Synthesis of Carboxymethyl GLA-60 Ether Derivatives Containing an Olefin in Their Chains and Their LPS-Antagonistic Activities

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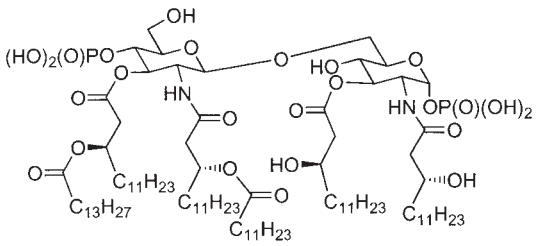
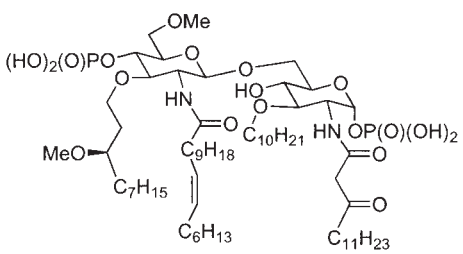
Anomeric carboxymethyl GLA-60 olefine derivatives having ether chains instead of ester chains in their side chains were synthesized and their biological activities toward both human U937 cells and mouse PEC-macrophage cells were measured. The species-specific behavior of these compounds in humans (LPS-antagonistic) and mice (very weak LPS-antagonistic, but almost inactive) found this time was different from that in humans (LPS-antagonistic) and mice (endotoxic) found in the biosynthetic precursor of lipid A, such as lipid IVa. However, this fact also shows, interestingly enough, that a difference exists in the molecular recognition between human and mouse LPS receptors.

Since Shiba and Kusumoto's total synthesis of lipid A (Fig. 1),[1] a toxic component of endotoxin (lipopolysaccharide; LPS) existing in the outer surface membrane of Gram-negative bacteria, the study of endotoxin has developed extensively.[2] Endotoxin and its related compounds have been investigated as anticancer drugs[2] by stimulating the immune system.[3] Also, in recent years, lipid A-related compounds have been studied as LPS-antagonists, which may have potential as immunosuppressants in autoimmune diseases and septicemia by deactivating the LPS-induced immune system.[2] In fact, a nontoxic natural lipid A-related compound isolated from *Rhodobacter sphaeroides*[4] showed a LPS-antagonistic activity, and the Eisai group has developed a related compound, E5564, as a highly potent anti-septicemia drug.[5]

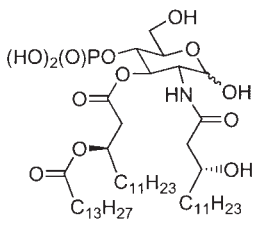
During our investigation of the biological activities of compounds related to GLA-60,[6] which is a nonreducing distal subunit of a lipid A analogue, we found that some anomeric α-carboxymethyl GLA-60 analogues had LPS-antagonistic activity toward human U937 cells.[7] It was also found that LPS-antagonist for human macrophages obtained from *Rhodobacter sphaeroides* having a unique structural feature, that is, containing an olefine and a ketone in its long chains of fatty acids, does not show LPS-agonistic activity toward mouse macrophages.[8] Furthermore, it was revealed by the Eisai group that many lipid A-related compounds having an olefinic double bond in their molecules behave as LPS-antagonists to-



Lipid A



E-5564



GLA-60

Fig. 1. Structures of Lipid A, E-5564 and GLA-60.

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ward both human and murine macrophages.[8] Therefore, we were interested in the LPS-antagonistic activity of the anomeric carboxymethyl GLA-60 ether derivatives containing an olefin in their molecules. We synthesized compounds **16**, **17**, **24**, **25**, **37**, and **38** to measure the LPS-antagonistic activity toward human U937 cells and mouse PEC-macrophage cells. We would like to describe the synthesis of the compounds and their LPS-antagonistic activity in this paper.

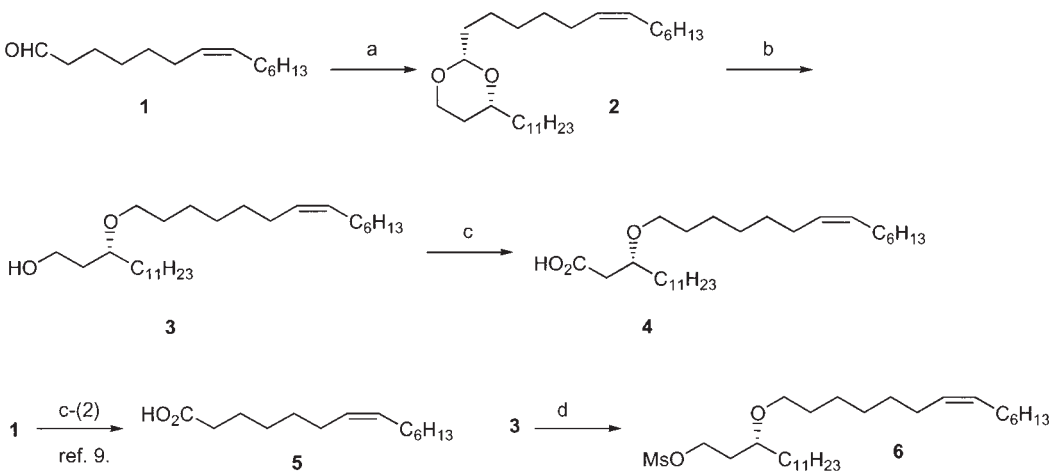
Results and Discussion

Firstly, we tried the synthesis of C-6 hydroxy compound **16** and C-6 methoxy compound **17** possessing four chains in their molecules. The compounds (*R*)-3-(alkenyloxy)tetradecanoic acid **4**, carboxylic acid **5**[9] and mesylate **6** for side chains were synthesized from aldehyde **1**, which was obtained by a modification of reported methods[10] (Scheme 1). The acetal **2** of (*R*)-tetradecane-1,3-diol[11] and aldehyde **1**, obtained by a treatment with camphorsulfonic acid (CSA) as a catalyst, were converted to primary alcohol **3** by hydridodiisobutylaluminum (DIBAL) reduction. A Swern oxidation of alcohol **3** with oxalyl chloride and dimethyl sulfoxide (DMSO) yielded an aldehyde, which was further oxidized to carboxylic acid **4** by sodium chlorite in the presence of 2-methyl-2-butene and sodium dihydrogenphosphate in *t*-butyl alcohol.[9] The same treatment of aldehyde **1** also gave carboxylic acid **5**.[9] On the other hand, the mesylation of **3** by methanesulfonyl chloride (MsCl) using *N*,*N*-diisopropylethylamine (i-Pr_2NEt) as a base gave mesylate **6**.

