesized by olefin cross metathesis reaction between the deuterated pentadecene (**3** or **7**) and olefin **8** that was prepared as previously reported by Katsumura's group (Yamamoto, T., Hasegawa, H., Hakogi, T., and Katsumura, S. (2006) *Org. Lett.* 8, 5569–5572). Introduction of a choline group and a stearoyl acyl chain furnished 5-*d*- and 6-*d*-SSMs (Scheme S2).

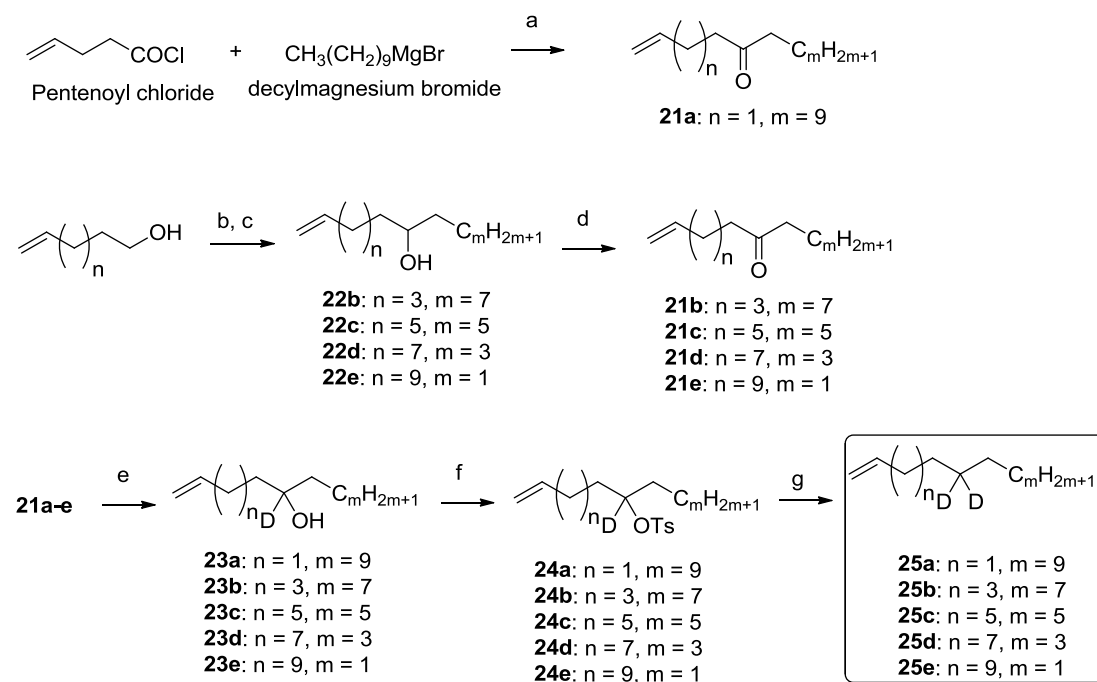
Scheme S2. Synthesis of 5-*d*- and 6-*d*-SSMs.



Reagents and conditions: (a) Grubbs 2nd, *p*-quinone, **3**, CH₂Cl₂, 55 °C, 1.5 h, 53%; (b) TBAF, THF, rt, 25 min, quant; (c) PivCl, pyridine, -30 °C, 2.5 h, 94%; (d). TBSOTf, 2,6-lutidine, CH₂Cl₂, 0 °C, 30 min, 88%; (e) DBU, MeOH, rt, 14 h, 98%; (f) 2-chloro-1,3,2-dioxaphospholane 2-oxide, Et₃N, DMAP, benzene, 0 °C, 1 h; (g) Me₃N, CH₃CN, 85 °C, 72 h, 68% (2 steps); (h) TFA, CH₂Cl₂, 0 °C, 30 min; (i) *p*-nitrophenyl stearate, Et₃N, DMAP, THF, rt, 13 h; (j) TBAF, THF, rt, 26 h, 73% (3 steps); (k) Grubbs 2nd, *p*-quinone, **7**, CH₂Cl₂, 55 °C, 1.5 h, 53%.

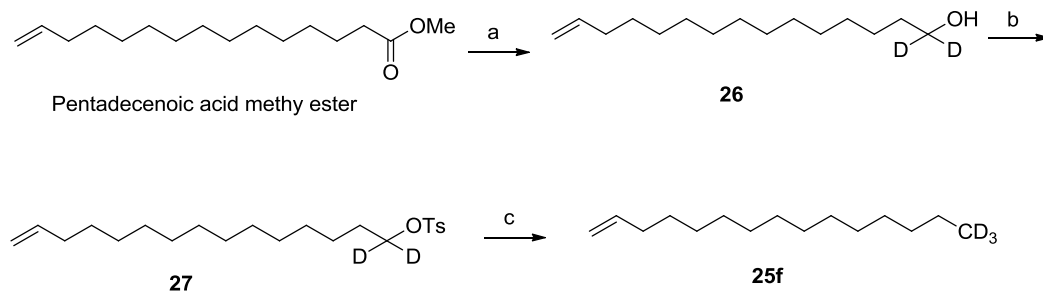
4-2. Synthesis of 8-*d*₂-, 10-*d*₂-, 12-*d*₂-, 14-*d*₂-, 16-*d*₂-, and 18-*d*₃-SSMs. Preparation procedure of these labeled SSMs was slightly modified from that of 6-*d*₂-SSM in order to introduce deuterated olefins by Grubbs olefination at a nearly final step of the synthesis (Schemes S3 to S5). Briefly, 5-*d*₂-, 7-*d*₂-, 9-*d*₂-, and 11-*d*₂-pentadecenes **25a-e** were prepared from corresponding keto-olefins **21a-e** by reduction with NaBD₄ and then with LiAlD₄ (Schemes S3). 18-*d*₃-pentadecene **25f** was prepared from 14-pentadecenoic acid methyl ester, which was reduced with LiAlD₄ to give primary alcohol **26** (Scheme S4). The alcohol was converted to tosylate **27**, which was further reduced with LiAlD₄ to provide 18-*d*₃-pentadecene **25f** (Scheme S4).

Scheme S3 Synthesis of pentadecenes-*d*₂ **25a-e**



Reagents and conditions: (a) CH₃(CH₂)₉MgBr, CuI, THF, 0 °C, 2 h; (b) IBX, DMSO, rt, 4 h; (c) CH₃(CH₂)_mMgBr (m=10-n), diethyl ether, rt, 18 h, 69-85% (2 steps); (d) SO₃·Py, Et₃N, DMSO, CH₂Cl₂, rt, 14 h, 86%; (e) NaBD₄, CHCl₃/MeOH, 0 °C, 1 h, 97% f) TsCl, Et₃N, DMAP, CH₂Cl₂, rt, 5.5 h, 81%; (g) LiAlD₄, diethyl ether, rt, 4 h, 88%.

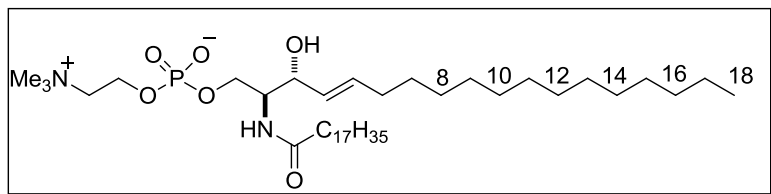
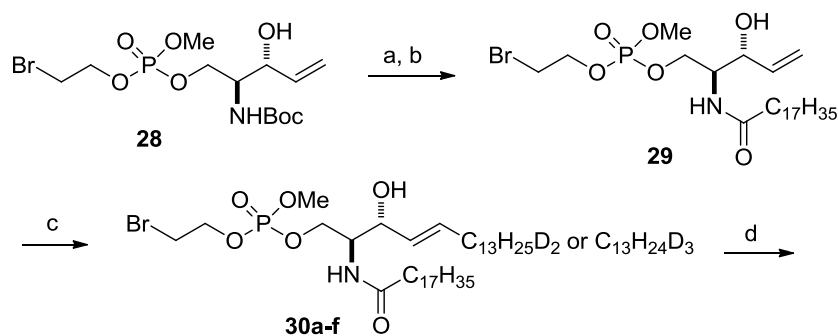
Scheme S4. Synthesis of pentadecenes- d_3 **25f**



Reagents and conditions: (a) LiAlD_4 , THF, rt, 30 min, 87%; (b) TsCl , Et_3N , DMAP, CH_2Cl_2 , rt, 3 h, 95%; (c) LiAlD_4 , ether, rt, 4 h, 79%.

Following the reported procedure (Yamamoto, T., et al. (2006) *Org. Lett.* 8, 5569–5572), cross metathesis of these deuterated pentadecenes **25a-f** with **29** prepared from known compound **28** afforded **30a-f**. Treatment with trimethyl amine furnished objective deuterated SSMs (Scheme S5).

Scheme S5. Completion of synthesis of 8- d_2 -, 10- d_2 -, 12- d_2 -, 14- d_2 -, 16- d_2 -, and 18- d_3 -SSM

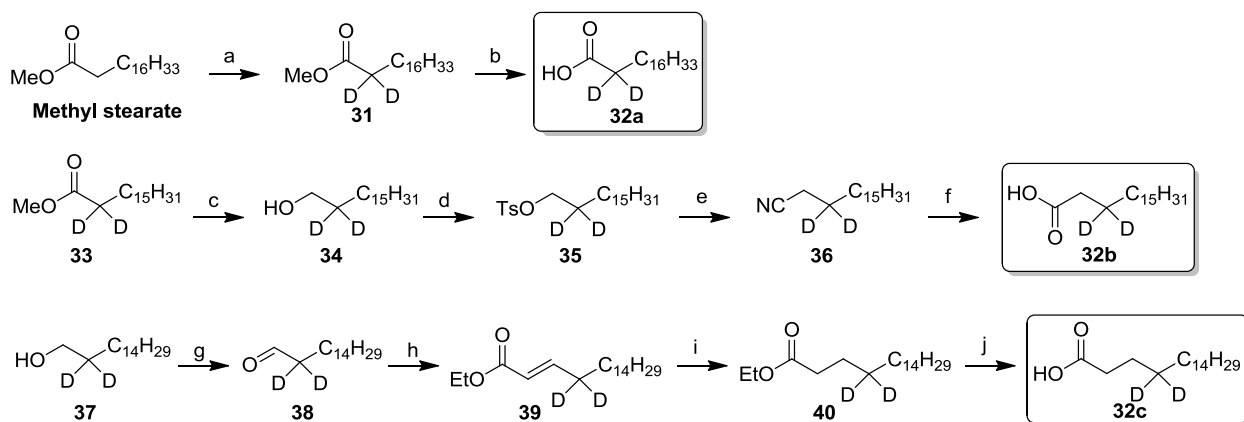


8- d_2 -, 10- d_2 -, 12- d_2 -, 14- d_2 -, 16- d_2 - and 18- d_3 -SSM

Reagents and conditions: (a) TFA, CH_2Cl_2 , 0 °C, 90 min; (b) *p*-nitrophenyl stearate, DIPEA, DMAP, THF, rt, 2 h; (c) Grubbs 2nd, *p*-quinone, **25a-f**, CH_2Cl_2 , 55 °C, 2 h; (d) trimethylamine, MeOH, rt.

4-3. Synthesis of 2'-d₂-, 3'-d₂-, 4'-d₂-, 6'-d₂-, 8'-d₂-, 10'-d₂-, 12'-d₂-, 14'-d₂-, 16'-d₂-, 18'-d₃-SSMs. A series of deuterated stearic acids (**32a-c** and **45a-f**) were synthesized as shown in Schemes S6 and S7. 2-d₂-stearic acid **32a** was synthesized from methyl stearate, whose α-methylene hydrogens were replaced by deuterons with excess MeOD in the presence of MeONa to afford **31**. Alkaline hydrolysis of **31** with NaOH produced 2-d₂-stearic acid **32a** with 71-97% deuterium labeling rate (Scheme S6). Synthesis of 3-d₂-stearic **32b** commenced with methyl 2-d₂-heptadecanoic acid **33**, which was prepared from methyl heptadecanoate in a similar manner to the synthesis of **31**. Reduction of **33** with LiAlH₄ gave 2-d₂-hexadecanol **34**, which was converted to nitrile **36** via tosyl ester **35**. Hydrolysis of **36** with NaOH gave 3-d₂-stearic acid **32b** (Scheme S6). 4-d₂-stearic acid **32c** was synthesized via 2-d₂-hexadecanol **37** obtained from methyl palmitate in a similar manner to the preparation of **34**. Oxidation of the alcohol **37** to aldehyde **38**, followed by Wittig reaction with Ph₃P=CHCO₂Et, provided olefin **39**. Hydrogenation of the olefin **39** gave **40**, which was hydrolyzed with NaOH to afford 4-d₂-stearic acid **32c** (Scheme S6).

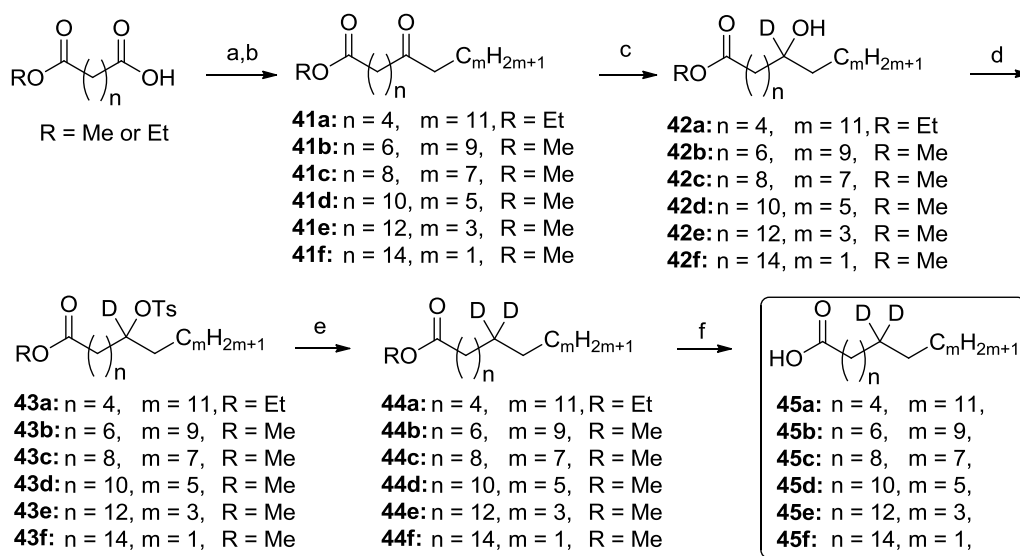
Scheme S6. Preparation of 2-d₂-, 3-d₂-, and 4-d₂-stearic acids (**32a-c**)



Reagents and conditions: (a) MeONa, MeOD, 80 °C, 24 h, 88%; (b) NaOH, MeOH-THF, 60 °C, 2 h, 94%; (c) LiAlH₄, THF, rt, 7 h, 93%; (d) TsCl, Et₃N, DMAP, CH₂Cl₂, rt, 9 h, quant.; (e) KCN, 18-crown-6, DMF, rt, 2 h, 59%; (f) NaOH, EtOH, 100 °C, 16 h, 91%; (g) SO₃·Py, Et₃N, DMSO, CH₂Cl₂, rt, 2 h; (h) Ph₃P=CHCO₂Et, THF, rt, 4 h then 40 °C, 17.5 h, 77% (2 steps); (i) H₂, Pd-C, THF, rt, 24 h, 94%; (j) NaOH, MeOH, THF, 60 °C, 2 h, quant.

As shown in Scheme S7, 6-*d*₂-, 8-*d*₂-, 10-*d*₂-, 12-*d*₂-, 14-*d*₂-, and 16-*d*₂-stearic acids (**45a-f**) were prepared from corresponding known keto-stearic acid esters (**41a-f**), which were obtained from mono esters of dicarboxylic acids, except commercially available **41c**, through conversion to acid chlorides and coupling with Grignard reagents. The keto esters **41a-f** were reduced by NaBD₄, and the resulting mono-deuterated alcohols (**42a-f**) were converted to tosyl esters (**43a-f**), which were again reduced with NaBD₄ in DMSO at 80 °C to give *d*₂-stearic acid esters (**44a-f**). Hydrolysis of the esters with NaOH afforded *d*₂-stearic acids (**45a-f**).