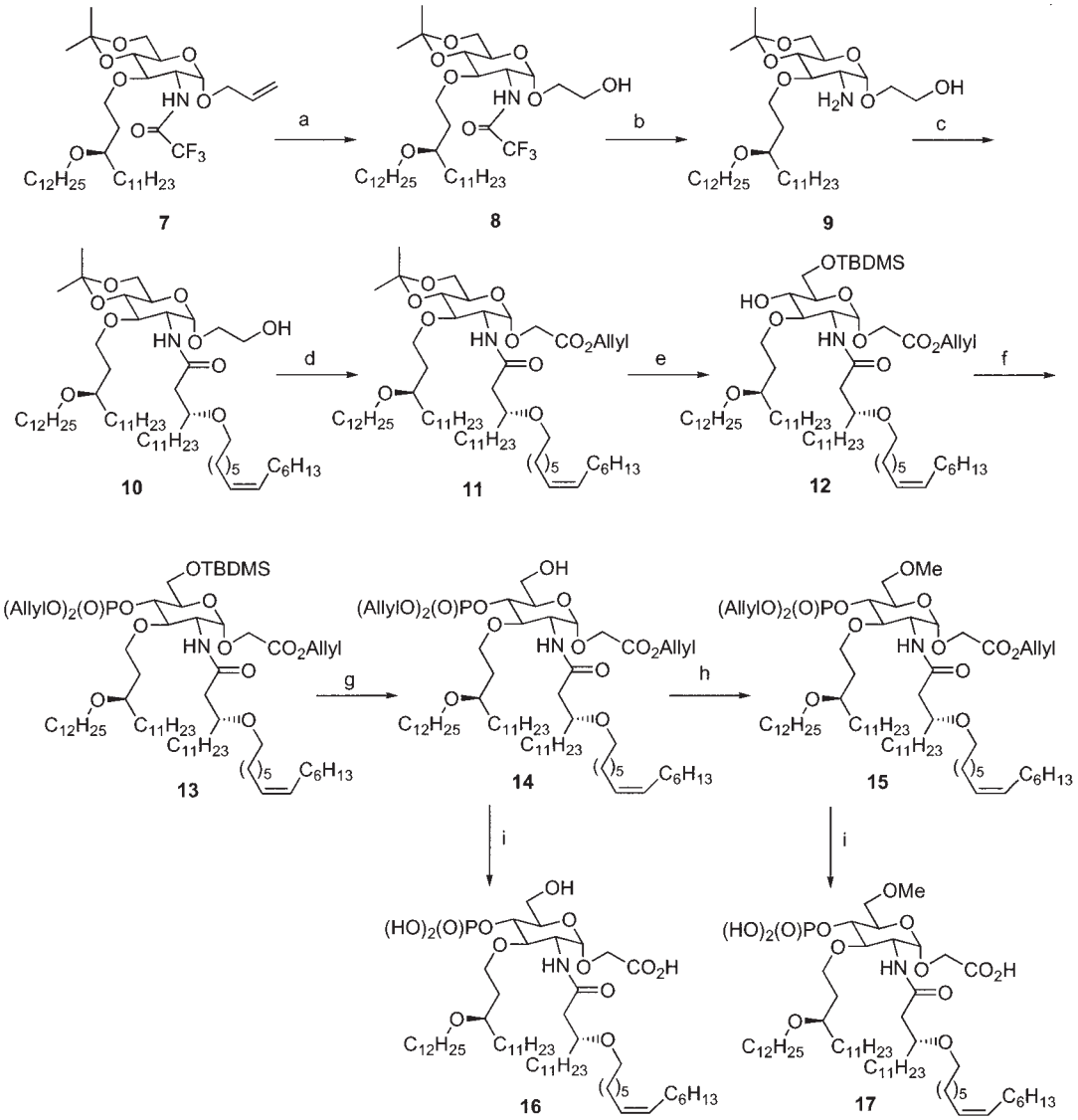
The starting allyl 2-deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-4,6-*O*-isopropylidene-2-trifluoroacetamido-α-D-glucopyranoside **7**[12] was converted to anomeric 2-hydroxyethyl compound **8** by OsO_4–$NaIO_4$ cleavage of the allylic double bond and successive reduction of the resulting aldehyde with $NaBH_4$ according to the reported procedure.[13] Saponification of trifluoroacetamide group of **8** with methanolic NaOH gave amine **9**. A treatment of **9** with carboxylic acid **4** and dicyclohexylcarbodiimide (DCC) gave amide **10**. Dess-Martin periodinane oxidation[14] of alcohol **10**, successive treatments of the resulting aldehyde with sodium chlorite in the presence of 2-methyl-2-butene and sodium dihydrogenphosphate in *t*-BuOH,[13] and finally esterification of the resulting carboxylic acid with allyl bromide and Et_3N gave allyl ester **11**. Acetonide of **11** was cleaved with CSA in MeOH, and successive silylation of the C-6 primary alcohol of the resulting diol with *t*-butyldimethylsilyl chloride (TBDMSCl) and imidazole as a base yielded silyl ether **12**. The treatment of the C-4 secondary alcohol of **12** with diallyl diisopropylphosphoramidite and 1*H*-tetrazole, and a successive treatment of the resulting phosphite with 40% H_2O_2 afforded phosphate **13**. The treatment of **13** with 5% aq. H_2SO_4 in acetone for desilylation gave alcohol **14**, which was treated with trimethyloxonium tetrafluoroborate (Me_3OBF_4) in the presence of 2,6-di-*t*-butyl-4-methylpyridine (DTBMP) to give a C-6 methoxy compound **15**. The allyl protecting groups of **14** and **15** were cleaved by reactions with PPh_3, Et_3N–HCOOH, and $[Pd(PPh_3)_4]$ in tetrahydrofuran (THF) at 50 °C for 4 hours to give deprotected compounds **16** and **17**, respectively[15] (Scheme 2).

Secondly, we tried to synthesize the C-6 hydroxy compound **24** and the C-6 methoxy compound **25** possessing three chains in their molecules. The reaction of **9** with (*Z*)-tetradec-7-enoic acid using DCC as a condensing reagent gave amide **18**, which was converted to allyl ester **19** according to the same procedure from **10** to **11**. The same procedure of **19** from **11** to **16** or **17** via compounds **12**, **13** and **14** or compounds **12**, **13**, **14** and **15** gave **24** or **25** through compounds **20**, **21**, and **22** or compounds **20**, **21**, **22**, and **23**, respectively (Scheme 3).

Thirdly, we tried to synthesize C-6 hydroxy compound **37** and C-6 methoxy compound **38** possessing three chains in their molecules. Compound **26**[16] was converted to alcohol **27** according to the same procedure from **7** to **8**. Selective protection of the primary alcohol of **27** with 4-methoxybenzyl chloride in DMF using NaH as a base gave ether **28**. A treatment of the secondary alcohol **28** in DMF with mesylate **6** using NaH as a base gave ether **29**. The trifluoroacetyl protecting group of amide **29** was cleaved by 1 M aq NaOH in EtOH, giving free amine, which was treated with 2,2-difluorotetradecanoic acid using DCC as a condensing reagent and 4-dimethylaminopyridine (DMAP) to afford amide **30**. Deprotection of the 4-



Scheme 1. Reagents and conditions: a) (*R*)-tetradecane-1,3-diol, CSA, benzene, azeotropic distillation, 100%; b) DIBAL, toluene, 50 °C, 10 h, 81%; c) (1) $(COCl)_2$, DMSO, CH_2Cl_2, −78 °C, 1 h, then Et_3N, 0 °C, 1 h; (2) $NaClO_2$, 2-methyl-2-butene, NaH_2PO_4, *t*-BuOH, rt, 3 h, two steps 88% (**4**) and 96% (**5**); d) $MeSO_2Cl$, i-Pr_2NEt, CH_2Cl_2, rt, 4 h, 100%.