Scheme S7. Preparation of 6-*d*₂-, 8-*d*₂-, 10-*d*₂-, 12-*d*₂-, 14-*d*₂- and 16-*d*₂-stearic acids (**45a-f**)

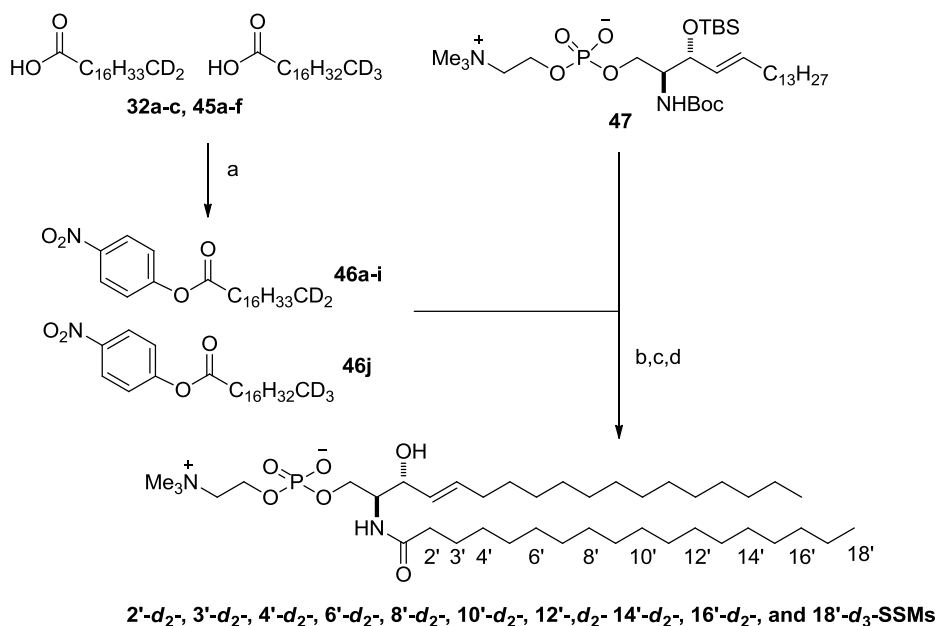


Reagents and conditions: (a) SOCl₂, 80 °C, 16 h; (b) CH₃(CH₂)_mMgBr (m=15-n), CuI, THF, rt, 2 h, 30-64 % (2 steps); (c) NaBD₄, MeOH-CHCl₃ (2:1), 0 °C, 2 h, 93%; (d) TsCl, Et₃N, DMAP, CH₂Cl₂, rt, 45 h, 74%; (e) NaBD₄, DMSO, 80 °C, 14 h, 54%; (f) NaOH, MeOH, THF, 60 °C, 2 h, quant.

Thus obtained *d*₂-stearic acids **32a-c** and **45a-f** were converted to *p*-nitrophenyl stearates (**46a-i**). Similarly commercially available 18-*d*₃-stearic acid was transformed to *p*-nitrophenyl stearates (**46j**). These *p*-nitrophenyl stearates (**46a-j**) were coupled with **47** (Scheme S8), which was prepared in a similar manner

to the synthesis of **14** in Scheme S2. Removal of the TBS group completed the synthesis of site-specifically deuterated *N*-stearoyl SMs (Scheme S8).

Scheme S8. Completion of synthesis of site specifically deuterated sphingomyelins



Reagents and conditions: (a) DCC, *p*-nitrophenol, THF, rt, 18 h, 80%; (b) TFA, CH₂Cl₂, 0 °C, 3 h; (c) Et₃N, DMAP, THF, rt, 15 h; (d) TBAF, THF 0 °C, 17 h.

5. Experimental section of synthesis

General information for synthesis. Chemicals and solvents were purchased from Nacalai Tesque, Aldrich, TCI, and KANTO Chemicals Inc., and used without further purification unless otherwise noted. Thin layer chromatography and column chromatography were Merck precoated silica gel 60 F-254 plates and silica gel 60 (100-200 μm), respectively. Proton nuclear magnetic resonance spectra were collected on a JEOL ECA 500 (500 MHz) or a JEOL ECS 400 (400 MHz). Mass spectrometry was performed using LCQ-DECA (Thermo quest), and high resolution mass spectra (HRMS) were recorded on a LTQ-Orbitrap XL.

Synthesis of 1. To a solution of methyl myristate (3.09 g, 13 mmol) in THF (40 ml) was added LiAlD_4 (410 mg, 9.77 mmol) at 0 °C. After the reaction mixture was stirred for 30 min at 0 °C, the reaction was quenched with water. The resulting mixture was filtered with Celite, and the filtrate was evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 5/1) afforded **1** (2.69 g, 12 mmol, quant) as a white solid. R_f 0.25 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 1.56 (2H, t, $J = 7.5$ Hz, H2), 1.23 (22H, m, H3~H13), 0.89 (3H, t, $J = 7.0$ Hz, H14).

Synthesis of 2. To a solution of **1** (2.67 g, 12 mmol) in dichloromethane (38 ml) were added DMSO (6.2 ml, 96 mmol), triethylamine (8.4 ml, 60 mmol), and $\text{SO}_3\text{-Py}$ (6.2 g, 38 mmol) at 0 °C. After the reaction mixture was stirred for 1.5 h at room temperature, the reaction was quenched with saturated aqueous NH_4Cl , and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with saturated aqueous NH_4Cl and brine, dried over Na_2SO_4 , filtered, and evaporated to give the crude products. Purification by flash silica gel column chromatography (hexane/ethyl acetate = 20/1) afforded aldehyde **2** as a white solid. R_f 0.50 (silica gel, hexane/ethyl acetate = 10/1).

Synthesis of 3. To a stirred solution of $\text{CH}_3\text{Ph}_3\text{P}^+\text{Br}^-$ and *t*-BuOK in THF (30 ml) was added aldehyde **2** at 0 °C. After the reaction mixture was stirred for 1 h at 0 °C, the reaction was quenched with water, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with brine, dried over Na_2SO_4 , filtered, and evaporated to give the crude products. Purification by flash silica gel column chromatography (hexane) afforded olefin **3** as a colorless oil (1.83 g, 8.66 mmol, 72%, 2 steps). R_f 0.87 (silica gel, hexane); ^1H NMR (500 MHz, CDCl_3) δ 4.96 (1H, s, H1), 4.91 (1H, , s, H1), 2.01 (2H, t, $J = 7.0$ Hz, H3), 1.35 (2H, tt, $J = 7.0, 7.0$ Hz, H4), 1.24 (20H, m, H5-H14), 0.86 (3H, t, $J = 7.0$ Hz, H15); MS (ESI) no detection.

Synthesis of 4. To a solution of methyl myristate (2.02 g, 8.3 mmol) in CH_3OD (30 ml) was added CH_3ONa (700 mg, 12.9 mmol) at room temperature. After the reaction mixture was refluxed for 24 h, the reaction was quenched with 1 M DCl in D_2O , and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with brine, dried over Na_2SO_4 , filtered, and evaporated to give the crude products. Purification by flash silica gel column chromatography (hexane) afforded **4** as a colorless solid (1.28 g, 5.2 mmol, 63%, deuterium rate 80%). R_f 0.61 (silica gel, hexane/ethyl acetate = 10/1); ^1H NMR (500 MHz, CDCl_3) δ 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 1.58 (2H, t, $J = 6.5$ Hz, H3), 1.28 (2H, m, H4-H13), 0.86 (3H, t, $J = 7.0$ Hz, H14).

Synthesis of 5. To a solution of methyl myristate **4** (50 mg, 0.20 mmol) in THF (2 ml) was added LiAlH_4 (13.4 mg, 0.35 mmol) at 0 °C. After the reaction mixture was stirred for 2 h at 0 °C, the reaction was quenched with water. The resulting mixture was filtered with Celite, and the filtrate was evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 5/1) afforded **5** (44.8 mg, 0.2 mmol, quant) as a white solid. R_f 0.25 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 3.62 (2H, brt, $J = 5.5$ Hz, H1), 1.28 (22H, m, H3~H13), 0.86 (3H, t, $J = 7.0$ Hz, H14).

Synthesis of 6. To a solution of alcohol **5** (44.8 mg, 0.20 mmol) and TEMPO (2.98 mg, 19 μ mol) in dichloromethane (4.7 ml) was added $\text{PhI}(\text{OAc})_2$ (204 mg, 0.63 mmol) at 0 °C. After the reaction mixture was stirred for 4 h at 0 °C, the reaction was quenched with saturated aqueous NaHCO_3 and saturated aqueous $\text{Na}_2\text{S}_2\text{O}_3$, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with brine, dried over Na_2SO_4 , filtered and evaporated to the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **6** as a yellow oil. R_f 0.50 (silica gel, hexane/ethyl acetate = 10/1).

Synthesis of 7. To a stirred solution of $\text{CH}_3\text{Ph}_3\text{P}^+\text{Br}^-$ (189.3 mg, 0.52 mmol) and *t*-BuOK (52.7 mg, 0.47 mmol) in THF (1 ml) was added aldehyde **6** at 0 °C. After the reaction mixture was stirred for 1 h at 0 °C, the reaction was quenched with water, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with brine, dried over Na_2SO_4 , filtered, and evaporated to give the crude products. Purification by flash silica gel column chromatography (hexane) afforded olefin **7** as a colorless oil (23.9 mg, 0.11 mmol, 52%, 2 steps). R_f 0.87 (silica gel, hexane); ^1H NMR (500 MHz, CDCl_3) δ 5.80 (1H, dd, $J = 16.5, 14$ Hz, H2), 4.97 (1H, dd, $J = 17.0, 2.5$ Hz, H1), 4.91 (1H, dd, $J = 10.0, 2.0$ Hz, H1), 1.34 (2H, m, H4), 1.24 (20H, m, H5-H14), 0.86 (3H, t, $J = 7.0$ Hz, H15); MS (ESI) no detection.

Synthesis of 9. Compound **8** was prepared as previously reported (*Org. Lett.* **2006**, *8*, 5569–5572). To a solution of **8** (200 mg, 0.6 mmol) and **3** (890 mg, 4.2 mmol) in dichloromethane (3 ml) were added Grubbs 2nd catalyst (25.5 mg, 30 μ mol) and *p*-benzoquinone (9.0 mg, 83 μ mol) in dichloromethane (3 ml) by cannula at 55 °C. After the reaction mixture was stirred for 2 h at 55 °C, solvent was removed by evaporation to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 20/1) afforded *trans* olefin **9** (164.3 mg, 0.32 mmol, 53%) as a brown oil; R_f 0.27 (silica gel,

hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 5.46 (1H, d, J = 6.0 Hz, H4), 5.20 (1H, d, J = 5.5 Hz, NH), 4.16 (1H, brm, H3), 3.90 (1H, dd, J = 3.0, 10.5 Hz, H1), 3.71 (1H, dd, J = 2.0, 11.0 Hz, H1), 3.53 (1H, brm, H2), 2.01 (2H, t, J = 7.5 Hz, H6), 1.41 (9H, s, Boc), 1.33 (2H, tt, J = 7.0, 7.0 Hz, H7), 1.22 (20H, m, H8-H17), 0.87 (9H, s, *Si*-Bu), 0.84 (3H, t, J = 7.0 Hz, H18), 0.037 (6H, s, SiMe), 0.034 (6H, s, SiMe); ^{13}C NMR (125 MHz, CDCl_3) δ 155.7, 132.9, 132.7, 132.6, 129.3, 79.4, 74.5, 63.4, 54.5, 32.2, 31.9, 29.7, 29.6, 29.6, 29.5, 29.3, 29.2, 29.1, 28.4, 25.8, 22.7, 18.1, 14.1, -5.65; MS (ESI) m/z calcd for $\text{C}_{29}\text{H}_{59}\text{DNO}_4\text{SiNa}$ ($\text{M}+\text{Na}^+$) 537.4, found 537.3.

Synthesis of 10. To a solution of **9** (164.3 mg, 0.32 mmol) in THF (5 ml) was added TBAF (1.0 M in THF, 0.64 ml, 0.64 mmol) at 0 °C. After the reaction mixture was stirred for 25 min at room temperature, the reaction was quenched with water, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with brine, dried over Na_2SO_4 , and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 1/1) afforded diol **10** (133.3 mg, quant) as a white solid; R_f 0.31 (silica gel, hexane/ethyl acetate = 1/1); ^1H NMR (500 MHz, CDCl_3) δ 5.48 (1H, d, J = 6.5 Hz, H4), 5.31 (1H, brm, NH), 4.27 (1H, dd, J = 6.5, 6.5 Hz, H3), 3.89 (1H, dd, J = 11.0, 3.0 Hz, H1), 3.71 (1H, dd, J = 11.0, 3.5 Hz, H1), 3.56 (1H, brm, H2), 2.83 (2H, brm, OH), 2.01 (2H, t, J = 7.5 Hz, H6), 1.42 (9H, s, Boc), 1.34 (2H, tt, J = 7.0, 7.0 Hz, H7), 1.23 (20H, m, H8-H17), 0.85 (3H, t, J = 7.0 Hz, H18); ^{13}C NMR (125 MHz, CDCl_3) δ 156.2, 134.0, 133.7, 133.5, 128.8, 79.8, 74.6, 62.6, 55.4, 32.1, 31.9, 29.7, 29.6, 29.5, 29.3, 29.2, 29.1, 28.4, 22.7, 14.1; MS (ESI) m/z calcd for $\text{C}_{23}\text{H}_{44}\text{DNNaO}_4^+$ ($\text{M}+\text{Na}^+$) 423.3, found 423.3.

Synthesis of 11. To a solution of **10** (133.3 mg, 0.33 mmol) in pyridine (4.5 ml) was added PivCl (62 μl , 0.50 mmol) at -30 °C. After the reaction mixture was stirred for 30 min at -30 °C, another PivCl (62 μl , 0.50 mmol) was added, and the mixture was stirred for further 2 h at -30 °C. The reaction was quenched

with H₂O, and the resultant mixture was extracted with ethyl acetate. The organic layer was washed with saturated aqueous KHSO₄ and brine, dried over Na₂SO₄, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded pivarate **11** (150.9 mg, 0.31 mmol, 94%) as a white solid; R_f 0.71 (silica gel, hexane/ethyl acetate = 3/1); ¹H NMR (500 MHz, CDCl₃) δ 5.44(1H, d, *J* = 6.5 Hz, H4), 4.85 (1H, d, *J* = 7.5 Hz, NH), 4.21 (1H, brm, H2), 4.12 (1H, brm, H3), 4.08 (1H, dd, *J* = 11.5, 4.0 Hz, H1), 3.90 (1H, brm, H2), 2.00 (2H, t, *J* = 7.0 Hz, H6), 1.39 (9H, s, Boc), 1.33 (2H, tt, *J* = 6.5, 6.5 Hz, H7), 1.21 (20H, m, H8~H17), 1.16 (9H, s, Piv), 0.83 (3H, t, *J* = 7.0 Hz, H18); ¹³C NMR (125 MHz, CDCl₃) δ 178.6, 155.8, 134.3, 134.1, 134.0, 128.1, 79.7, 73.3, 63.0, 54.2, 38.8, 32.2, 31.9, 29.6, 29.5, 29.4, 29.3, 29.2, 29.0, 28.3, 27.1, 27.0, 22.6, 14.1; MS (ESI) *m/z* calcd for C₂₈H₅₂DNNaO₅⁺ (M+Na⁺) 507.4, found 507.2

Synthesis of 12. To a solution of pivarate **11** (150.9 mg, 0.31 mmol) in dichloromethane (6 ml) were added 2,6-lutidine (64 μl, 0.55 mmol) and TBSOTf (0.10 ml, 0.44 mmol) at 0 °C under Ar atmosphere. After the reaction mixture was stirred for 30 min at 0 °C, the reaction was quenched with water, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with 1 M aqueous KHSO₄ and brine, dried over Na₂SO₄, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded TBS ether **12** (163.2 mg, 0.27 mmol, 88%) as a colorless oil; R_f 0.48 (silica gel, hexane/ethyl acetate = 4/1); ¹H NMR (500 MHz, CDCl₃) δ 5.36 (1H, d, *J* = 6.5 Hz, H4), 4.62 (1H, d, *J* = 9.0 Hz, NH), 4.21 (2H, brm, H1, H3), 4.08 (1H, dd, *J* = 11.5, 4.5 Hz, H1), 3.82 (1H, brm, H2), 1.98 (2H, t, *J* = 7.0 Hz, H6), 1.39 (9H, s, Boc), 1.33 (2H, tt, *J* = 7.0, 7.0 Hz, H7), 1.23 (20H, m, H8~H17), 1.17 (9H, s, Piv), 0.86 (9H, s, *Sit*-Bu), 0.86 (3H, t, *J* = 7.5 Hz, H18), 0.014 (3H, s, SiMe), -0.016 (3H, s, SiMe).

Synthesis of 13. To a solution of TBS ether **12** (83.5 mg, 0.14 mmol) in methanol (1.3 ml) was added DBU

(83.5 mg, 0.14 mmol) under Ar at 0 °C. After the reaction mixture was stirred for 14 h at room temperature, the reaction was quenched with saturated aqueous NH₄Cl, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with saturated aqueous NH₄Cl and brine, dried over MgSO₄, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 5/1) afforded primary alcohol **13** (67.4 mg, 0.13 mmol, 98%) as a colorless oil; R_f 0.33 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 5.42 (1H, d, *J* = 6.0 Hz, H4), 5.32 (1H, d, *J* = 8.0 Hz, NH), 4.45 (1H, brm, H3), 4.01 (1H, d, *J* = 11.5 Hz, H1), 3.54 (1H, ddd, *J* = 11.0, 11.0, 3.0 Hz, H1), 3.43 (1H, brm, OH), 2.97 (1H, d, *J* = 9.5 Hz, 2H), 2.00 (2H, t, *J* = 7.0 Hz, H6), 1.43 (9H, s, Boc), 1.33 (2H, tt, *J* = 7.0, 7.0 Hz, H7), 1.23 (20H, m, H8~H17), 0.88 (9H, s, Sit-Bu), 0.85 (3H, t, *J* = 7.0 Hz, H18), 0.049 (3H, s, SiMe), 0.06 (3H, s, SiMe); ¹³C NMR (125 MHz, CDCl₃) δ 155.7, 133.1, 133.0, 132.8, 129.2, 79.4, 76.3, 62.3, 55.2, 32.1, 29.7, 29.6, 29.5, 29.4, 29.1, 29.0, 28.4, 25.8, 22.7, 18.0, 14.1, -4.54, -5.17; MS (ESI) *m/z* calcd for C₂₉H₅₉DNO₄SiNa (M+Na⁺) 537.4, found 537.5.

Synthesis of 14. To a solution of primary alcohol **13** (67.4 mg, 0.13 mmol) in benzene (1.3 ml) were added Et₃N (0.18 ml, 1.3 mmol), DMAP (4.4 mg, 0.036 mmol), and 2-chloro-1,3,2-dioxaphospholane-2-oxide (80 μl, 0.87 mmol) at 0 °C. After the reaction mixture was stirred for 1 h at 0 °C, the reaction mixture was filtered through glass filter under Ar atmosphere. The filtrate was azeotropically evaporated with toluene three times. The residue was dissolved in acetonitrile (20 ml) and transferred to a pressure bottle. Anhydrous Me₃N dried over metallic sodium was added to the solution by cannula at -78 °C. After the reaction mixture was stirred at 80 °C for 72 h, the resulting mixture was evaporated to give the crude products. Purification by silica gel column chromatography (CHCl₃/MeOH/H₂O = 65/25/4) afforded sphingosine-1-phosphoryl choline **14** (54.4 mg, 0.08 mmol, 68%, 2 steps) as a colorless oil; R_f 0.28 (silica gel, CHCl₃/MeOH/H₂O = 65/25/4); ¹H NMR (500 MHz, CD₃OD) δ 5.56 (1H, d, *J* = 8.5 Hz, H5), 4.24 (2H, br, α), 4.20 (1H, dd, *J* = 7.0, 7.0 Hz, H3), 4.00 (1H, m, H1), 3.94 (1H, m, H1), 3.61 (3H, m, β and H2),

3.21 (9H, s, N⁺Me₃), 2.02 (2H, dt, $J = 7.0, 7.0$ Hz, H6), 1.46 (9H, s, Boc), 1.28 (22H, m, H7~H17), 0.90 (9H, s, *Sit*-Bu), 0.89 (3H, t, $J = 7.0$ Hz, H18), 0.083 (3H, s, SiMe), 0.032 (3H, s, SiMe); MS (ESI) m/z calcd for C₃₄H₇₀DN₂NaO₇PSi⁺ (M+Na⁺) 702.5, found 702.3.

Synthesis of 5-*d*-SSM. To a solution of sphingosine-1-phosphorylcholine **14** (54.4 mg, 80 μ mol) in CH₂Cl₂ (2 ml) was added trifluoroacetic acid (0.36 ml, 4.8 mmol) at 0 °C. After the reaction mixture was stirred for 30 min at 0 °C, the reaction was quenched with Et₃N (2 ml), and the resulting mixture was evaporated to give the crude amine, which was subjected to the next reaction without further purification. To a solution of the crude amine in THF (2 mL) were added DMAP (13.5 mg, 0.11 mmol), and *p*-nitrophenyl octadecanoate (63.8 mg, 0.11 mmol). After the reaction mixture was stirred for 13 h at room temperature, the mixture was evaporated. Purification by silica gel column chromatography (CHCl₃/MeOH/H₂O = 65/25/4) afforded TBS-protected 5-*d*-SSM as a white solid. To a solution of the protected 5-*d*-SSM in THF (1 ml) was added TBAF (1.0 M in THF, 0.32 ml, 0.32 mmol) at 0 °C. After the reaction mixture was stirred for 21 h at room temperature, MeOH was added to the solution and the solvent was evaporated. The resultant residue was washed with acetone on Celite-Pad and dissolved with CHCl₃/MeOH (1/1). After the solvent was removed by evaporation, purification by silica gel column chromatography (CHCl₃/MeOH/H₂O = 65/25/4) afforded 5-*d*-SSM (42.3 mg, 58 μ mol, 73%, 3step) as a white solid; R_f 0.31 (silica gel, CHCl₃/MeOH/H₂O = 65/45/8); ¹H NMR (500 MHz, CD₃OD) δ 5.43 (1H, d, $J = 8.0$ Hz, H4), 4.27 (2H, brm, α), 4.08 (1H, m, 1H), 4.04 (1H, dd, $J = 7.5, 7.5$ Hz, H3), 3.96 (2H, m, H1 and H2), 3.62 (2H, t, $J = 7.0$ Hz, β), 3.21 (9H, s, N⁺Me₃), 2.17 (2H, td, $J = 7.0, 3.0$ Hz, H2'), 2.01 (2H, t, $J = 7.0$ Hz, H6), 1.35 (2H, m, H7), 1.41 (48H, m, H8~H17 and H4'~H17'), 0.89 (6H, t, $J = 7.0$ Hz, H18 and H18'); ¹³C NMR (125 MHz, CD₃OD) δ 175.9, 134.7, 131.2, 72.5, 67.5, 65.9, 65.8, 60.4, 60.4, 55.3, 55.2, 54.7, 37.4, 33.4, 33.1, 30.8, 30.7, 30.7, 30.5, 30.5, 30.4, 27.2, 23.8, 14.5; ESI-HRMS m/z calcd for C₄₁H₈₂DN₂O₆PNa(M+Na)⁺ 754.5944, found 754.5953.

Synthesis of 15. Compound **15** was prepared from **8** and **7** in the same manner as the synthesis of **9**.

15: brown oil; R_f 0.27 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 5.73 (1H, d, J = 15.5 Hz, H5), 5.59 (1H, dd, J = 15.5, 5.5 Hz, H4), 5.21 (1H, brm, NH), 4.17 (1H, brm, H3), 3.92 (1H, dd, J = 10.5, 2.0 Hz, H1), 3.73 (1H, d, J = 9.5 Hz, H1), 3.55 (1H, brm, OH), 3.30 (1H, d, J = 8.0 Hz, H2), 1.53 (9H, s, Boc), 1.34 (2H, m, H7), 1.26 (20H, m, H8-H17), 0.88 (9H, s, Sit-Bu), 0.86 (3H, t, J = 7.0 Hz, H18), 0.053 (3H, s, SiMe), 0.049 (3H, s, SiMe); ^{13}C NMR (125 MHz, CDCl_3) δ 155.7, 133.0, 129.4, 79.4, 74.5, 63.4, 54.5, 31.9, 30.0, 29.5, 29.3, 29.1, 29.0, 28.4, 25.8, 22.7, 18.1, 14.1; MS (ESI) m/z calcd for $\text{C}_{29}\text{H}_{57}\text{D}_2\text{NNaO}_4\text{Si}^+$ ($\text{M}+\text{Na}^+$) 538.3, found 538.2.

Synthesis of 16. Compound **16** was prepared from **15** in the same manner as the synthesis of **10**.

16: white solid; R_f 0.31 (silica gel, hexane/ethyl acetate = 1/1); ^1H NMR (500 MHz, CDCl_3) δ 5.75 (1H, d, J = 15.5 Hz, H5), 5.51 (1H, dd, J = 15.5, 6.5 Hz), 5.28 (1H, d, J = 6.0 Hz, NH), 4.30 (1H, brm, H3), 3.91 (1H, brd, J = 12.0 Hz, H1), 3.68 (1H, brm, H1), 3.58 (1H, brm, H2), 2.51 (2H, brm, OH), 1.43 (9H, s, Boc), 1.33 (2H, m, H7), 1.23 (20H, m, H8-H17), 0.86 (3H, t, J = 7.0 Hz, H18); ^{13}C NMR (125 MHz, CDCl_3) δ 156.2, 134.0, 129.0, 79.7, 74.6, 62.5, 55.4, 31.9, 30.0, 29.5, 29.3, 29.1, 29.0, 28.9, 28.3, 22.7, 14.1; MS (ESI) m/z calcd for $\text{C}_{23}\text{H}_{43}\text{D}_2\text{NNaO}_4^+$ ($\text{M}+\text{Na}^+$) 424.3, found 424.2.

Synthesis of 17. Compound **17** was prepared from **16** in the same manner as the synthesis of **11**.

17: white solid; R_f 0.71 (silica gel, hexane/ethyl acetate = 3/1); ^1H NMR (500 MHz, CDCl_3) δ 5.72 (1H, d, J = 15.5 Hz, H5), 5.45 (1H, dd, J = 15.5, 6.5 Hz, H4), 4.81 (1H, d, J = 8.0 Hz, NH), 4.25 (1H, dd, J = 11.5, 7.0 Hz, H1), 4.14 (1H, brm, H3), 4.11 (1H, dd, J = 11.5, 4.5 Hz, H1), 3.92 (1H, brm, H2), 2.42 (1H, brm, OH), 1.42 (9H, s, Boc), 1.39 (2H, m, H7), 1.27 (20H, m, H8-H17), 1.19 (9H, s, Piv), 0.86 (3H, t, J = 7.0 Hz, H18); ^{13}C NMR (125 MHz, CDCl_3) δ 178.6, 155.8, 134.4, 128.1, 79.7, 73.3, 63.0, 54.2, 38.8, 31.9,

29.6, 29.4, 29.3, 29.1, 29.0, 28.9, 28.3, 27.1, 22.6, 14.1; MS (ESI) m/z calcd for $C_{28}H_{51}D_2NNaO_5^+$ ($M+Na^+$) 508.4, found 508.3.

Synthesis of 18. Compound **18** was prepared from **17** in the same manner as the synthesis of **12**.

18: colorless oil; R_f 0.48 (silica gel, hexane/ethyl acetate = 4/1); 1H NMR (500 MHz, $CDCl_3$) δ 5.65 (1H, d, $J = 15.5$ Hz, H5), 5.40 (1H, dd, $J = 15.5, 6.5$ Hz, H4), 4.65 (1H, d, $J = 9.0$ Hz, NH), 4.25 (1H m, H1), 4.25 (1H, m, H2), 4.11 (1H, $J = 11.5, 4.0$ Hz, H1), 3.85 (1H, brm, 3H), 1.42 (9H, s, Boc), 1.34 (2H, m, H7), 1.26 (20H, m, H8-H17), 1.20 (9H, s, Piv), 0.89 (9H, s, *Si**t*-Bu), 0.88 (3H, t, $J = 7.0$ Hz, H18), 0.044 (3H, s, *Si*Me), 0.014 (3H, s, *Si*Me); ^{13}C NMR (125 MHz, $CDCl_3$) δ 178.6, 155.3, 133.4, 129.2, 79.2, 74.0, 62.6, 54.7, 38.7, 31.9, 29.6, 29.5, 29.3, 29.2, 29.0, 28.9, 28.4, 27.1, 25.8, 22.7, 18.1, 14.1, -4.26, -5.05.

Synthesis of 19. Compound **19** was prepared from **18** in the same manner as the synthesis of **13**.

19: colorless oil; R_f 0.27 (silica gel, hexane/ethyl acetate = 5/1); 1H NMR (500 MHz, $CDCl_3$) δ 5.42 (1H, d, $J = 6.0$ Hz, H4), 5.32 (1H, d, $J = 8.0$ Hz, NH), 4.45 (1H, brm, H3), 4.01 (1H, d, $J = 11.5$ Hz, H1), 3.54 (1H, ddd, $J = 3.0, 10, 10$ Hz, H1), 3.43 (1H, brm, OH), 2.97 (1H, d, $J = 9.5$ Hz, 2H), 2.00 (2H, t, $J = 7.0$ Hz, H6), 1.43 (9H, s, Boc), 1.33 (2H, tt, $J = 7.0, 7.0$ Hz, H7), 1.23 (20H, m, H8-H17), 0.88 (9H, s, *Si**t*-Bu), 0.85 (3H, t, $J = 7.0$ Hz, H18), 0.049 (3H, s, *Si*Me), 0.06 (3H, s, *Si*Me); ^{13}C NMR (125 MHz, $CDCl_3$) δ 155.7, 133.2, 129.3, 79.4, 76.3, 62.3, 55.1, 31.9, 29.6, 29.5, 29.3, 29.1, 28.9, 28.4, 25.8, 22.7, 18.0, 14.1, -4.55, -5.18; MS (ESI) m/z calcd for $C_{29}H_{57}D_2NNaO_4Si^+$ ($M+Na^+$) 538.4, found 538.2.

Synthesis of 20. Compound **20** was prepared from **19** in the same manner as the synthesis of **14**.

20: colorless oil; R_f 0.28 (silica gel, $CHCl_3/MeOH/H_2O = 65/25/4$); 1H NMR (500 MHz, CD_3OD) δ 5.66 (1H, d, $J = 15.5$ Hz, H5), 5.42 (1H, dd, $J = 15.5, 7.0$ Hz, H4), 4.25 (2H, br, α), 4.20 (1H, t, $J = 7.0$ Hz, H3), 4.00 (1H, m, H1), 3.93 (1H, m, H1), 3.63 (3H, brt, $J = 4.0$ Hz, β and H2), 3.21 (9H, s, N^+Me_3), 2.03 (2H,

dt, $J = 7.0$, 7.0 Hz, H6), 1.41 (9H, s, Boc), 1.28 (22H, m, H7-H17), 0.90 (9H, s, *Sit*-Bu), 0.89 (3H, t, $J = 7.0$ Hz, H18), 0.087 (3H, s, SiMe), -0.025 (3H, s, SiMe); ^{13}C NMR (125 MHz, CDCl_3) δ 185.3, 156.1, 152.1, 87.7, 81.4, 72.1, 69.6, 63.3, 63.2, 59.8, 56.1, 29.1, 26.2, 26.1, 25.9, 25.6, 25.5, 23.9, 20.9, 19.3, 17.5, 11.5, 5.89, -17.0, -17.9; MS (ESI) m/z calcd for $\text{C}_{34}\text{H}_{69}\text{D}_2\text{N}_2\text{NaO}_7\text{PSi}^+$ ($\text{M}+\text{Na}^+$) 703.5, found 703.2.

Synthesis of 6- d_2 -SSM. 6- d_2 -SSM was prepared from **20** in the same manner as the synthesis of 5- d -SSM.

6- d_2 -SSM: white solid; R_f 0.31 (silica gel, $\text{CHCl}_3/\text{MeOH}/\text{H}_2\text{O} = 65/45/8$); ^1H NMR (500 MHz, CD_3OD) δ 5.67 (1H, d, $J = 14.5$ Hz, H5), 5.42 (1H, dd, $J = 15.5$, 7.5 Hz, H4), 4.26 (2H, br, α), 4.10 (1H, m, H1), 4.03 (1H, dd, $J = 8.0$, 8.0 Hz, H3), 3.96 (2H, m, H1 and H2), 3.63 (3H, t, $J = 5.0$ Hz, β), 3.21 (9H, s, N^+Me_3), 2.17 (2H, dt, $J = 7.5$, 2.5 Hz, H2'), 1.57 (2H, m, H3'), 1.36 (2H, m, H7'), 1.28 (48H, m, H7-H17 and H4'-H17'), 0.90 (6H, t, $J = 7.0$ Hz, H18 and H18'); ^{13}C NMR (125 MHz, CDCl_3) δ 155.7, 133.0, 129.4, 79.4, 74.5, 63.4, 54.5, 31.9, 30.0, 29.5, 29.3, 29.1, 29.0, 28.4, 25.8, 22.7, 18.1, 14.1; ESI-HRMS m/z calcd for $\text{C}_{41}\text{H}_{81}\text{D}_2\text{N}_2\text{O}_6\text{PNa}(\text{M}+\text{Na})^+$ 755.6006, found 755.6012.

Synthesis of 21a. To a suspension of dried copper(I) iodide (461 mg, 2.42 mmol) in THF (23.6 mmol) was added 4-pentenoyl chloride (2.80 ml, 22.0 mmol). To the reaction mixture was slowly added decylmagnesium bromide (22.0 ml, 22.0 mmol) at 0 °C. After the reaction mixture was stirred for 2 h at 0 °C, the reaction was quenched with 1 M HCl, and the resulting mixture was extracted with diethyl ether. The organic layer was washed with saturated aqueous NaHCO_3 and brine, dried over Na_2SO_4 , and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **21a** (6.53 g, impure) as yellow oil. R_f 0.57 (silica gel, hexane/ethyl acetate = 5/1).