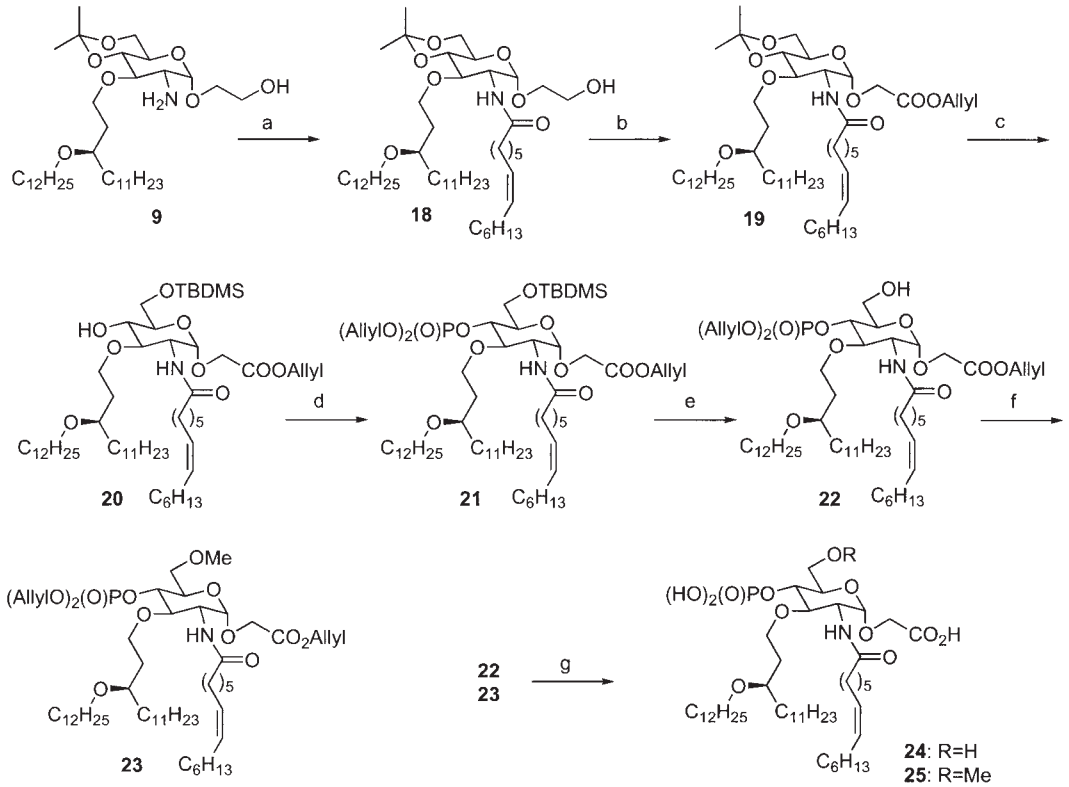


Scheme 2. Reagents and conditions: Allyl = $CH_2CH{=}CH_2$; TBDMS = *t*-butyldimethylsilyl; a) (1) OsO_4, $NaIO_4$, acetone–H_2O, rt, 1.5 h, (2) $NaBH_4$, MeOH, 0 °C, 30 min, 2 steps 47%; b) NaOH, MeOH–H_2O, 50 °C, 5 h, 93%; c) **4**, DCC, CH_2Cl_2, 0 °C, 4 h, 85%; d) (1) Dess-Martin periodinane, 0 °C, 3 h, (2) $NaClO_2$, NaH_2PO_4, 2-methyl-2-butene, *t*-BuOH–H_2O, rt, 2 h, (3) $CH_2{=}CHCH_2Br$, Et_3N, DMF, 50 °C, 4 h, three steps, 46%; e) CSA, MeOH, rt, 2 h, then TBDMSCl, imidazole, DMF, rt, 2.5 h, 87%; f) *i*-$Pr_2NP(OCH_2CH{=}CH_2)_2$, 1*H*-tetrazole, THF, rt, 1.5 h, then aq H_2O_2, rt, 1.5 h, 81%; g) aq 5% H_2SO_4, acetone, rt, 4.5 h, 80%; h) Me_3OBF_4, DTBMP, CH_2Cl_2, rt, 54%; i) [$Pd(PPh_3)_4$], PPh_3, Et_3N–HCOOH, THF, 50 °C, 4 h, 78% (**16**) and 85% (**17**).

methoxybenzyl ether of **30** with 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) and H_2O was performed to yield alcohol **31**, which was converted to allyl ester **32** according to the same procedure from **10** to **11**. The acetonide of **32** was deprotected by a treatment with 80% aq. AcOH at 60 °C to afford diol **33**. The same procedure of **33** from **11** to **13** gave diallyl phosphate **34**. A treatment of **34** with 3 M aq HCl in THF afforded alcohol **35**, which was further converted to C-6-methyl ether **36** by Me_3OBF_4 treatment. The deprotection of allyl groups from **35** and **36** with [$Pd(PPh_3)_4$], PPh_3, Et_3N, and HCOOH in THF yielded C-4 phosphono carboxylic acids **37** and **38**, respectively (Scheme 4).

Biological activity: The inhibitory activity of the six synthesized compounds (**16**, **17**, **24**, **25**, **37**, and **38**) on LPS-induced TNFα production was investigated in vitro using both human monoblastic U937 cells and mouse PEC-macrophage cells. The IC_{50} values (nM) of these six compounds (**16**, **17**, **24**, **25**, **37**, and **38**) toward human monoblastic U937 cells were 20.0, 13.0, 8.6, 37.0, 7.1, and 15.0, respectively. The activity toward human monoblastic U937 cells of these six monosaccharide compounds having a double bond in their three or four long chains were disappointingly much less active than that of our previously reported lipid A-type pyran carboxylic acids (IC_{50} = 0.6–6.4 nM).[12] In conclusion, it might be true that even though the compounds have a double bond in the long chains, if they are monosaccharides the LPS-antagonistic activity toward human monoblastic U937 cells would not exceed that of the lipid A-type disaccharides.

Additionally, the IC_{50} values (nM) of these six compounds (**16**, **17**, **24**, **25**, **37**, and **38**) toward mouse PEC-macrophage



Scheme 3. Reagents and conditions: Allyl = $CH_2CH{=}CH_2$; TBDMS = *t*-butyldimethylsilyl; a) **5**, DCC, CH_2Cl_2, 0 °C, 2.5 h, 84%; b) (1) Dess-Martin periodinane, CH_2Cl_2, 0 °C, 3.5 h, 0 °C, 3 h, (2) $NaClO_2$, NaH_2PO_4, 2-methyl-2-butene, *t*-BuOH–H_2O, rt, 1 h, (3) $CH_2{=}CHCH_2Br$, Et_3N, DMF, rt, 11 h, three steps 51%; c) (1) CSA, MeOH–THF, rt, 1 h; (2) TBDMS-Cl, imidazole, DMF, rt, 1 h, two steps, 99%; d) *i*-$Pr_2NP(OCH_2CH{=}CH_2)_2$, 1*H*-tetrazole, THF, rt, 3 h, and then 40% aq H_2O_2, 1 h, 85%; e) 5% aq H_2SO_4, acetone, rt, 5 h, 76%; f) Me_3OBF_4, DTBMP, CH_2Cl_2, rt, 4 h, 54%; g) [$Pd(PPh_3)_4$], PPh_3, Et_3N–HCOOH, THF, 50 °C, 7 h, 78% (**24**) and 85% (**25**).

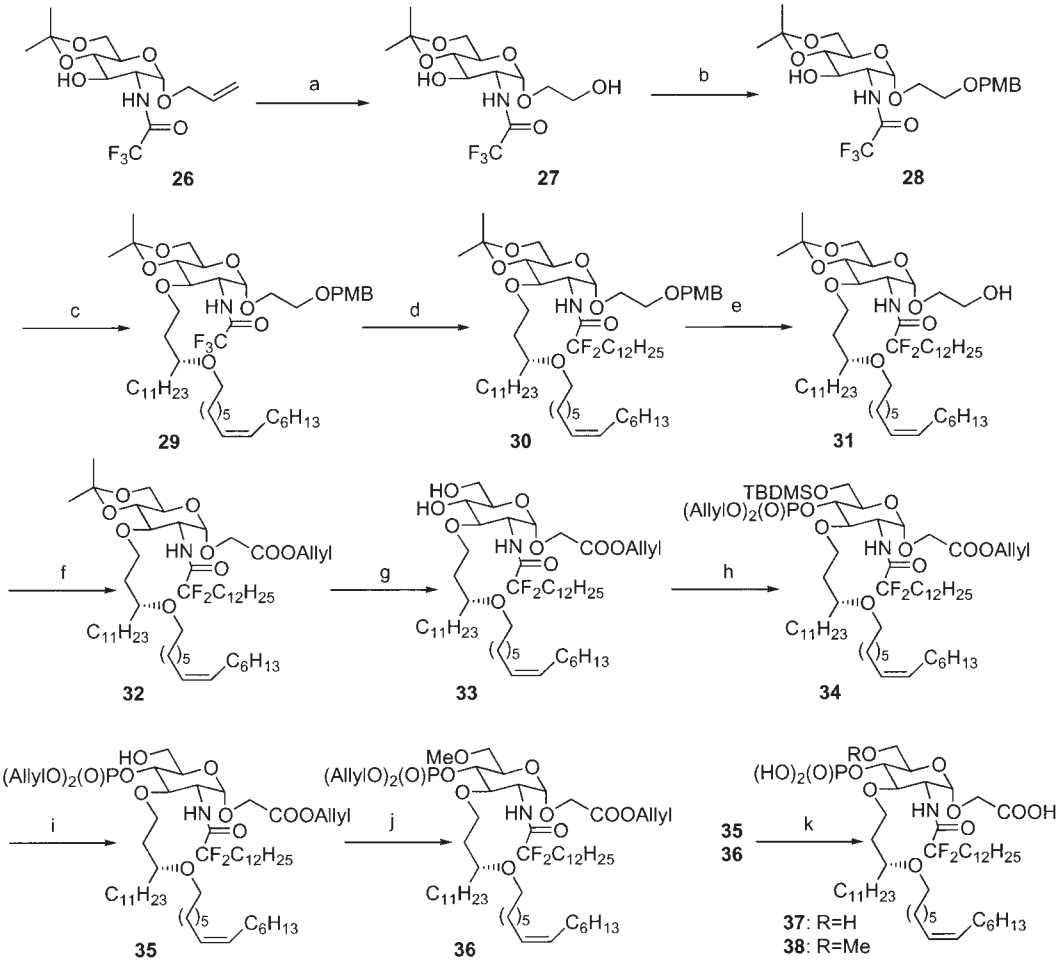
cells were >10000, >10000, 1686, 1907, 6141, and 8259, respectively. Among these compounds, compounds **16** and **17** having four chains in their molecules are completely lacking any activity. The other four compounds (**24**, **25**, **37**, and **38**) have three chains in their molecules. Also, the activity of compounds **37** and **38**, containing two fluorines, is much weaker than compounds **24** and **25**. The difference of C-6 OH and OMe does not much affect the activity. In conclusion, as compared with the activity toward human monoblastic U937 cells, the activity of these six compounds toward mouse PEC-macrophage cells is almost inactive.