Synthesis of 22b. To a solution of 6-hepten-1-ol (2.36 g, 20.7 mmol) in DMSO (70 ml) was added IBX (10.4 g, 37.1 mmol). After the reaction mixture was stirred for 4 h at room temperature, the reaction was

quenched with water. The resulting mixture was filtered, extracted with diethyl ether, dried over Na₂SO₄, and filtered. To the filtrate was slowly added Grignard reagent (60.0 ml, 120 mmol) at 0 °C. After the reaction mixture was stirred for 18 h at room temperature, the reaction was quenched with 1 M HCl, and the resulting mixture was extracted with diethyl ether. The organic layer was dried over Na₂SO₄, filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **22b** (3.51 g, 15.5 mmol, 75% for 2 steps) as a colorless oil. R_f 0.43 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.79 (1H, ddt, *J* = 17.2, 10.4, 3.6 Hz, H2), 4.98 (1H, dd, *J* = 17.2, 1.6, 1.6 Hz, H1-cis), 4.92 (1H, ddt, *J* = 10.4, 2.0, 0.8 Hz, H1-trans), 3.56 (1H, m, H7), 2.02 (2H, q, *J* = 6.0 Hz, H3), 1.54-1.38 (6H, m, H4, H6, H8), 1.24 (14H, s, H5, H9-H14), 0.86 (3H, t, *J* = 7.2 Hz, H15).

Synthesis of 22c. To a solution of 8-nonen-1-ol (3.12 g, 21.9 mmol) in DMSO (70 ml) was added IBX (11.4g, 40.7 mmol). After the reaction mixture was stirred for 3 h at room temperature, the reaction was quenched with water. The resulting mixture was filtered, extracted with diethyl ether, dried over Na₂SO₄, and filtered. To the filtrate was slowly added Grignard reagent (70.0 ml, 140 mmol) at 0 °C. After the reaction mixture was stirred for 18 h at room temperature, the reaction was quenched with 1 M HCl, and the resulting mixture was extracted with diethyl ether. The organic layer was dried over Na₂SO₄, filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **22c** (3.40 g, 15.0 mmol, 69% for 2 steps) as a colorless oil. R_f 0.50 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.79 (1H, ddt, *J* = 17.2, 10.4, 3.6 Hz, H2), 4.97 (1H, dd, *J* = 17.2, 1.6, 1.6 Hz, H1-cis), 4.91 (1H, ddt, *J* = 10.4, 2.0, 0.8 Hz, H1-trans), 3.56 (1H, m, H9), 2.02 (2H, m, H3), 1.40 (6H, m, H4, H8, H10), 1.24 (14H, s, H5-H7, H11-H14), 0.86 (3H, t, *J* = 7.2 Hz, H15).

Synthesis of 22d. To a solution of 10-undecen-1-ol (3.52 g, 20.7 mmol) in DMSO (60 ml) was added IBX (9.55g, 34.1 mmol). After the reaction mixture was stirred for 4 h at room temperature, the reaction was quenched with water. The resulting mixture was filtered, extracted with diethyl ether, dried over Na₂SO₄, and filtered. To the filtrate was slowly added Grignard reagent (65.0 ml, 130 mmol) at 0 °C. After the reaction mixture was stirred for 18 h at room temperature, the reaction was quenched with 1 M HCl, and the resulting mixture was extracted with diethyl ether. The organic layer was dried over Na₂SO₄, filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **22d** (3.99 g, 17.2 mmol, 85% for 2 steps) as a colorless oil. R_f 0.41 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.79 (1H, ddt, *J* = 17.2, 10.0, 3.2 Hz, H2), 4.97 (1H, dd, *J* = 17.2, 1.6, 1.6 Hz, H1-cis), 4.91 (1H, ddt, *J* = 10.4, 2.0, 0.8 Hz, H1-trans), 3.56 (1H, m, H11), 2.01 (2H, m, H3), 1.40 (6H, m, H4, H10, H12), 1.24 (14H, s, H5-H9, H13, H14), 0.89 (3H, t, *J* = 7.2 Hz, H15).

Synthesis of 22e. To a solution of 12-tridecen-1-ol (6.26 g, 31.6 mmol) in DMSO (105 ml) was added IBX (17.7 g, 63.2 mmol). After the reaction mixture was stirred for 3 h at room temperature, the reaction was quenched with water. The resulting mixture was filtered, extracted with diethyl ether, dried over Na₂SO₄, and filtered. To the filtrate was slowly added Grignard reagent (60.0 ml, 180 mmol) at 0 °C. After the reaction mixture was stirred for 6 h at room temperature, the reaction was quenched with 1 M HCl, and the resulting mixture was extracted with diethyl ether. The organic layer was dried over Na₂SO₄, filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **22e** (5.36 g, 23.7 mmol, 75% for 2 steps) as a colorless oil. R_f 0.41 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.79 (1H, ddt, *J* = 17.2, 10.0, 3.2 Hz, H2), 4.97 (1H, dd, *J* = 17.2, 1.6, 1.6 Hz, H1-cis), 4.90 (1H, ddt, *J* = 10.4, 2.0, 0.8 Hz, H1-trans), 3.49 (1H, m, H13), 2.01 (2H, m, H3), 1.38 (6H, m, H4, H12, H14), 1.24 (14H, s, H5-H11), 0.92 (3H, t, *J* = 7.6 Hz, H15).

Synthesis of 21b-e. Compounds **21b-e** were synthesized from hydroxyl pentadecene **22b-e** in the following manner. To a solution of respective **22** (3.51 g, 15.5 mmol) in dichloromethane (10.9 ml) were added dimethyl sulfoxide (8.81 ml, 124 mmol), triethylamine (8.63 ml, 62.0 mmol), and SO₃-Py (7.40 g, 46.5 mmol) at 0 °C. After the reaction mixture was stirred for 14 h at room temperature, saturated aqueous Na₂S₂O₃ was added, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with brine, dried over Na₂SO₄, and evaporated to give the crude products. Purification by flash silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **21** as a colorless oil.

21b: colorless oil (83%) R_f 0.61 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.77 (1H, ddt, *J* = 17.2, 10.4, 3.6 Hz, H2), 4.97 (1H, dd, *J* = 17.2, 2.0, 1.6 Hz, H1-cis), 4.91 (1H, ddt, *J* = 10.4, 2.0, 1.2 Hz, H1-trans), 2.36 (4H, d, *J* = 6.0 Hz, H6, H8), 2.03 (2H, q, *J* = 6.0 Hz, H3), 1.54 (4H, m, H5, H9), 1.37 (2H, m, H4), 1.24 (10H, s, H10-H14), 0.86 (3H, t, *J* = 7.2 Hz, H15).

21c: colorless oil (85%) R_f 0.73 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.78 (1H, ddt, *J* = 17.2, 10.4, 3.6 Hz, H2), 4.97 (1H, dd, *J* = 17.2, 2.0, 1.6 Hz, H1-cis), 4.91 (1H, ddt, *J* = 10.4, 2.0, 1.2 Hz, H1-trans), 2.36 (4H, d, *J* = 6.0 Hz, H8, H10), 2.01 (2H, q, *J* = 6.0 Hz, H3), 1.54 (4H, m, H7, H11), 1.35 (2H, m, H4), 1.24 (10H, s, -CH₂-), 0.86 (3H, t, *J* = 7.2 Hz, H15).

21d: colorless oil (86%) R_f 0.58 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.78 (1H, ddt, *J* = 17.2, 10.4, 3.6 Hz, H2), 4.96 (1H, dd, *J* = 17.2, 2.0, 1.6 Hz, H1-cis), 4.90 (1H, ddt, *J* = 10.4, 2.0, 1.2 Hz, H1-trans), 2.36 (4H, t, *J* = 7.6 Hz, H10, H12), 2.01 (2H, q, *J* = 7.2 Hz, H3), 1.52 (4H, m, H9, H13), 1.33 (2H, m, H4), 1.24 (10H, s, -CH₂-), 0.88 (3H, t, *J* = 7.2 Hz, H15).

21e: colorless oil (72%) R_f 0.66 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.78 (1H, ddt, *J* = 17.2, 10.4, 3.6 Hz, H2), 4.97 (1H, dd, *J* = 17.2, 2.0, 1.6 Hz, H1-cis), 4.90 (1H, ddt, *J* = 10.4, 2.0, 1.2 Hz, H1-trans), 2.39 (4H, m, H12, H14), 2.01 (2H, q, *J* = 7.2 Hz, H3), 1.55 (2H, br, H11), 1.33 (2H, br, H4), 1.24 (10H, s, -CH₂-), 1.02 (3H, t, *J* = 7.2 Hz, H15).

Synthesis of 23a-e. Compounds **23a-e** were synthesized from oxopentadecene **21a-e** in the following manner. To a solution of respective oxopentadecene **21** (6.53 g, impure) in MeOH (22 ml) and CHCl₃ (11 ml) was added sodium borodeuteride (1.35 g, 33.0 mmol) at 0 °C. After the reaction mixture was stirred for 1 h at 0 °C, the reaction was quenched with CH₃COOH, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with saturated aqueous NaHCO₃ and brine, dried over Na₂SO₄, filtrated, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 20/1) afforded **23** as a colorless oil.

23a: colorless oil (70% for 2 steps) R_f 0.47 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.83 (1H, ddt, *J* = 17.2, 10.4, 3.6 Hz, H2), 5.04 (1H, d, *J* = 1.2 Hz, H1-cis), 4.96 (1H, d, *J* = 1.2 Hz, H1-trans), 2.02 (2H, m, H3), 1.54-1.38 (4H, m, H4, H6), 1.24 (16H, s, H7-H14), 0.86 (3H, t, *J* = 7.2 Hz, H15).

23b: colorless oil (96%) R_f 0.41 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.80 (1H, ddt, *J* = 17.2, 10.4, 3.6 Hz, H2), 4.98 (1H, dd, *J* = 17.2, 2.0, 1.6 Hz, H1-cis), 4.92 (1H, ddt, *J* = 10.4, 2.0, 1.2 Hz, H1-trans), 2.05 (2H, m, H3), 1.40 (6H, m, H4, H6, H8), 1.23 (14H, s, H5, H9-H14), 0.86 (3H, t, *J* = 7.2 Hz, H15).

23c: colorless oil (93%) R_f 0.41 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.79 (1H, ddt, *J* = 17.2, 10.4, 3.6 Hz, H2), 4.97 (1H, dd, *J* = 17.2, 2.0, 1.6 Hz, H1-cis), 4.91 (1H, ddt, *J* = 10.4, 2.0, 1.2 Hz, H1-trans), 2.02 (2H, m, H3), 1.40 (6H, m, H4, H8, H10), 1.23 (14H, s, -CH₂-), 0.87 (3H, t, *J* = 7.2 Hz, H15).

23d: colorless oil (93%) R_f 0.44 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.79 (1H, ddt, *J* = 17.2, 10.4, 3.6 Hz, H2), 4.96 (1H, dd, *J* = 17.2, 2.0, 1.6 Hz, H1-cis), 4.90 (1H, ddt, *J* = 10.4, 2.0, 1.2 Hz, H1-trans), 2.01 (2H, m, H3), 1.40 (6H, m, H4, H10, H12), 1.24 (14H, s, -CH₂-), 0.89 (3H, t, *J* = 7.2 Hz, H15).

23e: colorless oil (97%) R_f 0.37 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (400 MHz, CDCl_3) δ 5.79 (1H, ddt, $J = 17.2, 10.4, 3.6$ Hz, H2), 4.96 (1H, dd, $J = 17.2, 2.0, 1.6$ Hz, H1-cis), 4.90 (1H, ddt, $J = 10.4, 2.0, 1.2$ Hz, H1-trans), 2.01 (2H, m, H3), 1.40 (6H, m, H4, H12, H14), 1.24 (14H, s, $-\text{CH}_2-$), 0.92 (3H, t, $J = 7.2$ Hz, H15).

Synthesis of 24a-e. Compounds **24a-e** were synthesized from **23a-e**, respectively, in the following manner. To a solution of hydroxypentadecene **23** (3.11 g, 13.7 mmol) in dichloromethane (46.2 ml) were added triethylamine (44 ml, 24.7 mmol), *N,N*-dimethyl-4-aminopyridine (2.00 g, 16.4 mmol), and *p*-toluenesulfonyl chloride (3.93g, 20.6 mmol) at 0 °C. After being stirred for 5.5 h at room temperature, the reaction was quenched with saturated aqueous NaHCO_3 , and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with saturated aqueous NaHCO_3 and brine, dried over Na_2SO_4 , filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 30/1) afforded respective tosylate **24**.

24a: yellow oil (82%) R_f 0.51 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (400 MHz, CDCl_3) δ 7.77 (2H, d, $J = 8.4$ Hz, *o*-ArH), 7.30 (2H, d, $J = 8.0$ Hz, *m*-ArH), 5.67 (1H, ddt, $J = 16.8, 10.8, 4.4$ Hz, H2), 4.96 (1H, d, $J = 1.2$ Hz, H1-cis), 4.92 (1H, d, $J = 1.2$ Hz, H1-trans), 2.42 (3H, s, $-\text{CH}_3$), 2.02 (2H, m, H3), 1.62 (4H, m, H4, H6), 1.20 (16H, s, H7-H14), 0.88 (3H, t, $J = 7.2$ Hz, H15).

24b: yellow oil (80%) R_f 0.58 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (400 MHz, CDCl_3) δ 7.77 (2H, d, $J = 8.0$ Hz, *o*-ArH), 7.30 (2H, d, $J = 8.4$ Hz, *m*-ArH), 5.72 (1H, ddt, $J = 17.2, 10.4, 3.6$ Hz, H2), 4.96 (1H, dd, $J = 17.2, 2.0, 1.6$ Hz, H1-cis), 4.91 (1H, ddt, $J = 10.4, 2.0, 1.2$ Hz, H1-trans), 2.42 (3H, s, $-\text{CH}_3$), 1.94 (2H, m, H3), 1.54 (4H, m, H6, H8), 1.23 (16H, s, H4, H5, H9-H14), 0.86 (3H, t, $J = 7.2$ Hz, H15).

24c: yellow oil (81%) R_f 0.55 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (400 MHz, CDCl_3) δ 7.77 (2H, d, $J = 8.0$ Hz, *o*-ArH), 7.30 (2H, d, $J = 8.4$ Hz, *m*-ArH), 5.77 (1H, ddt, $J = 17.2, 10.4, 3.6$ Hz, H2),

4.97 (1H, dd, $J = 17.2, 2.0, 1.6$ Hz, H1-cis), 4.91 (1H, ddt, $J = 10.4, 2.0, 1.2$ Hz, H1-trans), 2.42 (3H, s, -CH₃), 2.00 (2H, m, H3), 1.54 (4H, m, H8, H10), 1.23 (16H, s, -CH₂-), 0.83 (3H, t, $J = 7.2$ Hz, H15).

24d: yellow oil (37%) R_f 0.52 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 7.77 (2H, d, $J = 8.0$ Hz, *o*-ArH), 7.30 (2H, d, $J = 8.4$ Hz, *m*-ArH), 5.79 (1H, ddt, $J = 17.2, 10.4, 3.6$ Hz, H2), 4.97 (1H, dd, $J = 17.2, 2.0, 1.6$ Hz, H1-cis), 4.91 (1H, ddt, $J = 10.4, 2.0, 1.2$ Hz, H1-trans), 2.42 (3H, s, -CH₃), 2.00 (2H, m, H3), 1.54 (4H, m, H10, H12), 1.15 (16H, s, -CH₂-), 0.80 (3H, t, $J = 7.2$ Hz, H15).

24e: yellow oil (78%) R_f 0.54 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 7.77 (2H, d, $J = 8.0$ Hz, *o*-ArH), 7.29 (2H, d, $J = 8.4$ Hz, *m*-ArH), 5.79 (1H, ddt, $J = 17.2, 10.4, 3.6$ Hz, H2), 4.97 (1H, dd, $J = 17.2, 2.0, 1.6$ Hz, H1-cis), 4.91 (1H, ddt, $J = 10.4, 2.0, 1.2$ Hz, H1-trans), 2.42 (3H, s, -CH₃), 2.00 (2H, m, H3), 1.54 (4H, m, H12, H14), 1.15 (16H, s, -CH₂-), 0.80 (3H, t, $J = 7.2$ Hz, H15).

Synthesis of 25a-e. Compounds **25a-e** were synthesized from **24a-e**, respectively, in the following manner.

To a solution of tosylate **24** (2.07 mg, 5.41 mmol) in diethyl ether (20 ml) was added lithium aluminium deuteride (853 mg, 20.3 mmol). After the reaction mixture was stirred for 4 h at room temperature, the reaction was quenched with ethanol. The resulting mixture was filtered through Celite and washed with ethyl acetate, and the filtrate was evaporated to give the crude products. Purification by silica gel column chromatography (hexane) afforded **25** as a white solid.

25a: colorless oil (88%) R_f 0.87 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.80 (1H, ddt, $J = 17.2, 10.0, 6.6$ Hz, H2), 4.97 (1H, ddt, $J = 17.6, 3.6, 1.6$ Hz, H1-cis), 4.91 (1H, dt, $J = 10.0, 2.0, 1.2$ Hz, H1-trans), 2.02 (2H, br q, $J = 6.7$ Hz, H3), 1.35 (2H, m, H4), 1.21 (18H, s, H6-H14), 0.86 (3H, t, $J = 7.2$ Hz, H15).