The species-specific behavior of these compounds in humans (LPS-antagonistic) and mice (very weak LPS-antagonistic, but almost inactive) found this time was different from that in humans (LPS-antagonistic) and mice (endotoxic) found in the biosynthetic precursor of lipid A, such as lipid IVa.[17] However, this fact also shows, interestingly enough, that a difference exists in the molecular recognition between human and mouse LPS receptors.

Experimental

^{1}H NMR spectra were recorded with JEOL-GSX 400 and JNM-ECT 500 spectrometers using TMS as an internal standard. IR absorption spectra were measured with an IR A-2 spectrophotometer, and mass spectra were obtained with a JMS-700 mass spectrometer. Separation of compounds by column chromatography was performed with silica gel 60 (230–400 mesh ASTM) under a slightly elevated pressure (1.1–1.8 atm) for easy elution. Commercially available anhydrous THF and dichloromethane were used for the reactions. DMF and pyridine were dried by storage over 4 Å molecular sieves.

(*Z*)-7-Tetradecenal (1). (i) To a solution of 1-octyne (45.1 mL, 307 mmol) in THF (450 mL) was slowly added a 1.5 M solution of *n*-BuLi in hexane (204 mL, 307 mmol) at −45 °C. After stirring for 10 min at 0 °C, this reaction mixture was cooled to −45 °C and hexamethylphosphoric triamide (HMPA) (53.4 mL, 307 mmol) and 6-bromohexan-1-ol tetrahydropyranyl ether (67.8 g, 256 mmol) were added. After stirring for 22 h at room temperature, to the resulting mixture was slowly added sat. aq NH_4Cl (50 mL), and the mixture was poured into water. After extraction with ether, the organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product, which was dissolved in MeOH (400 mL) containing CSA (2.2 g, 10 mmol). After stirring for 24 h at room temperature, to the reaction mixture was added Et_3N (50 mL), and the mixture was evaporated in vacuo to give a crude product, which was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford 65.4 g of a crude product, which was purified by distillation under reduced pressure (bp 130–138 °C at 4 mmHg) to give tetradec-7-yn-1-ol (47.7 g, 88%) as a colorless oil; bp 130–138 °C (at 4 mmHg). IR (neat) 3341, 2932, 2859, 1464 cm^{-1}. ^{1}H NMR (500 MHz, $CDCl_3$) δ



Scheme 4. Reagents and conditions: Allyl = $CH_2CH{=}CH_2$; PMB = 4-methoxybenzyl; TBDMS = *t*-butyldimethylsilyl; a) (1) OsO_4, $NaIO_4$, acetone–H_2O–*t*-BuOH, rt, 1.5 h, (2) $NaBH_4$, MeOH, 0 °C, 30 min; 2 steps 70%; b) PMBCl, NaH, DMF, 0 °C, 15 min, 87%; c) **6**, NaH, DMF, rt, 3 h, 85%; d) (1) aq 1 M NaOH, EtOH, 60 °C, 4 h, (2) $HOOCCF_2C_{12}H_{25}$, DCC, DMAP, CH_2Cl_2, rt, 1 h, two steps, 96%; e) DDQ, CH_2Cl_2, H_2O, rt, 2 h, 84%; f) (1) Dess-Martin periodinane, 0 °C, 3 h, (2) $NaClO_2$, NaH_2PO_4, 2-methyl-2-butene, *t*-BuOH–H_2O, rt, 2 h, (3) $CH_2{=}CHCH_2Br$, Et_3N, DMF, 50 °C, 4 h, three steps, 71%; g) aq 80% AcOH, 60 °C, 3 h, 93%; h) (1) TBDMS-Cl, DMAP, CH_2Cl_2, rt, 3 h, (2) *i*-$Pr_2NP(OCH_2CH{=}CH_2)_2$, 1*H*-tetrazole, THF, rt, 1.5 h, then aq 30% H_2O_2, 0 °C, 1 h, 84%; i) 3 M aq HCl, THF, rt, 5 h, 91%; j) Me_3OBF_4, DTBMP, CH_2Cl_2, rt, 4 h, 77%; k) $[Pd(PPh_3)_4]$, PPh_3, Et_3N–HCOOH, THF, 50 °C, 7 h, 85% (**37**) and 92% (**38**), respectively.

0.89 (3H, t, $J = 7.8$ Hz), 1.23–1.53 (16H, m), 1.58 (2H, quintet, $J = 7.8$ Hz), 2.12–2.18 (4H, m), 3.65 (2H, t, $J = 5.9$ Hz). HRFABMS *m/z* (positive-ion); Calcd for $C_{14}H_{27}O$ $(M + H)^+$: 211.2062. Found: 211.2053.

(ii) To a solution of tetradec-7-yn-1-ol (10.6 g, 50 mmol) in hexane (200 mL) was added Lindlar's catalyst (1.0 g). After stirring for 9 h at room temperature under a hydrogen atmosphere, the reaction mixture was filtered using Celite. The filtrate was concentrated in vacuo to afford a crude product of (*Z*)-tetradec-7-en-1-ol. To a solution of $(COCl)_2$ (8.7 mL, 100 mmol) in CH_2Cl_2 (200 mL) was slowly added DMSO (14.2 mL, 200 mmol) at −78 °C. After stirring for 10 min, to this reaction mixture was added a solution of the previously obtained (*Z*)-tetradec-7-en-1-ol (10.7 g, 50 mmol) in CH_2Cl_2 (200 mL). After stirring for 1 h, the reaction mixture was slowly added Et_3N (55 mL, 400 mmol) and stirred for 1 h at 0 °C. The mixture was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product, which was purified by silica-gel column chromatography. Elution with hexane gave **1** (8.7 g, 83%) as a colorless oil. IR (neat) 1728 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (3H, t, $J = 6.6$ Hz), 1.23–1.40 (12H, m), 1.64 (2H, quintet, $J = 7.3$ Hz), 1.95–2.07 (4H, m), 2.42 (2H, dt, $J = 1.5$ Hz, 6.6 Hz), 5.29–5.40 (2H, m), 9.77 (1H, d, $J = 1.5$ Hz). HRFABMS *m/z* (positive-ion); Calcd for $C_{14}H_{26}O$ $(M)^{+\bullet}$: 210.1984. Found: 210.1980.

(2*S*,4*R*)-4-Undecyl-2-[(*Z*)-tridec-6-enyl]-1,3-dioxane (2). To a solution of **1** (10.1 g, 48 mmol) and (*R*)-tetradecan-1,3-diol (12.2 g, 53 mmol) in benzene (100 mL) was added CSA (0.50 g, 2.2 mmol). After refluxing for 2 h at 50 °C at 70 mmHg to remove H_2O azeotropically, the reaction mixture was cooled to room temperature and sat. aq $NaHCO_3$ (10 mL) was added. The resulting mixture was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc:hexane (0:10, and then 1:9) afforded **2** (21.4 g, quantitatively) as a colorless oil. $[\alpha]_D$ −1.1 (c 0.8, $CHCl_3$). IR

(neat) 2925, 2855, 1465 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (3H, t, $J = 6.8$ Hz), 0.89 (3H, t, $J = 6.8$ Hz), 1.23–1.46 (34H, m), 1.54–1.68 (4H, m), 1.97–2.05 (4H, m), 3.52–3.59 (1H, m), 3.71 (1H, dt, $J = 11.7$ Hz, 2.9 Hz), 4.09 (1H, dd, $J = 4.9$ Hz, 11.7 Hz), 4.49 (1H, t, $J = 4.9$ Hz), 5.30–5.40 (2H, m). HRFABMS m/z (positive-ion); Calcd for $C_{28}H_{54}O_2$ $(M)^{+\bullet}$: 422.4124. Found: 422.4116.

(*R*)-3-[(*Z*)-Tetradec-7-enyloxy]tetradecan-1-ol (3). To a solution of **2** (21.4 g, 48 mmol) in toluene was added a 1.0 M solution of DIBAL in toluene. After stirring 10 h at 50 °C, the reaction mixture was cooled to room temperature and sat. aq NH_4Cl (20 mL), sat. aq Rochelle salt (30 mL) and H_2O (100 mL) were slowly added. The resulting mixture was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (1:9) afforded **3** (16.5 g, 81%) as a colorless oil. $[\alpha]_D$ −19.7 (c 1.0, $CHCl_3$). IR (neat) 3375, 1466 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (6H, t, $J = 6.8$ Hz), 1.23–1.38 (32H, m), 1.42–1.50 (1H, m), 1.51–1.62 (3H, m), 1.65–1.73 (1H, m), 1.74–1.81 (1H, m), 1.98–2.05 (4H, m), 2.79 (1H, t, $J = 6.9$ Hz), 3.39 (1H, dt, $J = 7.8$ Hz, 8.8 Hz), 3.45–3.55 (2H, m), 3.70–3.84 (2H, m), 5.30–5.40 (2H, m). HRFABMS m/z (positive-ion); Calcd for $C_{28}H_{57}O_2$ $(M + H)^+$: 425.4359. Found: 425.4356.

(*R*)-3-[(*Z*)-Tetradec-7-enyloxy]tetradecanoic acid (4). To a solution of oxalyl chloride (0.83 mL, 9.6 mmol) in CH_2Cl_2 (10 mL) was slowly added DMSO (1.4 mL, 19 mmol) at −78 °C. After stirring for 10 min, to the reaction mixture was added a solution of **3** (2.0 g, 4.8 mmol) in CH_2Cl_2. After stirring for 1 h, to the reaction mixture was slowly added Et_3N (5.3 mL, 38 mmol) and the reaction mixture was stirred for 1 h at 0 °C. The mixture was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product. To a solution of this crude product in *t*-BuOH (4.5 mL) and H_2O (1.5 mL) were added 2-methyl-2-butene (2.6 mL, 24 mmol), NaH_2PO_4 (1.1 g, 7.2 mmol), and $NaClO_2$ (1.3 g, 14 mmol). After stirring for 3 h at room temperature, the reaction mixture was poured into 1 M aq HCl and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (1:9) afforded **4** (1.9 g, 88%) as a colorless oil. $[\alpha]_D$ −5.6 (c 1.1, $CHCl_3$). IR (neat) 1712 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (6H, t, $J = 6.8$ Hz), 1.22–1.40 (32H, m), 1.46–1.65 (4H, m), 1.94–2.06 (4H, m), 2.54 (2H, d, $J = 6.8$ Hz), 3.46–3.55 (2H, m), 3.69 (1H, quintet, $J = 5.9$ Hz), 5.30–5.40 (2H, m). HRFABMS m/z (positive-ion); Calcd for $C_{28}H_{55}O_3$ $(M + H)^+$: 439.4151. Found: 439.4131.

(*Z*)-Tetradec-7-enoic acid (5). To a solution of aldehyde **1** (1.6 g, 7.6 mmol) in *t*-BuOH (7.5 mL) and H_2O (2.5 mL) were added 2-methyl-2-butene (4.0 mL, 38 mmol), NaH_2PO_4 (1.8 g, 11.4 mmol), and $NaClO_2$ (2.6 g, 22.8 mmol). After stirring for 3 h at room temperature, the reaction mixture was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (1:9, and then 1:4) afforded **5** (1.6 g, 96%) as a colorless oil. IR (neat) 1710 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (3H, t, $J = 6.8$ Hz), 1.22–1.40 (12H, m), 1.60–1.69 (2H, m), 1.86–2.07 (4H, m), 2.35 (2H, t, $J = 7.8$ Hz), 5.32–5.39 (2H, m). HRFABMS m/z (positive-ion); Calcd for $C_{14}H_{26}O_2$ $(M)^{+\bullet}$: 226.1933. Found: 226.1923.

1-Methylsulfonyloxy-3-[(*R*)-[(*Z*)-tetradec-7-enyloxy]]tetradecane (6). To a solution of **3** (6.6 g, 15.5 mmol) in CH_2Cl_2 (20 mL) was added *N*,*N*-diisopropylethylamine (4.0 mL, 23.3 mmol) and MsCl (1.4 mL, 18.6 mmol). After stirring for 4 h at room temperature, the reaction mixture was added sat. aq $NaHCO_3$ (5 mL). The resulting mixture was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (1:9) afforded **6** (7.8 g, quantitatively) as a colorless oil. $[\alpha]_D$ −18.7 (c 0.2, $CHCl_3$). IR (neat) 1466, 1416, 1359, 1179 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (6H, t, $J = 7.8$ Hz), 1.22–1.39 (32H, m), 1.40–1.48 (1H, m), 1.50–1.58 (3H, m), 1.79–1.87 (1H, m), 1.98–2.05 (4H, m), 3.00 (3H, s), 3.35–3.43 (2H, m), 3.47 (1H, dt, $J = 6.8$ Hz, 8.8 Hz), 4.29–4.40 (2H, m), 5.30–5.40 (2H, m). HRFABMS m/z (positive-ion); Calcd for $C_{29}H_{59}O_4S$ $(M + H)^+$: 503.4134. Found: 503.4143.

2-Hydroxyethyl 2-Deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-4,6-*O*-isopropylidene-2-trifluoroacetamido-α-D-glucopyranoside (8). To a solution of **7** (6.6 g, 9.1 mmol) in acetone (15 mL) and H_2O (5 mL) were added $NaIO_4$ (7.8 g, 36.4 mmol) and a 2.5 wt% solution of OsO_4 in *t*-BuOH (0.5 mL). After stirring 1.5 h at room temperature, the reaction mixture was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford 7.0 g of a crude product. To a solution of this crude product in MeOH (10 mL) was slowly added $NaBH_4$ (0.41 g, 10.9 mmol) at 0 °C. After stirring for 0.5 h, the reaction mixture was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford 6.4 g of a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (1:9, and then 3:7) afforded **8** (3.2 g, 47%) as a colorless oil. $[\alpha]_D$ +24.4 (c 1.0, $CHCl_3$). IR (neat) 3435, 3309, 3084, 1720, 1561, 1466, 1380, 1371 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (6H, t, $J = 7.8$ Hz), 1.22–1.36 (36H, m), 1.41 (3H, s), 1.49–1.55 (5H, m, involving 3H, s, at δ 1.51), 1.61–1.70 (4H, m), 3.30–3.40 (3H, m), 3.52–3.59 (2H, m), 3.60–3.66 (1H, m), 3.66–3.90 (8H, m), 4.15 (1H, dt, $J = 9.8$ Hz, 3.9 Hz), 4.93 (1H, d, $J = 3.9$ Hz), 6.90 (1H, d, $J = 7.8$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{39}H_{73}F_3NO_8$ $(M + H)^+$: 740.5288. Found: 740.5269.