25b: colorless oil (86%) R_f 0.71 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (400 MHz, CDCl₃) δ 5.79 (1H, ddt, $J = 17.2, 10.4, 3.6$ Hz, H2), 4.97 (1H, dd, $J = 17.2, 2.0, 1.6$ Hz, H1-cis), 4.91 (1H, ddt, $J = 10.4, 2.0, 1.2$ Hz, H1-trans), 2.02 (2H, m, H3), 1.23 (20H, s, -CH₂-), 0.86 (3H, t, $J = 7.2$ Hz, H15).

25c: colorless oil (81%) R_f 0.79 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (400 MHz, CDCl_3) δ 5.79 (1H, ddt, $J = 17.2, 10.4, 3.6$ Hz, H2), 4.97 (1H, dd, $J = 17.2, 2.0, 1.6$ Hz, H1-cis), 4.91 (1H, ddt, $J = 10.4, 2.0, 1.2$ Hz, H1-trans), 2.03 (2H, m, H3), 1.23 (20H, s, $-\text{CH}_2-$), 0.86 (3H, t, $J = 7.2$ Hz, H15).

25d: colorless oil (57%) R_f 0.79 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (400 MHz, CDCl_3) δ 5.79 (1H, ddt, $J = 17.2, 10.4, 3.6$ Hz, H2), 4.97 (1H, dd, $J = 17.2, 2.0, 1.6$ Hz, H1-cis), 4.90 (1H, ddt, $J = 10.4, 2.0, 1.2$ Hz, H1-trans), 2.01 (2H, m, H3), 1.23 (20H, s, $-\text{CH}_2-$), 0.86 (3H, t, $J = 7.2$ Hz, H15).

25e: colorless oil (81%) R_f 0.74 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (400 MHz, CDCl_3) δ 5.79 (1H, ddt, $J = 17.2, 10.4, 3.6$ Hz, H2), 4.97 (1H, dd, $J = 17.2, 2.0, 1.6$ Hz, H1-cis), 4.90 (1H, ddt, $J = 10.4, 2.0, 1.2$ Hz, H1-trans), 2.02 (2H, m, H3), 1.24 (20H, s, $-\text{CH}_2-$), 0.86 (3H, t, $J = 7.2$ Hz, H15).

Synthesis of 26. To a solution of 14-pentadecenoic acid methyl ester (1.02 g, 4.01 mmol) in THF (30 ml) was added lithium aluminium deuteride (201 mg, 4.81 mmol). After the reaction mixture was stirred for 30 min at room temperature, the reaction was quenched with water. The resulting mixture was extracted with ethyl acetate and washed with brine, and the filtrate was evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 5/1) afforded **26** (798 mg, 3.50 mmol, 87%) as a colorless oil; R_f 0.32 (silica gel, hexane/ethyl acetate = 3/1); $^1\text{H NMR}$ (400 MHz, CDCl_3) δ 5.79 (1H, ddt, $J = 17.2, 10.0, 3.2$ Hz, H1), 4.97 (1H, ddt, $J = 17.2, 1.6, 1.6$ Hz, H2-cis), 4.91 (1H, ddt, $J = 10.4, 2.0, 0.8$ Hz, H2-trans), 2.01 (2H, m, H3), 1.55 (2H, t, $J = 6.0$ Hz, H14), 1.24 (20H, s, H4-H13).

Synthesis of 27. Compound **27** was prepared from **26** (798 mg, 3.50 mmol) in the same way as the synthesis of **24**.

27: (1.28 g, 3.35 mmol, 95%) yellow oil; R_f 0.58 (silica gel, hexane/ethyl acetate = 3/1); $^1\text{H NMR}$ (400 MHz, CDCl_3) δ 7.77 (2H, d, $J = 8.0$ Hz, *o*-ArH), 7.29 (2H, d, $J = 8.4$ Hz, *m*-ArH), 5.79 (1H, ddt, $J = 17.2, 10.4, 3.6$ Hz, H2), 4.97 (1H, dd, $J = 17.2, 2.0, 1.6$ Hz, H1-cis), 4.91 (1H, ddt, $J = 10.4, 2.0, 1.2$ Hz,

H1-trans), 2.43 (3H, s, -CH₃), 2.00 (2H, m, H3), 1.60 (2H, t, *J* = 6.0 Hz, H14), 1.22 (22H, s, -CH₂-).

Synthesis of 25f. Compound **25f** was prepared from **27** (1.28 g, 3.35 mmol) in the same way as the synthesis of other **25**.

25f: (563 mg, 2.64 mmol, 79%) colorless oil; *R*_f 0.81 (silica gel, hexane/ethyl acetate = 3/1); ¹H NMR (400 MHz, CDCl₃) δ 5.79 (1H, ddt, *J* = 17.2, 10.4, 3.6 Hz, H2), 4.97 (1H, dd, *J* = 17.2, 2.0, 1.6 Hz, H1-cis), 4.90 (1H, ddt, *J* = 10.4, 2.0, 1.2 Hz, H1-trans), 2.02 (2H, m, H3), 1.24 (22H, s, -CH₂-).

Synthesis of 29. A known bromide **28** was synthesized as previously reported (*Org. Lett.* **2006**, *8*, 5569–5572). To a solution of bromide **28** (309 mg, 0.74 mmol) in CH₂Cl₂ (7.46 ml) was added trifluoroacetic acid (1.64 ml, 22.2 mmol) at 0 °C. After the reaction mixture was stirred for 90 min at 0 °C, DMAP (90 mg, 0.38 mmol), *p*-nitrophenyl octadecanoate (360 mg, 0.89 mmol) and DIPEA (4.25 ml, 24.2 mmol) were added. After the reaction mixture was stirred for 2 h at room temperature, the reaction was quenched with saturated NaHCO₃, and the resulting mixture was extracted with CHCl₃. The organic layer was washed with 1M NaOH, saturated KHSO₄, and brine, dried over Na₂SO₄, filtered and evaporated to give the crude products. Purification by silica gel column chromatography (CHCl₃/MeOH = 150/1) afforded amide **29** (330 mg, 0.56 mmol, 77%) as a white solid; *R*_f 0.47 (silica gel, CHCl₃/MeOH = 10/1); ¹H NMR (400 MHz, CDCl₃) δ 6.20 (1H, d, *J* = 6.8 Hz, N-H), 5.90 (1H, ddd, *J* = 17.2, 10.8, 6.0 Hz, H4), 5.37 (1H, d, *J* = 16.8 Hz, H5trans), 5.22 (1H, d, *J* = 10.4 Hz, H5cis), 4.32 (3H, m, H3, α), 4.22 (1H, br, H1), 4.14 (2H, m, H1,H2), 3.79 (3H, dd, *J* = 11.2, 0.8 Hz, -OMe), 3.53 (2H, t, *J* = 6.4 Hz, β), 2.20 (2H, t, *J* = 6.0 Hz, H2'), 1.60 (2H, brm, H3'), 1.23 (28H, s, -CH₂-), 0.86 (3H, t, *J* = 6.8 Hz, H18').

Synthesis of 30a-f. To a solution of **29** (138 mg, 0.24 mmol) in CH₂Cl₂ (4.8 ml) was added deuterated 1-pentadecene **25a** (252 mg, 1.19 mmol) and Grubbs catalyst 2nd generation (9.17 mg, 0.01 mmol) at

room temperature. After reaction mixture was refluxed for 2h at 55 °C, the resulting mixture was evaporated to give crude products. Purification by silica gel column chromatography (CHCl₃/MeOH = 150/1) afforded **30a** as white solids. Compounds **30b-f** were prepared from **25b-f** in a similar manner.

30a: white solid (100 mg, 0.13 mmol, 57%); R_f 0.51 (silica gel, CHCl₃/MeOH = 10/1); ¹H NMR (400 MHz, CDCl₃) δ 6.10 (1H, d, *J* = 7.2 Hz, N-H), 5.73 (1H, dt, *J* = 15.6, 6.4 Hz, H5), 5.46 (1H, dd, *J* = 15.2, 6.8 Hz, H4), 4.32 (3H, m, H3, α), 4.15 (1H, br, H1, H2), 3.79 (3H, dd, *J* = 11.2, 2.8 Hz, -OMe), 3.53 (2H, t, *J* = 6.4 Hz, β), 2.18 (2H, t, *J* = 8.0 Hz, H2'), 2.00 (2H, dt, *J* = 6.8, 7.2 Hz, H6), 1.60 (2H, brm, H3'), 1,23 (48H, s, -CH₂-), 0.86 (6H, t, *J* = 6.8 Hz, H18, 18').

30b: white solid (136 mg, 0.18 mmol, 49%); R_f 0.51 (silica gel, CHCl₃/MeOH = 10/1); ¹H NMR (400 MHz, CDCl₃) δ 6.10 (1H, d, *J* = 7.2 Hz, N-H), 5.73 (1H, dt, *J* = 15.6, 6.4 Hz, H5), 5.46 (1H, dd, *J* = 15.2, 6.8 Hz, H4), 4.32 (3H, m, H3, α), 4.15 (1H, br, H1, H2), 3.79 (3H, dd, *J* = 11.2, 2.8 Hz, -OMe), 3.53 (2H, t, *J* = 6.4 Hz, β), 2.18 (2H, t, *J* = 8.0 Hz, H2'), 2.00 (2H, dt, *J* = 6.8, 7.2 Hz, H6), 1.60 (2H, brm, H3'), 1,23 (48H, s, -CH₂-), 0.86 (6H, t, *J* = 6.8 Hz, H18, 18').

30c: white solid (100 mg, 0.13 mmol, 47%); R_f 0.51 (silica gel, CHCl₃/MeOH = 10/1); ¹H NMR (400 MHz, CDCl₃) δ 6.11 (1H, d, *J* = 7.2 Hz, N-H), 5.73 (1H, dt, *J* = 15.6, 6.4 Hz, H5), 5.46 (1H, dd, *J* = 15.2, 6.8 Hz, H4), 4.32 (3H, m, H3, α), 4.14 (1H, br, H1, H2), 3.80 (3H, dd, *J* = 11.2, 2.8 Hz, -OMe), 3.53 (2H-OMe), 3.53 (2H, t, *J* = 6.4 Hz, β), 2.18 (2H, t, *J* = 8.0 Hz, H2'), 2.01 (2H, t, *J* = 6.4 Hz, β), 2.17 (2H, t, *J* = 8.0 Hz, H2'), 2.00 (2H, dt, *J* = 6.8, 7.2 Hz, H6), 1.58 (2H, brm, H3'), 1,23 (48H, s, -CH₂-), 0.86 (6H, t, *J* = 6.8 Hz, H18, 18').

30d: white solid (160 mg, 0.21 mmol, 65%); R_f 0.51 (silica gel, CHCl₃/MeOH = 10/1); ¹H NMR (400 MHz, CDCl₃) δ 6.08 (1H, d, *J* = 7.2 Hz, N-H), 5.73 (1H, dt, *J* = 15.6, 6.4 Hz, H5), 5.46 (1H, dd, *J* = 15.2, 6.8 Hz, H4), 4.32 (3H, m, H3, α), 4.15 (1H, br, H1, H2), 3.80 (3H, dd, *J* = 11.2, 2.8 Hz, = 6.8, 7.2 Hz, H6), 1.59 (2H, brm, H3'), 1,23 (48H, s, -CH₂-), 0.86 (6H, t, *J* = 6.8 Hz, H18, 18').

30e: white solid (138 mg, 0.18 mmol, 60%); R_f 0.51 (silica gel, CHCl₃/MeOH = 10/1); ¹H NMR (400 MHz,

CDCl₃) δ 6.10 (1H, d, *J* = 7.2 Hz, N-H), 5.73 (1H, dt, *J* = 15.6, 6.4 Hz, H5), 5.46 (1H, dd, *J* = 15.2, 6.8 Hz, H4), 4.32 (3H, m, H3, α), 4.15 (1H, br, H1, H2), 3.79 (3H, dd, *J* = 11.2, 2.8 Hz, -OMe), 3.53 (2H, t, *J* = 6.4 Hz, β), 2.18 (2H, t, *J* = 8.0 Hz, H2'), 2.00 (2H, dt, *J* = 6.8, 7.2 Hz, H6), 1.60 (2H, brm, H3'), 1,23 (48H, s, -CH₂-), 0.86 (6H, t, *J* = 6.8 Hz, H18, 18').

30f: white solid (191 mg, 0.25 mmol, 63%); R_f 0.51 (silica gel, CHCl₃/MeOH = 10/1); ¹H NMR (400 MHz, CDCl₃) δ 6.10 (1H, d, *J* = 7.2 Hz, N-H), 5.73 (1H, dt, *J* = 15.6, 6.4 Hz, H5), 5.46 (1H, dd, *J* = 15.2, 6.8 Hz, H4), 4.32 (3H, m, H3, α), 4.15 (1H, br, H1, H2), 3.79 (3H, dd, *J* = 11.2, 2.8 Hz, -OMe), 3.53 (2H, t, *J* = 6.4 Hz, β), 2.18 (2H, t, *J* = 8.0 Hz, H2'), 2.00 (2H, dt, *J* = 6.8, 7.2 Hz, H6), 1.60 (2H, brm, H3'), 1,23 (50H, s, -CH₂-), 0.86 (3H, t, *J* = 6.8 Hz, H18').

Synthesis of 8-*d*₂-, 10-*d*₂-, 12-*d*₂-, 14-*d*₂-, 16-*d*₂-, and 18-*d*₃- SSMs. To a solution of **30a** (100 mg, 0.13 mmol) in methanol (2.5 ml) was added 30% trimethylamine solution (1.4 ml) at room temperature. After the reaction mixture was stirred for 14 h at same temperature, solvent was evaporated to give the crude products. Purification by silica gel column chromatography (CHCl₃/MeOH = 3/1 to CHCl₃/MeOH/H₂O = 65/25/4) afforded 8-*d*₂-SSMs as a white solid. 10-*d*₂-, 12-*d*₂-, 14-*d*₂-, 16-*d*₂-, and 18-*d*₃- SSMs were prepared from **30b-f** in a similar manner.

8-*d*₂-SSM: white solid (44.2 mg, 0.04 mmol, 46%), R_f 0.24 (silica gel, CHCl₃/MeOH/H₂O = 65/25/4); ¹H NMR (400 MHz, CD₃OD) δ 5.69 (1H, td, *J* = 15.6, 6.8 Hz, H5), 5.43 (1H, dd, *J* = 15.2, 7.6 Hz, H4), 4.26 (2H, br, α), 4.10 (1H, m, H1), 4.02 (1H, dd, *J* = 8.0 Hz, H3), 3.95 (2H, m, H1, H2), 3.63 (2H, t, *J* = 5.2 Hz, β), 3.21 (9H, s, -N⁺Me₃), 2.17 (2H, br, H2'), 2.01 (2H, br, H6), 1.57 (2H, m, H3'), 1.28 (48H, s, -CH₂-), 0.89 (6H, t, *J* = 7.2 Hz, H18, H18'); ESI-HRMS *m/z* calcd for C₄₁H₈₁D₂N₂O₆PNa(M+Na)⁺ 755.6006, found 755.6018.

10-*d*₂-SSM: white solid (56.1 mg, 0.08 mmol, 43%). R_f 0.23 (silica gel, CHCl₃/MeOH/H₂O = 65/25/4); ¹H NMR (400 MHz, CD₃OD) δ 5.68 (1H, td, *J* = 15.5, 6.5 Hz, H5), 5.42 (1H, dd, *J* = 15.5, 7.5 Hz, H4), 4.25

(2H, br, α), 4.08 (1H, m, H1), 4.02 (1H, dd, $J = 8.7, 7.3$ Hz, H3), 3.94 (2H, m, H1, H2), 3.62 (2H, t, $J = 4.5$ Hz, β), 3.20 (9H, s, $-\text{N}^+\text{Me}_3$), 2.15 (2H, br, H2'), 2.00 (2H, br, H6), 1.56 (2H, m, H3'), 1.26 (48H, s, $-\text{CH}_2-$), 0.88 (6H, t, $J = 7.2$ Hz, H18, H18'); ESI-HRMS m/z calcd for $\text{C}_{41}\text{H}_{81}\text{D}_2\text{N}_2\text{O}_6\text{PNa}(\text{M}+\text{Na})^+$ 755.6006, found 755.6018.

12- d_2 -SSM: white solid (47.8 mg, 0.07 mmol, 50%). R_f 0.23 (silica gel, $\text{CHCl}_3/\text{MeOH}/\text{H}_2\text{O} = 65/25/4$); ^1H NMR (400 MHz, CD_3OD) δ 5.69 (1H, td, $J = 15.5, 6.0$ Hz, H5), 5.43 (1H, dd, $J = 14.3, 7.7$ Hz, H4), 4.27 (2H, br, α), 4.08 (1H, m, H1), 4.02 (1H, dd, $J = 8.7, 7.3$ Hz, H3), 3.95 (2H, m, H1, H2), 3.62 (2H, br, β), 3.21 (9H, s, $-\text{N}^+\text{Me}_3$), 2.19 (2H, br, H2'), 2.02 (2H, br, H6), 1.57 (2H, m, H3'), 1.28 (48H, s, $-\text{CH}_2-$), 0.89 (6H, t, $J = 7.0$ Hz, H18, H18'); ESI-HRMS m/z calcd for $\text{C}_{41}\text{H}_{81}\text{D}_2\text{N}_2\text{O}_6\text{PNa}(\text{M}+\text{Na})^+$ 755.6006, found 755.6016.