2-Hydroxyethyl 2-Amino-2-deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-4,6-*O*-isopropylidene-α-D-glucopyranoside (9). To a solution of **8** (3.2 g, 4.3 mmol) in MeOH (15 mL) and H_2O (2 mL) was added NaOH (0.34 g, 8.6 mmol). After stirring for 5 h at 50 °C, the reaction mixture was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to yield a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (3:7) afforded **9** (2.6 g, 93%) as a colorless oil. $[\alpha]_D$ +40.8 (c 1.2, $CHCl_3$). IR (neat) 3371, 3295, 1587, 1466, 1380, 1369, 1267 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (6H, t, $J = 6.8$ Hz), 1.23–1.38 (35H, m), 1.40 (3H, s), 1.42–1.57 (9H, m, involving a singlet at δ 1.49), 1.68–1.77 (4H, m), 2.77 (1H, dd, $J = 3.9$ Hz, 9.8 Hz), 3.29 (1H, t, $J = 8.8$ Hz), 3.33–3.45 (3H, m), 3.56 (1H, t, $J = 9.8$ Hz), 3.62–3.69 (2H, m), 3.69–3.74 (2H, m), 3.74–

3.86 (4H, m), 3.93 (1H, td, $J = 5.9$ Hz, 9.8 Hz), 4.92 (1H, d, $J = 3.9$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{37}H_{74}NO_7$ $(M + H)^+$: 644.5465. Found: 644.5466.

2-Hydroxyethyl 2-Deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-4,6-*O*-isopropylidene-2-[(*R*)-3-[(*Z*)-tetradec-7-enoyloxy]tetradecanamido]-α-D-glucopyranoside (10). To a solution of **9** (0.92 g, 1.4 mmol) and (*R*)-3-[(*Z*)-tetradec-7-enyloxy]tetradecanoic acid **4** (0.75 g, 1.7 mmol) in CH_2Cl_2 (3 mL) was added DCC (0.43 g, 2.1 mmol) at 0 °C. After stirring for 2.5 h, to the reaction mixture was added hexane (5 mL), and the mixture was filtered. The filtrate was concentrated in vacuo to give a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (1:4) afforded **10** (1.3 g, 84%) as a colorless oil. $[\alpha]_D$ +23.0 (c 0.9, $CHCl_3$). IR (neat) 3317, 1646, 1545, 1466 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (12H, t, $J = 6.8$ Hz), 1.22–1.66 (84H, m, involving 3H two singlets at δ 1.41 and δ 1.50), 1.67–1.75 (1H, m), 1.95–2.06 (4H, m), 2.34 (1H, dd, $J = 7.8$ Hz, 15.6 Hz), 2.41 (1H, dd, $J = 3.9$ Hz, 15.6 Hz), 3.31–3.39 (3H, m, involving a triplet at δ 3.37, $J = 6.8$ Hz), 3.42–3.49 (3H, m), 3.54–3.61 (2H, m), 3.66–3.86 (8H, m), 4.14–4.20 (1H, m), 4.88 (1H, d, $J = 3.9$ Hz), 5.30–5.40 (2H, m), 6.72 (1H, d, $J = 8.8$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{65}H_{126}NO_9$ $(M + H)^+$: 1064.9433. Found: 1064.9438.

(Allyloxycarbonyl)methyl 2-Deoxy-3-*O*-[(*R*)-3-(dodecyloxy)-tetradecyl]-4,6-*O*-isopropylidene-2-[(*R*)-3-[(*Z*)-tetradec-7-enoyloxy]tetradecanamido]-α-D-glucopyranoside (11). To a solution of **10** (1.2 g, 1.1 mmol) in CH_2Cl_2 (20 mL) was added Dess-Martin periodinane (1.2 g, 2.9 mmol) at 0 °C. After stirring for 3.5 h, to the reaction mixture were added sat. aq $NaHCO_3$ (1 mL) and sat. aq $Na_2S_2O_3$ (1 mL). The resulting mixture was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product. To a solution of this crude product in *t*-BuOH (3 mL) and H_2O (1 mL) were added 2-methyl-2-butene (0.5 mL, 4.6 mmol), NaH_2PO_4 (0.27 g, 1.7 mmol) and $NaClO_2$ (0.40 g, 3.5 mmol). After stirring for 1 h at room temperature, the reaction mixture was poured into water, and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product. Furthermore, to a solution of this crude product in DMF (2 mL) were added Et_3N (0.64 mL, 4.6 mmol) and allyl bromide (0.30 mL, 3.5 mmol). After stirring for 11 h at room temperature, sat. aq $NaHCO_3$ (1 mL) was added to the reaction mixture, which was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (1:9, and then 1:4) gave **11** (0.66 g, 51%) as a colorless oil. $[\alpha]_D$ +30.4 (c 0.3, $CHCl_3$). IR (neat) 3320, 1759, 1658, 1536, 1466, 1372 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (12H, t, $J = 6.8$ Hz), 1.21–1.60 (82H, m, involving 3H two singlets at δ 1.40 and 1.49, respectively), 1.68–1.75 (2H, m), 1.97–2.06 (4H, m), 2.39 (1H, dd, $J = 6.8$ Hz, 14.7 Hz), 2.44 (1H, dd, $J = 3.9$ Hz, 14.7 Hz), 3.30–3.51 (6H, m), 3.58 (1H, dt, $J = 9.8$ Hz, 6.8 Hz), 3.63–3.85 (6H, m), 4.16 (1H, d, $J = 3.9$ Hz), 5.27 (1H, d, $J = 8.8$ Hz), 5.30–5.40 (3H, m), 5.90 (1H, ddt, $J = 10.7$ Hz, 17.5 Hz, 5.9 Hz), 6.82 (1H, d, $J = 9.8$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{68}H_{128}NO_{10}$ $(M + H)^+$: 1118.9538. Found: 1118.9557.

(Allyloxycarbonyl)methyl 6-*O*-(*t*-Butyldimethylsilyl)-2-deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-2-[(*R*)-3-[(*Z*)-tetradec-7-enoyloxy]tetradecanamido]-α-D-glucopyranoside (12). To a solution of **11** (0.59 g, 0.53 mmol) in MeOH (3 mL) and THF (1 mL) was added CSA (45 mg, 0.19 mmol). After stirring for 1 h at room temperature, the reaction mixture was added Et_3N (1 mL) and evaporated in vacuo to give a crude diol. To a solution of this diol in DMF (1 mL) were added imidazole (72 mg, 1.1 mmol) and TBDMSCl (88 mg, 0.58 mmol). After stirring for 1 h at room temperature, sat. aq $NaHCO_3$ (1 mL) was added to the reaction mixture, which was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (1:9, and then 1:4) afforded **12** (0.62 g, 99%) as a colorless oil. $[\alpha]_D$ +26.3 (c 0.6, $CHCl_3$). IR (neat) 3353, 1758, 1655, 1535, 1465 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.08 (6H, s), 0.88 (12H, t, $J = 6.8$ Hz), 0.90 (9H, s), 1.22–1.60 (76H, m), 1.71 (2H, q, $J = 5.9$ Hz), 1.96–2.05 (4H, m), 2.37–2.45 (2H, m), 3.34–3.51 (6H, m), 3.56–3.71 (4H, m), 3.74–3.80 (1H, m), 3.82 (1H, dd, $J = 3.9$ Hz, 10.7 Hz), 3.85 (1H, dd, $J = 2.9$ Hz, 10.7 Hz), 4.18 (2H, s), 4.21 (1H, dt, $J = 3.9$ Hz, 9.8 Hz), 4.63 (2H, d, $J = 5.9$ Hz), 4.83 (1H, d, $J = 2.9$ Hz), 5.26 (1H, d, $J = 10.7$ Hz), 5.30–5.40 (3H, m), 5.90 (1H, ddt, $J = 17.5$, 10.7, 4.9 Hz), 6.81 (1H, d, $J = 9.8$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{71}H_{138}NO_{10}Si$ $(M + H)^+$: 1193.0090. Found: 1193.0084.