14- d_2 -SSM: white solid (75.5 mg, 0.10 mmol, 34%). R_f 0.23 ($\text{CHCl}_3/\text{MeOH}/\text{H}_2\text{O} = 65/25/4$); ^1H NMR (400 MHz, CD_3OD) δ 5.69 (1H, td, $J = 15.0, 7.0$ Hz, H5), 5.43 (1H, dd, $J = 14.5, 8.0$ Hz, H4), 4.26 (2H, br, α), 4.10 (1H, m, H1), 4.04 (1H, dd, $J = 8.7, 7.3$ Hz, H3), 3.95 (2H, m, H1, H2), 3.63 (2H, t, $J = 4.0$ Hz, β), 3.21 (9H, s, $-\text{N}^+\text{Me}_3$), 2.17 (2H, br, H2'), 2.01 (2H, br, H6), 1.57 (2H, m, H3'), 1.28 (48H, s, $-\text{CH}_2-$), 0.89 (6H, t, $J = 7.2$ Hz, H18, H18'); ESI-HRMS m/z calcd for $\text{C}_{41}\text{H}_{81}\text{D}_2\text{N}_2\text{O}_6\text{PNa}(\text{M}+\text{Na})^+$ 755.6006, found 755.6016.

16- d_2 -SSM: white solid (28.4 mg, 0.04 mmol, 26%). R_f 0.24 (silica gel, $\text{CHCl}_3/\text{MeOH}/\text{H}_2\text{O} = 65/25/4$); ^1H NMR (500 MHz, CD_3OD) δ 5.69 (1H, td, $J = 15.5, 6.5$ Hz, H5), 5.43 (1H, dd, $J = 15.0, 7.5$ Hz, H4), 4.27 (2H, br, α), 4.09 (1H, m, H1), 4.03 (1H, dd, $J = 8.7, 7.3$ Hz, H3), 3.95 (2H, m, H1, H2), 3.64 (2H, t, $J = 4.0$ Hz, β), 3.20 (9H, s, $-\text{N}^+\text{Me}_3$), 2.17 (2H, br, H2'), 2.03 (2H, br, H6), 1.56 (2H, m, H3'), 1.28 (48H, s, $-\text{CH}_2-$), 0.89 (6H, t, $J = 7.2$ Hz, H18, H18'); ESI-HRMS m/z calcd for $\text{C}_{41}\text{H}_{81}\text{D}_2\text{N}_2\text{O}_6\text{PNa}(\text{M}+\text{Na})^+$ 755.6006, found 755.6017.

18- d_3 -SSM: white solid (56.1 mg, 0.08 mmol, 31%). R_f 0.24 (silica gel, $\text{CHCl}_3/\text{MeOH}/\text{H}_2\text{O} = 65/25/4$); ^1H NMR (400 MHz, CD_3OD) δ 5.69 (1H, td, $J = 15.5, 6.5$ Hz, H5), 5.43 (1H, dd, $J = 15.0, 7.5$ Hz, H4), 4.27

(2H, br, α), 4.09 (1H, m, H1), 4.03 (1H, dd, $J = 8.7, 7.3$ Hz, H3), 3.95 (2H, m, H1, H2), 3.64 (2H, t, $J = 4.0$ Hz, β), 3.20 (9H, s, $-N^+Me_3$), 2.17 (2H, br, H2'), 2.03 (2H, br, H6), 1.56 (2H, m, H3'), 1.28 (48H, s, $-CH_2-$), 0.89 (6H, t, $J = 7.2$ Hz, H18'); ESI-HRMS m/z calcd for $C_{41}H_{80}D_3N_2O_6PNa(M+Na)^+$ 756.6069, found 756.6077.

Synthesis of 31. To a solution of methyl stearate (710 mg, 2.38 mmol) in MeOD (8 ml) was added sodium methoxide (193 mg, 3.57 mmol). After the reaction mixture was refluxed for 24 h at 80 °C, deuterium chloride, 35wt.% solution in D_2O was added. The resulting mixture was extracted with dichloromethane, dried over $MgSO_4$, filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **31** (421 mg, 1.40 mmol, 59%) as a white solid. R_f 0.73 (silica gel, hexane/ethyl acetate = 9/1); 1H NMR (500 MHz, $CDCl_3$) δ 3.64 (3H, s, $-CO_2Me$), 2.27 (3%*, br, H2), 1.58 (2H, q, $J = 7.0$ Hz, H3), 1.23 (28H, s, H4-H17), 0.86 (3H, t, $J = 6.5$ Hz, H18); *not deuterated H (%)

Synthesis of 32a. To a solution of **31** (337 mg, 1.12 mmol) in MeOH (1.9 ml) and THF (3.7 ml) was added 1 M NaOH solution (3.4 ml, 3.36 mmol). After the reaction mixture was stirred for 2 h at 60 °C, 1 M HCl was added, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with brine, dried over $MgSO_4$, filtered, and evaporated to give **32a** (302 mg, 1.06 mmol, 94%) as a white solid. R_f 0.09 (silica gel, hexane/ethyl acetate = 10/1); 1H NMR (500 MHz, $CDCl_3$) δ 2.33 (35%*, br, H2), 1.58 (2H, q, $J = 7.0$ Hz, H3), 1.26 (28H, s, H4-H17), 0.86 (3H, t, $J = 6.5$ Hz, H18). *not deuterated H (%).

Synthesis of 33. Compound **33** was prepared from heptadecanoic acid methyl ester in a similar manner to the preparation of **31** as white solid (195 mg, 0.68 mmol, 88%). R_f 0.46 (silica gel, hexane/ethyl acetate = 10/1); 1H NMR (500 MHz, $CDCl_3$) δ 3.64 (3H, s, $-CO_2Me$), 2.26 (15%*, br, H2), 1.58 (2H, q, $J = 6.9$ Hz,

H3), 1.26 (26H, s, H4-H16), 0.85 (3H, t, $J = 7.0$ Hz, H17). *not deuterated H (%).

Synthesis of 34. To a solution of **33** (195 mg, 0.68 mmol) in THF (3.5 ml) was added lithium aluminum hydride (49.6 mg, 1.31 mmol). After the mixture was stirred for 7 h at room temperature, the reaction was quenched with water. The resulting mixture was filtered through Celite and washed with ethyl acetate, and the filtrate was evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **34** (163 mg, 0.63 mmol, 93%) as a white solid. R_f 0.11 (silica gel, hexane/ethyl acetate = 10/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 3.61 (2H, s, H1), 1.29 (1H, br, -OH), 1.26 (28H, s, H3-H16), 0.85 (3H, t, $J = 7.0$ Hz, H17).

Synthesis of 35. To a solution of **34** (160 mg, 0.62 mmol) in dichloromethane (5.0 ml) were added triethylamine (258 μl , 1.86 mmol), *N,N*-dimethyl-4-aminopyridine (7.6 mg, 0.06 mmol), and *p*-toluenesulfonyl chloride (297 mg, 1.56 mmol) at 0 °C. After being stirred for 9 h at room temperature, the reaction was quenched with water, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with saturated aqueous NaHCO_3 and brine, dried over MgSO_4 , filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **35** (292 mg, 0.62 mmol, quant) as a white solid. R_f 0.36 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 7.76 (2H, d, $J = 8.3$ Hz, *o*-ArH), 7.30 (2H, d, $J = 7.9$ Hz, *m*-ArH), 3.82 (2H, s, H1), 2.42 (3H, s, - CH_3), 1.23 (28H, s, H3-H16), 0.86 (3H, t, $J = 7.2$ Hz, H17).

Synthesis of 36. To a solution of **35** (292 mg, 0.62 mmol) in *N,N*-dimethylformamide (3.0 ml) were added 18-crown-6-ether (323 mg, 0.93 mmol) and KCN (62.7 mg, 0.93 mmol) at 0 °C. After being stirred for 24 h at room temperature, the reaction was quenched with water, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with saturated aqueous NaHCO_3 and brine, dried over MgSO_4 ,

filtered, and evaporated to give the crude products. Purification by flash silica gel column chromatography (hexane/ethyl acetate = 100/1) afforded **36** (98.3 mg, 0.37 mmol, 59%) as a white solid. R_f 0.94 (silica gel, hexane/ethyl acetate = 50/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 2.25 (2H, s, H1), 1.23 (28H, s, H4-H17), 0.85 (3H, t, $J = 7.2$ Hz, H18).

Synthesis of 32b. To a solution of **36** (98.3 mg, 0.37 mmol) in EtOH (3.0 ml) was added 12.5 M NaOH solution (1.00 ml, 12.5 mmol). After being stirred for 16 h at 100 °C, the reaction was quenched with 1 M HCl, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with brine, dried over MgSO_4 , filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **32b** (96.3 mg, 0.34 mmol, 91%) as a white solid. R_f 0.09 (hexane/ethyl acetate = 10/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 2.34 (2H, t, $J = 7.6$ Hz, H2), 1.23 (28H, s, H4-H17), 0.86 (3H, t, $J = 7.2$ Hz, H18).

Synthesis of 37. 2,2- d_2 -palmitic acid methyl ester was prepared from palmitic acid methyl ester in a similar manner to the preparation of **31** as a white solid (689 mg, 2.53 mmol, 57%). R_f 0.53 (hexane/ethyl acetate = 10/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 2.26 (25%*, br, H2), 1.58 (2H, q, $J = 6.9$ Hz, H3), 1.26 (24H, s, H4-H15), 0.85 (3H, t, $J = 7.0$ Hz, H16). *not deuterated H (%).

From 2,2- d_2 -palmitic acid methyl ester, **37** was prepared in a similar manner to the preparation of **34** as a white solid (557 mg, 2.28 mmol, 91%). R_f 0.10 (silica gel, hexane/ethyl acetate = 10/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 3.61 (2H, s, H1), 1.29 (1H, br, $-\text{OH}$), 1.26 (26H, s, H3-H15), 0.85 (3H, t, $J = 7.0$ Hz, H16).

Synthesis of 38. To a solution of **37** (557 mg, 2.28 mmol) in dichloromethane (10.9 ml) were added dimethyl sulfoxide (1.30 ml, 18.2 mmol), triethylamine (1.27 ml, 9.12 mmol), and $\text{SO}_3\text{-Py}$ (1.11g, 6.84 mmol) at 0 °C. After being stirred for 2 h at room temperature, the reaction was quenched with saturated

aqueous $\text{Na}_2\text{S}_2\text{O}_3$, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with water and saturated aqueous NH_4Cl and brine, dried over MgSO_4 , filtered, and evaporated to give the crude products. Purification by flash silica gel column chromatography (hexane/ethyl acetate = 50/1) afforded **38** (548 mg, 2.24 mmol, 98%) as a white solid. R_f 0.28 (silica gel, hexane/ethyl acetate = 20/1); ^1H NMR (500 MHz, CDCl_3) δ 9.74 (1H, -CHO), 1.58 (2H, t, $J = 7.2$ Hz, H3), 1.23 (24H, s, H4-H15), 0.85 (3H, t, $J = 7.2$ Hz, H16).

Synthesis of 39. To a solution of **38** (548 mg, 2.24 mmol) in THF (33.7 ml) were added phosphonium ylides (2.35 g, 6.72 mmol). After the reaction mixture was stirred for 2 h at room temperature and then for 16 h at 40 °C, the solvent was removed by evaporation to give the crude products. Purification by silica gel column chromatography (hexane) afforded **39** (537 mg, 1.72 mmol, 77%) as a colorless oil. R_f 0.57 (silica gel, hexane/ethyl acetate = 10/1); ^1H NMR (500 MHz, CDCl_3) δ 6.94 (1H, dd, $J = 15.6, 5.6$ Hz, H2), 5.79 (1H, d, $J = 15.6$ Hz, H3), 4.15 (2H, q, $J = 7.2$ Hz, $-\text{OCH}_2-$), 1.43 (2H, m, H5), 1.27 (3H, t, $J = 7.1$ Hz, $-\text{OCH}_2\text{CH}_3$), 1.23 (24H, s, H6-H17), 0.86 (3H, t, $J = 7.2$ Hz, H18).

Synthesis of 40. To a solution of **39** (537 mg, 1.72 mmol) in ethyl acetate was added Pd-C (181 mg, 0.17 mmol). After being stirred for 24 h at room temperature under hydrogen atmosphere, the mixture was filtered through Celite and washed with ethyl acetate, and the filtrate was evaporated to give the crude products. Purification by flash silica gel column chromatography (hexane/ethyl acetate = 50/1) afforded **40** (500 mg, 1.59 mmol, 94%) as a white solid. R_f 0.46 (silica gel, hexane/ethyl acetate = 10/1); ^1H NMR (500 MHz, CDCl_3) δ 4.10 (2H, q, $J = 7.2$ Hz, $-\text{OCH}_2-$), 2.26 (2H, t, $J = 7.6$ Hz, H2), 1.57 (2H, m, H3), 1.28-1.21 (29H, m, H5-H17, $-\text{OCH}_2\text{CH}_3-$), 0.86 (3H, t, $J = 7.2$ Hz, H18).

Synthesis of 32c. To a solution of **40** (500 mg, 1.59 mmol) in MeOH (2.70 ml) and THF (6.30 ml) was

added 1 M NaOH solution (3.18 ml, 3.18 mmol). After being stirred for 5 h at 60 °C, the reaction was quenched with 1 M HCl, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with brine, dried over MgSO₄, filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **32c** (472 mg, 1.59 mmol, quant) as a white solid. R_f 0.07 (silica gel, hexane/ethyl acetate = 10/1); ¹H NMR (500 MHz, CDCl₃) δ 2.34 (2H, t, *J* = 7.6 Hz, H2), 1.59 (2H, m, H3), 1.23 (26H, s, H5-H17), 0.86 (3H, t, *J* = 7.2 Hz, H18).

Synthesis of 41a. To monoethyl adipate (656 μl, 4.00 mmol) was added thionyl chloride (0.87 ml, 12.0 mmol). After being refluxed for 16 h at 80 °C, the reaction mixture was azeotropically evaporated with toluene to give the acyl chloride. Separately, to a suspension of dried copper(I) iodide (84.4 mg, 0.44 mmol) in THF (2.0 ml), was slowly added Grignard reagent (8.50 ml, 8.00 mmol) at -15 °C to provide a cuprate solution. After the cuprate solution was stirred for 30 min at -15 °C, a solution of the acyl chloride in THF (1.80 ml) was added to the cuprate solution. After being stirred for 2 h at -15 °C, the reaction was quenched with saturated aqueous NH₄Cl, and the resulting mixture was extracted with hexane. The organic layer was washed with saturated aqueous NaHCO₃, saturated aqueous Na₂S₂O₃ and brine, dried over MgSO₄, filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 20/1) afforded **41a** (804 mg, 2.46 mmol, 61% for 2 steps, impure) as a white solid. R_f 0.43 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 4.12 (2H, q, *J* = 7.2 Hz, -CO₂CH₂CH₃), 2.40 (4H, m, H5, H7), 2.30 (2H, t, *J* = 7.6 Hz, H2), 1.63-1.53 (6H, m, H3, H4, H8), 1.25 (21H, m, -CH₂-, -CO₂CH₂CH₃), 0.87 (3H, t, *J* = 7.3 Hz, H18).

Synthesis of 41b. To monomethyl suberate (660 μl, 3.67 mmol) was added thionyl chloride (0.80 ml, 11.0 mmol). After being refluxed for 16 h at 80 °C, the reaction mixture was azeotropically evaporated with toluene to give the acyl chloride. Separately, to a suspension of dried copper(I) iodide (49.9 mg, 0.26

mmol) in THF (1.8 ml), was slowly added Grignard reagent (8.58 ml, 7.62 mmol) at -15 °C to provide a cuprate solution. After the cuprate solution was stirred for 30 min at -15 °C, a solution of the acyl chloride in THF (1.80 ml) was added to the cuprate solution. After being stirred for 2 h at -15 °C, the reaction was quenched with saturated aqueous NH₄Cl, and the resulting mixture was extracted with hexane. The organic layer was washed with saturated aqueous NaHCO₃, saturated aqueous Na₂S₂O₃ and brine, dried over MgSO₄, filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 20/1) afforded **41b** (344 mg, 1.10 mmol, 30% for 2 steps, impure) as a white solid. R_f 0.47 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 3.64 (3H, s, -CO₂Me), 2.36 (4H, m, H7, H9), 2.28 (2H, t, *J* = 7.6 Hz, H2), 1.63-1.53 (6H, m, H3, H6, H10), 1.23 (18H, s, -CH₂-), 0.87 (3H, t, *J* = 7.3 Hz, H18).

Synthesis of 41d. To a solution of 12-hydroxyoctadecanoic acid methyl ester (843 mg, 2.68 mmol) in dichloromethane (24.3 ml) were added dimethyl sulfoxide (1.52 ml, 21.4 mmol), triethylamine (1.49 ml, 10.7 mmol), and SO₃-Py (1.28g, 8.04 mmol) at 0 °C. After being stirred for 18 h at room temperature, the reaction was quenched with saturated aqueous Na₂S₂O₃, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with saturated aqueous NH₄Cl and brine, dried over Na₂SO₄, filtered, and evaporated to give the crude products. Purification by flash silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **41d** (693 mg, 2.22 mmol, 83%) as a white solid. R_f 0.60 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 3.64 (3H, s, -CO₂Me), 2.36 (4H, m, H11, H13), 2.28 (2H, t, *J* = 7.6 Hz, H2), 1.59 (1H, dt, *J* = 14.5, 7.5 Hz, H3), 1.53 (4H, m, H10, H14), 1.23 (18H, s, -CH₂-), 0.87 (3H, t, *J* = 7.3 Hz, H18).

Synthesis of 41e. To a solution of monomethyl tetradecanedioic acid (368 mg, 1.35 mmol) in toluene (2 ml) was added oxalyl chloride (0.16 ml, 1.76 mmol). After being refluxed for 1 h at room temperature, the

reaction mixture was evaporated to give the acyl chloride. Separately, to a suspension of dried copper(I) iodide (257 mg, 1.35 mmol) in THF (3.0 ml), was slowly added 1.6 M solution of *n*-BuLi in hexane (1.68 ml, 2.70 mmol) at 0 °C to provide a cuprate solution. After the cuprate solution was stirred for 30 min at 0 °C, a solution of the acyl chloride in THF (4.0 ml) was added to the cuprate solution. After being stirred for 1 h at 0 °C, the reaction was quenched with 1 M HCl, and the resulting mixture was extracted with diethyl ether. The organic layer was dried over Na₂SO₄, filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 50/1) afforded **41e** (192 mg, 0.61 mmol, 64% for 2 steps, impure) as a white solid. R_f 0.63 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 3.65 (3H, s, -CO₂Me), 2.36 (4H, m, H13, H15), 2.27 (2H, t, *J* = 7.6 Hz, H2), 1.60-1.49 (6H, m, H3, H12, H16), 1.23 (18H, s, -CH₂-), 0.87 (3H, t, *J* = 7.3 Hz, H18).

Synthesis of 41f. To a solution of monomethyl hexadecanedioic acid (726 mg, 2.42 mmol) in toluene (20 ml) was added oxalyl chloride (0.50 ml, 5.83 mmol). After being refluxed for 1 h at room temperature, the reaction mixture was evaporated to give the acyl chloride. Separately, to a suspension of dried copper(I) iodide (461 mg, 2.42 mmol) in THF (5.0 ml), was slowly added 1.0 M solution of ethyl magnesium bromide in THF (4.84 ml, 4.84 mmol) at -78 °C to provide a cuprate solution. After being stirred for 30 min at -78 °C, a solution of the acyl chloride in THF (4.0 ml) was added to the cuprate solution. After the mixture was stirred for 1 h at -78 °C, the reaction was quenched with saturated aqueous NH₄Cl, and the resulting mixture was extracted with diethyl ether. The organic layer was dried over Na₂SO₄, filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 20/1) afforded **41f** (484 mg, 1.55 mmol, 64% for 2 steps) as a white solid. R_f 0.63 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 3.64 (3H, s, -CO₂Me), 2.38 (2H, m, H15, H17), 2.28 (2H, t, *J* = 7.6 Hz, H2), 1.59 (2H, m, H3), 1.52 (2H, m, H14), 1.23 (20H, s, H4-H13), 1.03 (3H, t, *J* = 7.3 Hz, H18).

Synthesis of 42a-f. Compounds **42a-f** were synthesized from corresponding oxooctadecanoic esters **41a-f** in the following manner. To a solution of respective ketoester **41a** (1.13 g, 3.45 mmol) in methanol (30 ml) and CHCl_3 (15 ml) was added sodium borodeuteride (434 mg, 10.4 mmol) at 0 °C. After being stirred for 1 h at the same temperature, the reaction was quenched with acetic acid, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with saturated aqueous NaHCO_3 and brine, dried over MgSO_4 , filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **42a**.

42a: white solid (1.05g, 3.23 mmol, 93%) R_f 0.25 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 4.12 (2H, q, $J = 7.2$ Hz, $-\text{CO}_2\text{CH}_2\text{CH}_3$), 2.30 (2H, t, $J = 7.6$ Hz, H2), 1.64 (2H, m, H3), 1.55 (4H, m, H5, H7), 1.24 (22H, s, $-\text{CH}_2-$, $-\text{CO}_2\text{CH}_2\text{CH}_3$), 0.86 (3H, t, $J = 7.2$ Hz, H18).

42b: white solid (500 mg, 1.58 mmol, 85%) R_f 0.25 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 3.65 (3H, s, $-\text{CO}_2\text{Me}$), 2.28 (2H, t, $J = 7.6$ Hz, H2), 1.62 (2H, m, H3), 1.41 (4H, m, H7, H9), 1.24 (22H, s, $-\text{CH}_2-$), 0.86 (3H, t, $J = 7.2$ Hz, H18).

42c: (450 mg, 1.10 mmol, quant) R_f 0.42 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 2.28 (2H, t, $J = 7.6$ Hz, H2), 1.59 (2H, m, H3), 1.53 (1H, s, $-\text{OH}$), 1.40 (4H, m, H9, H11), 1.27 (22H, s, $-\text{CH}_2-$), 0.86 (3H, t, $J = 7.2$ Hz, H18).

42d: white solid (553 mg, 1.75 mmol, 93%) R_f 0.33 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 2.28 (2H, t, $J = 7.6$ Hz, H2), 1.60 (2H, m, H3), 1.40 (4H, m, H11, H13), 1.24 (22H, s, $-\text{CH}_2-$), 0.89 (3H, t, $J = 7.2$ Hz, H18).

42e: white solid (94.3 mg, 0.30 mmol, 68%) R_f 0.33 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 2.28 (2H, t, $J = 7.6$ Hz, H2), 1.59 (2H, m, H3), 1.41 (4H, m, H13, H15), 1.24 (22H, s, $-\text{CH}_2-$), 0.89 (3H, t, $J = 7.2$ Hz, H18).

42f: white solid (447 mg, 1.42 mmol, 92%) R_f 0.29 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 2.28 (2H, t, $J = 7.6$ Hz, H2), 1.59 (2H, m, H3), 1.41 (4H, m, H15, H17), 1.27 (22H, s, $-\text{CH}_2-$), 0.92 (3H, t, $J = 7.2$ Hz, H18).

Synthesis of 43a-f. Compounds **43a-f** were synthesized from corresponding **42a-f** in the following manner. To a solution of hydroxyoctadecanoic acid ester **42a** (1.05g, 3.20 mmol) in dichloromethane (31 ml) were added triethylamine (4.04 ml, 28.8 mmol), *N,N*-dimethyl-4-aminopyridine (480 mg, 3.93 mmol), and *p*-toluenesulfonyl chloride (3.69g, 19.4 mmol) at 0 °C. After being stirred for 40 h at room temperature, the reaction was quenched with water, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with saturated aqueous NaHCO_3 and brine, dried over MgSO_4 , filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded tosylate **43a**.

43a: yellow oil (1.15 g, 2.68 mmol, 74%) R_f 0.43 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 7.78 (2H, d, $J = 8.3$ Hz, *o*-ArH), 7.32 (2H, d, $J = 8.0$ Hz, *m*-ArH), 4.12 (2H, q, $J = 7.2$ Hz, $-\text{CO}_2\text{CH}_2\text{CH}_3$), 2.44 (3H, s, $-\text{CH}_3$), 2.21 (2H, t, $J = 7.6$ Hz, H2), 1.60 (2H, m, H3), 1.53 (4H, m, H5, H7), 1.26 (25H, m, $-\text{CH}_2-$, $-\text{CO}_2\text{CH}_2\text{CH}_3$), 0.88 (3H, t, $J = 7.2$ Hz, H18).

43b: yellow oil (613 mg, 1.31 mmol, 78%) R_f 0.41 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 7.77 (2H, d, $J = 8.3$ Hz, *o*-ArH), 7.30 (2H, d, $J = 8.0$ Hz, *m*-ArH), 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 2.42 (3H, s, $-\text{CH}_3$), 2.28 (2H, t, $J = 7.6$ Hz, H2), 1.59 (2H, m, H3), 1.53 (4H, m, H7, H9), 1.20 (22H, s, $-\text{CH}_2-$), 0.87 (3H, t, $J = 7.2$ Hz, H18).

43c: colorless oil (377 mg, 0.80 mmol, 93%) R_f 0.21 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 7.76 (2H, d, $J = 8.3$ Hz, *o*-ArH), 7.30 (2H, d, $J = 7.9$ Hz, *m*-ArH), 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 2.42 (3H, s, $-\text{CH}_3$), 2.28 (2H, t, $J = 7.6$ Hz, H2), 1.57 (2H, m, H3), 1.53 (4H, m, H9, H11), 1.19 (22H, s, $-\text{CH}_2-$), 0.86 (3H, t, $J = 7.2$ Hz, H18).

43d: yellow oil (922 mg, 1.96 mmol, 95%) R_f 0.21 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 7.77 (2H, d, J = 8.3 Hz, *o*-ArH), 7.30 (2H, d, J = 7.9 Hz, *m*-ArH), 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 2.42 (3H, s, $-\text{CH}_3$), 2.28 (2H, t, J = 7.6 Hz, H2), 1.60 (2H, m, H3), 1.52 (4H, m, H11,H13), 1.19 (22H, s, $-\text{CH}_2-$), 0.86 (3H, t, J = 7.2 Hz, H18).

43e: yellow solid (413 mg, 0.88 mmol, 79%) R_f 0.37 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 7.77 (2H, d, J = 8.3 Hz, *o*-ArH), 7.30 (2H, d, J = 8.0 Hz, *m*-ArH), 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 2.42 (3H, s, $-\text{CH}_3$), 2.28 (2H, t, J = 7.6 Hz, H2), 1.59 (2H, m, H3), 1.53 (4H, m, H13, H15), 1.20 (22H, s, $-\text{CH}_2-$), 0.80 (3H, t, J = 7.2 Hz, H18).

43f: yellow solid (668 mg, 1.42 mmol, 72%) R_f 0.34 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 7.77 (2H, d, J = 8.3 Hz, *o*-ArH), 7.30 (2H, d, J = 8.0 Hz, *m*-ArH), 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 2.42 (3H, s, $-\text{CH}_3$), 2.28 (2H, t, J = 7.6 Hz, H2), 1.59 (2H, m, H3), 1.53 (4H, m, H15, H17), 1.24 (22H, s, $-\text{CH}_2-$), 0.81 (3H, t, J = 7.2 Hz, H18).

Synthesis of 44a-f. To a solution of tosylate **43a** (1.15g, 0.54 mmol) in dimethyl sulfoxide (23.8 ml) was added sodium borodeuteride (403 mg, 9.50 mmol) at 0 °C. After being stirred for 12 h at 80 °C, the reaction was quenched with acetic acid, and the resulting mixture was extracted with ethyl acetate. The organic layer was washed with saturated aqueous NaHCO_3 and brine, dried over MgSO_4 , filtered, and evaporated to give the crude products. Purification by silica gel column chromatography (hexane/ethyl acetate = 10/1) afforded **44a**. **44b-f** were prepared in a similar manner.

44a: white solid (399 mg, 1.27 mmol, 54%) R_f 0.68 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 4.12 (2H, q, J = 7.2 Hz, $-\text{CO}_2\text{CH}_2\text{CH}_3$), 2.28 (2H, t, J = 7.6 Hz, H2), 1.61 (2H, m, H3), 1.25 (29H, m, $-\text{CH}_2-$, $-\text{CO}_2\text{CH}_2\text{CH}_3$), 0.88 (3H, t, J = 6.9 Hz, H18).

44b: white solid (160 mg, 0.53 mmol, 49%) R_f 0.73 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 3.65 (3H, s, $-\text{CO}_2\text{Me}$), 2.28 (2H, t, $J = 7.6$ Hz, H2), 1.60 (2H, m, H3), 1.24 (26H, s, $-\text{CH}_2-$), 0.86 (3H, t, $J = 6.9$ Hz, H18).

44c: white solid (149 mg, 0.50 mmol, 43%) R_f 0.73 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 3.65 (3H, s, $-\text{CO}_2\text{Me}$), 2.33 (2H, t, $J = 7.6$ Hz, H2), 1.61 (2H, m, H3), 1.19 (26H, s, $-\text{CH}_2-$), 0.86 (3H, t, $J = 7.2$ Hz, H18).

44d: white solid 180 mg, 0.60 mmol, 47%). R_f 0.72 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 2.33 (2H, t, $J = 7.6$ Hz, H2), 1.61 (2H, m, H3), 1.19 (26H, s, $-\text{CH}_2-$), 0.86 (3H, t, $J = 7.2$ Hz, H18).

44e: white solid (133 mg, 0.44 mmol, 50%) R_f 0.71 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 2.28 (2H, t, $J = 7.6$ Hz, H2), 1.60 (2H, m, H3), 1.24 (26H, s, $-\text{CH}_2-$), 0.86 (3H, t, $J = 7.2$ Hz, H18).

44f: white solid (224 mg, 0.75 mmol, 53%) R_f 0.71 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 3.64 (3H, s, $-\text{CO}_2\text{Me}$), 2.28 (2H, t, $J = 7.6$ Hz, H2), 1.60 (2H, m, H3), 1.24 (26H, s, $-\text{CH}_2-$), 0.86 (3H, t, $J = 7.2$ Hz, H18).

Synthesis of 45a-f. Synthesis of **45a-f** was completed by hydrolysis of **44a-f** in a similar manner to the synthesis of compound **32a**.

45a: white solid (365 mg, 1.27 mmol, quant) R_f 0.27 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 2.35 (2H, t, $J = 7.6$ Hz, H2), 1.63 (2H, m, H3), 1.26 (26H, s, $-\text{CH}_2-$), 0.88 (3H, t, $J = 6.9$ Hz, H18).

45b: white solid (140 mg, 0.49 mmol, 92%) R_f 0.21 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (500 MHz, CDCl_3) δ 2.35 (2H, t, $J = 7.6$ Hz, H2), 1.63 (2H, m, H3), 1.23 (26H, s, $-\text{CH}_2-$), 0.86 (3H, t, $J = 6.9$ Hz, H18).

45c: (134 mg, 0.47 mmol, 94%) as a white solid. R_f 0.10 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 2.33 (2H, t, $J = 7.6$ Hz, H2), 1.61 (2H, m, H3), 1.19 (26H, s, -CH₂-), 0.86 (3H, t, $J = 7.2$ Hz, H18).

45d: white solid (156 mg, 0.54 mmol, 91%) R_f 0.10 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 2.33 (2H, t, $J = 7.6$ Hz, H2), 1.61 (2H, m, H3), 1.19 (26H, s, -CH₂-), 0.86 (3H, t, $J = 7.2$ Hz, H18).

45e: white solid (118 mg, 0.41 mmol, 94%) R_f 0.23 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 2.32 (2H, t, $J = 7.6$ Hz, H2), 1.61 (2H, m, H3), 1.23 (26H, s, -CH₂-), 0.86 (3H, t, $J = 7.2$ Hz, H18).

45f: white solid (203 mg, 0.71 mmol, 94%) R_f 0.23 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 2.33 (2H, t, $J = 7.6$ Hz, H2), 1.62 (2H, m, H3), 1.23 (26H, s, -CH₂-), 0.86 (3H, t, $J = 7.2$ Hz, H18).

Synthesis of 46a-j. To a solution of **32a** (174 mg, 0.61 mmol) in THF (1.2 ml) were added *p*-nitrophenol (110 mg, 0.79 mmol) and *N,N'*-dicyclohexylcarbodiimide (162 mg, 0.79 mmol) at 0 °C. After being stirred for 17 h at room temperature, the resulting mixture was evaporated to give the crude products. Purification by flash silica gel column chromatography (hexane/ethyl acetate = 100/1) afforded **46a**. Compounds **46b, c, d, e, f, g, h, i** were prepared from **32b, 32c, 45a, b, c, d, e, f**, respectively, in a similar manner. Compound **46j** was from commercially available 18-*d*₃-stearic acid.

46a: white solid (208 mg, 0.51 mmol, 84% from **32a**). R_f 0.69 (silica gel, hexane/ethyl acetate = 5/1); ^1H NMR (500 MHz, CDCl_3) δ 8.25 (2H, m, *m*-ArH), 7.25 (2H, m, *o*-ArH), 2.58 (29%*, 2H, t, $J = 7.5$ Hz, H2), 1.72 (2H, t, $J = 7.0$ Hz, H3), 1.26 (28H, s, H4-H17), 0.86 (3H, t, $J = 7.0$ Hz, H18). *not deuterated H (%).

46b: white solid (71% from **32b**). R_f 0.65 (silica gel, hexane/ethyl acetate = 3/1); ¹H NMR (500 MHz, CDCl₃) δ 8.25 (2H, m, *m*-ArH), 7.25 (2H, m, *o*-ArH), 2.57 (2H, t, *J* = 7.6 Hz, H2), 1.23 (28H, s, H4-H17), 0.86 (3H, t, *J* = 7.2 Hz, H18).