(Allyloxycarbonyl)methyl 6-*O*-*t*-Butyldimethylsilyl-2-deoxy-4-*O*-bis(allyloxy)phosphoryl-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-2-[(*R*)-3-[(*Z*)-tetradec-7-enoyloxy]tetradecanamido]-α-D-glucopyranoside (13). To a solution of **12** (0.62 g, 0.52 mmol) in THF (2 mL) were added diallyl diisopropylphosphoramidite (0.21 mL, 0.78 mmol) and 1*H*-tetrazole (73 mg, 1.0 mmol). After stirring for 3 h at room temperature, to the reaction mixture was added 40% aq H_2O_2 (0.3 mL). After further stirring for 1 h, sat. aq $Na_2S_2O_3$ (0.5 mL) was slowly added to the reaction mixture, which was poured into water and extracted with ether. The organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (1:9) afforded **13** (0.60 g, 85%) as a colorless oil. $[\alpha]_D$ +28.5 (c 0.7, $CHCl_3$). IR (neat) 3320, 1759, 1677, 1535, 1464 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.05 (6H, s), 0.88 (12H, t, $J = 6.8$ Hz), 0.89 (9H, s), 1.21–1.58 (74H, m), 1.66–1.80 (2H, m), 1.98–2.04 (4H, m), 2.39 (2H, d, $J = 5.9$ Hz), 3.24–3.36 (3H, m), 3.45 (2H, t, $J = 6.8$ Hz), 3.60–3.80 (7H, m), 3.95 (1H, d, $J = 9.8$ Hz), 4.15 (1H, d, $J = 16.6$ Hz), 4.19 (1H, d, $J = 16.6$ Hz), 4.23–4.31 (2H, m), 4.53–4.59 (4H, m), 4.63 (2H, d, $J = 5.9$ Hz), 4.84 (1H, d, $J = 3.9$ Hz), 5.28–5.40 (8H, m), 5.85–5.98 (3H, m), 6.84 (1H, d, $J = 9.8$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{77}H_{147}NO_{13}PSi$ $(M + H)^+$: 1353.0379. Found: 1353.0371.

(Allyloxycarbonyl)methyl 4-*O*-Bis(allyloxy)phosphoryl-2-deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-2-[(*R*)-3-[(*Z*)-tetradec-7-enoyloxy]tetradecanamido]-α-D-glucopyranoside (14). To a solution of **13** (0.60 g, 0.44 mmol) in acetone (1 mL) was added 5% aq H_2SO_4 (0.2 mL). After stirring for 5 h at room temperature, the reaction mixture was quenched with sat. aq $NaHCO_3$ (0.2 mL) and poured into water. After extraction with ether, the organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (3:7, and then 1:1) afforded **14** (0.42

g, 76%) as a colorless oil. $[\alpha]_D$ +19.9 (c 1.0, $CHCl_3$). IR (neat) 3319, 1757, 1655, 1541, 1465 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (12H, t, $J = 6.8$ Hz), 1.22–1.78 (78H, m), 1.96–2.06 (4H, m), 2.39 (2H, d, $J = 5.9$ Hz), 3.23–3.37 (3H, m), 3.45 (2H, t, $J = 6.8$ Hz), 3.56–3.64 (2H, m), 3.66–3.74 (2H, m), 3.74–3.83 (2H, m), 3.92–3.98 (2H, m), 4.15 (1H, d, $J = 16.6$ Hz), 4.20 (1H, d, $J = 16.6$ Hz), 4.29 (1H, td, $J = 3.9$ Hz, 10.7 Hz), 4.40 (1H, q, $J = 9.8$ Hz), 4.56 (2H, t, $J = 6.8$ Hz), 4.62–4.67 (4H, m), 4.85 (1H, d, $J = 3.9$ Hz), 5.24–5.41 (8H, m), 5.86–5.99 (3H, m), 6.81 (1H, d, $J = 9.8$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{71}H_{133}NO_{13}P$ $(M + H)^+$: 1238.9515. Found: 1238.9519.

(Allyloxycarbonyl)methyl 4-*O*-Bis(allyloxy)phosphoryl-2-deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-2-[(*R*)-3-[(*Z*)-tetradec-7-enoyloxy]tetradecanamido]-α-D-glucopyranoside (15). To a solution of **14** (120 mg, 0.094 mmol) and 2,6-di-*t*-butyl-4-methylpyridine (280 mg, 1.3 mmol) in CH_2Cl_2 (0.5 mL) was added Me_3OBF_4 (130 mg, 0.94 mmol). After stirring for 4 h at room temperature, the reaction mixture was quenched with sat. aq $NaHCO_3$ (0.5 mL) and poured into water. After extraction with ether, the organic layer was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to afford a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (3:7) afforded **15** (63 mg, 54%) as a colorless oil. $[\alpha]_D$ +27.0 (c 0.3, $CHCl_3$). IR (neat) 3321, 1757, 1675, 1537, 1465 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (12H, t, $J = 6.8$ Hz), 1.22–1.58 (78H, m), 1.66–1.80 (4H, m), 1.96–2.06 (4H, m), 2.38 (2H, d, $J = 5.9$ Hz), 3.24–3.30 (1H, m), 3.31–3.36 (2H, m), 3.39 (3H, s), 3.44 (2H, t, $J = 6.8$ Hz), 3.60–3.78 (6H, m), 3.89 (1H, dt, $J = 3.9$, 10.7 Hz), 4.20 (3H, s), 4.30 (1H, dt, $J = 3.9$, 10.7 Hz), 4.37 (1H, q, $J = 9.8$ Hz), 4.54–4.62 (m, 4H), 4.63 (2H, d, $J = 5.9$ Hz), 4.87 (1H, d, $J = 3.9$ Hz), 5.23–5.40 (8H, m), 5.85–5.99 (3H, m), 6.83 (1H, d, $J = 9.8$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{72}H_{135}NO_{13}P$ $(M + H)^+$: 1252.9671. Found: 1252.9677.

Carboxymethyl 2-Deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-4-*O*-phosphono-2-[(*R*)-3-[(*Z*)-tetradec-7-enoyloxy]tetradecanamido]-α-D-glucopyranoside (16). To a solution of **14** (94 mg, 0.076 mmol) and PPh_3 (10 mg, 0.038 mmol) in THF (0.3 mL) were added Et_3N (53 μL, 0.38 mmol), HCO_2H (29 μL, 0.76 mmol) and $[Pd(PPh_3)_4]$ (2 mg, 0.002 mmol). After stirring for 7 h at 50 °C, the reaction mixture was evaporated in vacuo to give a crude product, which was purified by DEAE-cellulose column chromatography. The column was eluted with $CHCl_3$–MeOH–0.1 M aq CH_3COONH_4 (2:1:0, and then 2:3:1). The product containing fractions were concentrated in vacuo to give a crude product, which was dissolved in $CHCl_3$ (4 mL), MeOH (8 mL) and 0.1 M aq HCl (3 mL). To this solution was added another volume of $CHCl_3$ (4 mL) and 0.1 M aq HCl (4 mL) to separate the solution into two phases. The lower $CHCl_3$ phase was collected and concentrated to give **16** (66 mg, 78%) as a white powder. $[\alpha]_D$ +25.6 (c 0.5, $CHCl_3$). IR ($CHCl_3$) 3634, 3340, 1754, 1663, 1521, 1466 cm^{-1}. ^1H NMR (500 MHz, CD_3OD) δ 0.90 (9H, t, $J = 6.8$ Hz), 1.25–1.57 (76H, m), 1.69–1.81 (2H, m), 1.98–2.08 (4H, m), 2.35 (1H, dd, $J = 5.9$ Hz, 14.7 Hz), 2.49 (1H, dd, $J = 14.7$ Hz, 6.8 Hz), 3.34–3.48 (4H, m), 3.50–3.56 (1H, m), 3.66 (2H, t, $J = 9.8$ Hz), 3.70–3.77 (2H, m), 3.77–3.83 (2H, m), 3.89 (1H, q, $J = 8.8$ Hz), 4.08 (1H, dd, $J = 3.9$ Hz, 10.7 Hz), 4.10–4.17 (2H, m, involving a doublet at δ 4.13, $J = 16.6$ Hz), 4.25 (1H, d, $J = 16.6$ Hz), 4.81 (1H, d, $J = 2.9$ Hz), 5.31–5.40 (2H, m). HRFABMS m/z (positive-ion); Calcd for $C_{62}H_{120}NO_{13}PNa$ $(M + Na)^+$: 1140.8395. Found: 1140.8383.