46c: white solid (467 mg, 1.15 mmol, 72% from **32c**). R_f 0.58 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 8.25 (2H, m, *m*-ArH), 7.25 (2H, m, *o*-ArH), 2.57 (2H, t, *J* = 7.6 Hz, H2), 1.72 (2H, m, H3), 1.23 (28H, s, H5-H17), 0.86 (3H, t, *J* = 7.2 Hz, H18).

46d: white solid (416 mg, 1.02 mmol, 80% from **45a**). R_f 0.67 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 8.26 (2H, m, *m*-ArH), 7.27 (2H, m, *o*-ArH), 2.59 (2H, t, *J* = 7.4 Hz, H2), 1.76 (2H, m, H3), 1.26 (26H, s, -CH₂-), 0.88 (3H, t, *J* = 6.9 Hz, H18).

46e: white solid (125 mg, 0.31 mmol, 79% from **45b**). R_f 0.59 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 8.25 (2H, m, *m*-ArH), 7.27 (2H, m, *o*-ArH), 2.61 (2H, t, *J* = 7.4 Hz, H2), 1.76 (2H, m, H3), 1.23 (26H, s, -CH₂-), 0.88 (3H, t, *J* = 6.9 Hz, H18).

46f: white solid (93.1 mg, 0.23 mmol, 79% from **45c**). R_f 0.61 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 8.25 (2H, m, *m*-ArH), 7.25 (2H, m, *o*-ArH), 2.57 (2H, t, *J* = 7.6 Hz, H2), 1.74 (2H, m, H3), 1.23 (26H, s, -CH₂-), 0.86 (3H, t, *J* = 7.2 Hz, H18).

46g: white solid (96.5 mg, 0.24 mmol, 56% from **45d**). R_f 0.56 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 8.25 (2H, m, *m*-ArH), 7.25 (2H, m, *o*-ArH), 2.57 (2H, t, *J* = 7.6 Hz, H2), 1.74 (2H, m, H3), 1.23 (26H, s, -CH₂-), 0.86 (3H, t, *J* = 7.2 Hz, H18).

46h: white solid (107 mg, 0.26 mmol, 64% from **45e**). R_f 0.58 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 8.25 (2H, m, *m*-ArH), 7.25 (2H, m, *o*-ArH), 2.58 (2H, t, *J* = 7.6 Hz, H2), 1.74 (2H, m, H3), 1.23 (26H, s, -CH₂-), 0.86 (3H, t, *J* = 7.2 Hz, H18).

46i: white solid (183 mg, 0.45 mmol, 63% from **45f**). R_f 0.58 (silica gel, hexane/ethyl acetate = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 8.25 (2H, m, *m*-ArH), 7.25 (2H, m, *o*-ArH), 2.58 (2H, t, *J* = 7.6 Hz, H2), 1.74 (2H, m, H3), 1.23 (28H, s, -CH₂-), 0.86 (3H, t, *J* = 7.2 Hz, H18).

46j: white solid (105 mg, 0.26 mmol, 81%), R_f 0.61 (silica gel, hexane/ethyl acetate = 5/1); $^1\text{H NMR}$ (400 MHz, CDCl_3) δ 8.25 (2H, m, *m*-ArH), 7.25 (2H, m, *o*-ArH), 2.58 (2H, t, $J = 7.6$ Hz, H2), 1.74 (2H, m, H3), 1.23 (28H, s, $-\text{CH}_2-$).

Synthesis of 47. Compound **47** was synthesized from L-serine in the same manner as the preparation of **14** as a colorless oil. R_f 0.47 (silica gel, $\text{CHCl}_3/\text{MeOH}/\text{H}_2\text{O} = 65/25/4$); $^1\text{H NMR}$ (500 MHz, CD_3OD) δ 5.67 (1H, td, $J = 15.2, 6.8$ Hz, H5), 5.42 (1H, dd, $J = 15.8, 7.8$ Hz, H4), 4.26 (2H, br, α), 4.21 (1H, dd, $J = 6.8, 6.6$ Hz, H3), 4.01 (1H, m, H1), 3.94 (2H, m, H1, H2), 3.62 (2H, br, β), 3.21 (9H, s, $-\text{N}^+\text{Me}_3$), 2.02 (2H, br, H6), 1.42 (9H, s, Boc-*t*-Bu), 1.28 (22H, s, H7-17), 0.90 (12H, m, TBS-*t*-Bu, H18), 0.08 (3H, s, TBS-Me), 0.03 (3H, s, TBS-Me).

Synthesis of 2'- d_2 -, 3'- d_2 -, 4'- d_2 -, 6'- d_2 -, 8'- d_2 -, 10'- d_2 -, 12'- d_2 -, 14'- d_2 -, 16'- d_2 -, and 18'- d_3 -SSMs. To a solution of **47** (97 mg, 0.14 mmol) in dichlorometane (3.0 ml) was added trifluoroacetic acid (750 μl , 10.1 mmol) at 0 °C. After being stirred for 3 h at 0 °C, triethylamine was added, and the reaction mixture was evaporated to give the crude products. To a solution of the crude products in THF (1.43 ml) were added triethylamine (60 μl , 0.43 mmol), *N,N*-dimethyl-4-aminopyridine (19.2 mg, 0.16 mmol), and *p*-nitrophenylester **46a** (70 mg, 0.17 mmol). After being stirred for 13 h at room temperature, the resulting mixture was evaporated and precipitated by acetone. The precipitate was isolated by Celite filtration and washed with $\text{CHCl}_3/\text{MeOH}$ (1/1) to give the crude products. Purification by silica gel column chromatography ($\text{CHCl}_3/\text{MeOH}/\text{H}_2\text{O} = 65/25/4$) afforded TBS-protected SM as a white solid.

To a solution of TBS-protected SM in THF (2.0 ml) was added TBAF (1.0 M in THF, 0.66 ml, 0.66 mmol) at 0 °C. After being stirred for 17 h at room temperature, MeOH was added to the solution. The resulting mixture was evaporated and precipitated by acetone. The precipitate was isolated by Celite filtration, and washed with CHCl_3 -MeOH (1:1) to give the crude products. Purification by silica gel

column chromatography (CHCl₃/MeOH/H₂O = 65/25/4) afforded 2'-d₂-SSM (62.6 mg, 0.09 mmol 60% for 3 steps) as a white solid. Other deuterated SSMs were prepared from **47** and corresponding **46b-j** in a similar manner.

2'-d₂-SSM: white solid (62.6 mg, 0.09 mmol 60% for 3 steps). R_f 0.23 (silica gel, CHCl₃/MeOH/H₂O = 65/25/4); ¹H NMR (500 MHz, CD₃OD) δ 5.69 (1H, td, *J* = 15.0, 6.5 Hz, H5), 5.43 (1H, dd, *J* = 15.0, 8.0 Hz, H4), 4.26 (2H, br, α), 4.10 (1H, m, H1), 4.03 (1H, dd, *J* = 8.5, 7.3 Hz, H3), 3.95 (2H, m, H1, H2), 3.62 (2H, br, β), 3.21 (9H, s, -N⁺Me₃), 2.14 (29%*, br, H2'), 2.02 (2H, br, H6), 1.56 (2H, br, H3'), 1.28 (50H, s, -CH₂-), 0.89 (6H, t, *J* = 6.5 Hz, H18, H18'). *not deuterated H (%); ESI-HRMS *m/z* calcd for C₄₁H₈₁D₂N₂O₆PNa(M+Na)⁺ 755.6006, found 755.6011.

3'-d₂-SSM: white solid (73.6 mg, 0.10 mmol, 51% for 3 steps). R_f 0.22 (silica gel, CHCl₃/MeOH/H₂O = 65/25/4); ¹H NMR (500 MHz, CD₃OD) δ 5.69 (1H, td, *J* = 15.5, 6.6 Hz, H5), 5.44 (1H, dd, *J* = 14.3, 7.7 Hz, H4), 4.26 (2H, br, α), 4.10 (1H, m, H1), 4.03 (1H, dd, *J* = 8.7, 7.3 Hz, H3), 3.94 (2H, m, H1, H2), 3.62 (2H, br, β), 3.21 (9H, s, -N⁺Me₃), 2.17 (2H, br, H2'), 2.01 (2H, br, H6), 1.56 (2H, m, H3'), 1.28 (50H, s, -CH₂-), 0.88 (6H, t, *J* = 7.2 Hz, H18, H18').

4'-d₂-SSM: white solid (68.5 mg, 0.09 mmol, 47% for 3 steps). R_f 0.23 (silica gel, CHCl₃/MeOH/H₂O = 65/25/4); ¹H NMR (500 MHz, CD₃OD) δ 5.69 (1H, td, *J* = 15.3, 6.6 Hz, H5), 5.44 (1H, dd, *J* = 14.3, 7.7 Hz, H4), 4.26 (2H, br, α), 4.11 (1H, m, H1), 4.03 (1H, dd, *J* = 8.7, 7.3 Hz, H3), 3.96 (2H, m, H1, H2), 3.62 (2H, br, β), 3.21 (9H, s, -N⁺Me₃), 2.17 (2H, br, H2'), 2.02 (2H, br, H6), 1.56 (2H, m, H3'), 1.28 (50H, s, -CH₂-), 0.89 (6H, t, *J* = 7.2 Hz, H18, H18'); ESI-HRMS *m/z* calcd for C₄₁H₈₁D₂N₂O₆PNa⁺ (M+Na)⁺ 755.6006, found 755.6014.

6'-d₂-SSM: white solid (31.4 mg, 0.04 mmol, 31% for 3 steps). R_f 0.24 (silica gel, CHCl₃/MeOH/H₂O = 65/25/4); ¹H NMR (500 MHz, CD₃OD) δ 5.68 (1H, td, *J* = 15.0, 6.5 Hz, H5), 5.42 (1H, dd, *J* = 15.0, 7.5 Hz, H4), 4.25 (2H, br, α), 4.08 (1H, m, H1), 4.02 (1H, dd, *J* = 8.7, 7.3 Hz, H3), 3.93 (2H, m, H1, H2), 3.61 (2H, t, *J* = 4.5 Hz, β), 3.20 (9H, s, -N⁺Me₃), 2.17 (2H, br, H2'), 2.01 (2H, br, H6), 1.55 (2H, m, H3'), 1.28

(50H, s, -CH₂-), 0.88 (6H, t, $J = 7.2$ Hz, H18, H18'); ESI-HRMS m/z calcd for C₄₁H₈₁D₂N₂O₆PNa⁺ (M+Na)⁺ 755.6006, found 755.6013.

8'-d₂-SSM: white solid (72.4 mg, 0.10 mmol, 70% for 3 steps). R_f 0.23 (silica gel, CHCl₃/MeOH/H₂O = 65/25/4); ¹H NMR (500 MHz, CD₃OD) δ 5.68 (1H, td, $J = 15.5, 6.5$ Hz, H5), 5.42 (1H, dd, $J = 15.5, 7.5$ Hz, H4), 4.25 (2H, br, α), 4.08 (1H, m, H1), 4.01 (1H, dd, $J = 8.7, 7.3$ Hz, H3), 3.92 (2H, m, H1, H2), 3.61 (2H, t, $J = 4.5$ Hz, β), 3.20 (9H, s, -N⁺Me₃), 2.16 (2H, br, H2'), 2.01 (2H, br, H6), 1.55 (2H, m, H3'), 1.28 (50H, s, -CH₂-), 0.88 (6H, t, $J = 7.2$ Hz, H18, H18'); ESI-HRMS m/z calcd for C₄₁H₈₁D₂N₂O₆PNa⁺ (M+Na)⁺ 755.6006, found 755.6013.

10'-d₂-SSM: white solid (77.9 mg, 0.11 mmol, 64% for 3 steps). R_f 0.22 (silica gel, CHCl₃/MeOH/H₂O = 65/25/4); ¹H NMR (500 MHz, CD₃OD) δ 5.69 (1H, td, $J = 15.5, 6.5$ Hz, H5), 5.44 (1H, dd, $J = 15.0, 7.5$ Hz, H4), 4.25 (2H, br, α), 4.08 (1H, m, H1), 4.02 (1H, dd, $J = 8.7, 7.3$ Hz, H3), 3.93 (2H, m, H1, H2), 3.61 (2H, br, β), 3.20 (9H, s, -N⁺Me₃), 2.15 (2H, br, H2'), 2.01 (2H, br, H6), 1.56 (2H, m, H3'), 1.27 (50H, s, -CH₂-), 0.89 (6H, t, $J = 7.2$ Hz, H18, H18'); ESI-HRMS m/z calcd for C₄₁H₈₁D₂N₂O₆PNa⁺ (M+Na)⁺ 755.6006, found 755.6035.

12'-d₂-SSM: white solid (65.3 mg, 0.10 mmol, 54% for 3 steps). R_f 0.23 (silica gel, CHCl₃/MeOH/H₂O = 65/25/4); ¹H NMR (500 MHz, CD₃OD) δ 5.68 (1H, td, $J = 15.5, 6.0$ Hz, H5), 5.44 (1H, dd, $J = 14.3, 7.7$ Hz, H4), 4.25 (2H, br, α), 4.08 (1H, m, H1), 4.02 (1H, dd, $J = 8.7, 7.3$ Hz, H3), 3.93 (2H, m, H1, H2), 3.62 (2H, br, β), 3.20 (9H, s, -N⁺Me₃), 2.15 (2H, br, H2'), 2.00 (2H, br, H6), 1.56 (2H, m, H3'), 1.27 (50H, s, -CH₂-), 0.88 (6H, t, $J = 7.0$ Hz, H18, H18'); ESI-HRMS m/z calcd for C₄₁H₈₁D₂N₂O₆PNa⁺ (M+Na)⁺ 755.6006, found 755.6012.

14'-d₂-SSM: white solid (75.5 mg, 0.10 mmol, 75%) for 3 steps. R_f 0.23 (silica gel, CHCl₃/MeOH/H₂O = 65/25/4); ¹H NMR (500 MHz, CD₃OD) δ 5.70 (1H, td, $J = 15.0, 7.0$ Hz, H5), 5.44 (1H, dd, $J = 14.5, 8.0$ Hz, H4), 4.26 (2H, br, α), 4.09 (1H, m, H1), 4.03 (1H, dd, $J = 8.7, 7.3$ Hz, H3), 3.93 (2H, m, H1, H2), 3.62 (2H, t, $J = 4.0$ Hz, β), 3.20 (9H, s, -N⁺Me₃), 2.16 (2H, br, H2'), 2.01 (2H, br, H6), 1.56 (2H, m, H3'), 1.27

(50H, s, -CH₂-), 0.88 (6H, t, $J = 7.2$ Hz, H18, H18'); ESI-HRMS m/z calcd for C₄₁H₈₁D₂N₂O₆PNa⁺ (M+Na)⁺ 755.6006, found 755.6046.

16'-d₂-SSM: white solid (74.1 mg, 0.10 mmol, 53% for 3 steps). R_f 0.24 (silica gel, CHCl₃/MeOH/H₂O = 65/25/4); ¹H NMR (500 MHz, CD₃OD) δ 5.68 (1H, td, $J = 15.5, 6.5$ Hz, H5), 5.44 (1H, dd, $J = 15.0, 7.5$ Hz, H4), 4.26 (2H, br, α), 4.09 (1H, m, H1), 4.02 (1H, dd, $J = 8.7, 7.3$ Hz, H3), 3.93 (2H, m, H1, H2), 3.62 (2H, t, $J = 4.0$ Hz, β), 3.20 (9H, s, -N⁺Me₃), 2.16 (2H, br, H2'), 2.01 (2H, br, H6), 1.56 (2H, m, H3'), 1.27 (50H, s, -CH₂-), 0.89 (6H, t, $J = 7.2$ Hz, H18, H18'); ESI-HRMS m/z calcd for C₄₁H₈₁D₂N₂O₆PNa⁺ (M+Na)⁺ 755.6006, found 755.6003.

18'-d₃-SSM: white solid (73.9 mg, 0.10 mmol, 63% for 2 steps). R_f 0.19 (silica gel, CHCl₃/MeOH/H₂O = 65/25/4); ¹H NMR (400 MHz, CD₃OD) δ 5.69 (1H, td, $J = 15.5, 6.5$ Hz, H5), 5.44 (1H, dd, $J = 15.0, 7.5$ Hz, H4), 4.27 (2H, br, α), 4.09 (1H, m, H1), 4.04 (1H, dd, $J = 8.7, 7.3$ Hz, H3), 3.95 (2H, m, H1, H2), 3.63 (2H, t, $J = 4.0$ Hz, β), 3.21 (9H, s, -N⁺Me₃), 2.17 (2H, br, H2'), 2.02 (2H, br, H6), 1.57 (2H, m, H3'), 1.29 (50H, s, -CH₂-), 0.89 (3H, t, $J = 7.2$ Hz, H18); ESI-HRMS m/z calcd for C₄₁H₈₀D₃N₂O₆PNa(M+Na)⁺ 756.6069, found 756.6078.