Carboxymethyl 2-Deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-6-*O*-methyl-4-*O*-phosphono-2-[(*R*)-3-[(*Z*)-tetradec-7-enoyloxy]tetradecanamido]-α-D-glucopyranoside (17). Compound **15** (63 mg, 0.050 mmol) was treated as described in the formation of **16** from **14** to give **17** (48 mg, 85%) as a white powder. $[\alpha]_D$ +33.1 (c 0.60, $CHCl_3$). IR ($CHCl_3$) 3691, 3607, 3341, 1736, 1671, 1603, 1525, 1467 cm^{-1}. ^1H NMR (500 MHz, CD_3OD) δ 0.90 (12H, t, $J = 6.8$ Hz), 1.24–1.58 (78H, m), 1.68–1.82 (2H, m), 2.00–2.07 (4H, m), 2.34 (1H, dd, $J = 4.9$ Hz, 14.7 Hz), 2.49 (1H, dd, $J = 14.7$ Hz, 6.8 Hz), 3.36–3.42 (5H, m, involving a singlet at δ 3.38), 3.44–3.48 (2H, m), 3.52 (1H, dt, $J = 5.9$ Hz, 11.7 Hz), 3.59 (1H, dd, $J = 5.9$ Hz, 10.7 Hz), 3.63–3.70 (2H, m), 3.70–3.77 (2H, m), 3.84–3.90 (2H, m), 4.08 (1H, dd, $J = 2.9$ Hz, 10.7 Hz), 4.12 (1H, d, $J = 16.6$ Hz), 4.14 (1H, q, $J = 9.8$ Hz), 4.24 (1H, d, $J = 16.6$ Hz), 4.79 (1H, d, $J = 2.9$ Hz), 5.31–5.40 (2H, m). HRFABMS m/z (positive-ion); Calcd for $C_{63}H_{122}NO_{13}PNa$ $(M + Na)^+$: 1154.8552. Found: 1154.8561.

2-Hydroxyethyl 2-Deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-4,6-*O*-isopropylidene-2-[(*Z*)-7-tetradecenamido]-α-D-glucopyranoside (18). To a solution of **9** (0.93 g, 1.4 mmol) in CH_2Cl_2 (3 mL) and (*Z*)-tetradec-7-enoic acid **5** (0.39 g, 1.7 mmol) was added DCC (0.45 g, 2.2 mmol) at 0 °C. After stirring for 4 h, the reaction mixture was diluted with hexane (5 mL) and filtered. The filtrate was concentrated in vacuo to give a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (1:4, and then 2:3) afforded **18** (1.0 g, 85%) as a colorless oil. $[\alpha]_D$ +29.1 (c 2.0, $CHCl_3$). IR (neat) 3306, 1644, 1544, 1465, 1379 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (9H, t, $J = 7.8$ Hz), 1.22–1.57 (58H, m, involving 3H, two singlets at δ 1.40 and δ 1.50, respectively), 1.60–1.73 (4H, m), 1.94–2.06 (4H, m), 2.19 (2H, t, $J = 7.8$ Hz), 3.34–3.43 (3H, m, involving a triplet at δ 3.36, $J = 5.9$ Hz), 3.44–3.49 (1H, m), 3.52–3.63 (2H, m), 3.65–3.87 (8H, m), 4.09–4.15 (1H, m), 4.97 (1H, d, $J = 2.9$ Hz), 5.29–5.39 (2H, m), 6.05 (1H, d, $J = 8.8$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{51}H_{97}NO_8$ $(M + H)^+$: 851.7214. Found: 852.7286.

(Allyloxycarbonyl)methyl 2-Deoxy-3-*O*-[(*R*)-3-(dodecyloxy)-tetradecyl]-4,6-*O*-isopropylidene-2-[(*Z*)-7-tetradecenamido]-α-D-glucopyranoside (19). Compound **18** (0.41 g, 0.48 mmol) was treated as described in the formation of **11** from **10** to afford **19** (0.18 g, 46%) as a colorless oil. $[\alpha]_D$ +32.0 (c 0.7, $CHCl_3$). IR (neat) 3306, 1759, 1650, 1544, 1465, 1379 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.88 (9H, t, $J = 6.8$ Hz), 1.21–1.46 (53H, m, involving a singlet at δ 1.40), 1.47–1.56 (5H, m, involving a singlet at δ 1.49), 1.58–1.72 (4H, m), 1.95–2.06 (4H, m), 2.22 (2H, dt, $J = 6.8$ Hz, 1.9 Hz), 3.32–3.39 (3H, m), 3.44–3.50 (1H, m), 3.54–3.60 (1H, m), 3.67–3.76 (3H, m), 3.76–3.84 (2H, m), 4.16 (1H, d, $J = 17.6$ Hz), 4.22 (1H, d, $J = 17.6$ Hz), 4.23 (1H, dt, $J = 10.7$ Hz, 3.9 Hz), 4.65 (2H, d, $J = 5.9$ Hz), 4.79 (1H, d, $J = 3.9$ Hz), 5.27 (1H, d, $J = 8.8$ Hz), 5.30–5.40 (3H, m), 5.91 (1H, ddt, $J = 10.7$ Hz, 17.5 Hz, 5.9 Hz), 6.09 (1H, d, $J = 8.8$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{54}H_{100}NO_9$ $(M + H)^+$: 906.7398. Found: 906.7397.

(Allyloxycarbonyl)methyl 6-*O*-(*t*-Butyldimethylsilyl)-2-deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-2-[(*Z*)-7-tetradecenamido]-α-D-glucopyranoside (20). Compound **19** (0.59 g, 0.65 mmol) was treated as described in the formation of **12** from **11** to afford a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (1:9, and then 1:4) afforded **20** (0.56 g, 87%) as a colorless oil. $[\alpha]_D$ +30.9 (c 0.5, $CHCl_3$). IR (neat) 3313, 1756, 1650, 1543, 1465 cm^{-1}.

^{1}H NMR (500 MHz, $CDCl_3$) δ 0.08 (6H, s), 0.88 (9H, t, $J = 6.8$ Hz), 0.90 (9H, s), 1.22–1.44 (48H, m), 1.49–1.75 (8H, m), 1.94–2.05 (4H, m), 2.20–2.27 (2H, m), 3.37 (2H, t, $J = 6.8$ Hz), 3.39–3.44 (1H, m), 3.48 (1H, dd, $J = 8.8$, 10.7 Hz), 3.60–3.78 (4H, m), 3.81–3.88 (2H, m), 4.17 (1H, d, $J = 16.6$ Hz), 4.20–4.25 (2H, m, involving 1H, d, $J = 16.6$ Hz, at δ 4.23), 4.64 (2H, d, $J = 5.9$ Hz), 4.78 (1H, d, $J = 3.9$ Hz), 5.27 (1H, d, $J = 10.7$ Hz), 5.30–5.40 (3H, m), 5.90 (1H, ddt, $J = 17.5$, 10.7, 4.9 Hz), 6.15 (1H, d, $J = 9.8$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{57}H_{110}NO_9Si$ $(M + H)^+$: 980.7950. Found: 980.7963.

(Allyloxycarbonyl)methyl 4-*O*-Bis(allyloxy)phosphoryl-6-*O*-(*t*-butyldimethylsilyl)-2-deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-2-[(*Z*)-7-tetradecenamido]-α-D-glucopyranoside (21). Compound **20** (0.55 g, 0.56 mmol) was treated as described in the formation of **13** from **12** to afford a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (1:9, and then 1:4) afforded **21** (0.52 g, 81%) as a colorless oil. $[\alpha]_D$ +31.5 (c 1.0, $CHCl_3$). IR (neat) 3308, 1757, 1660, 1541, 1464 cm^{-1}. ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.05 (6H, s), 0.88 (9H, t, $J = 6.8$ Hz), 0.89 (9H, s), 1.23–1.76 (56H, m), 1.95–2.05 (4H, m), 2.23 (2H, t, $J = 7.8$ Hz), 3.26–3.32 (1H, m), 3.33 (2H, t, $J = 6.8$ Hz), 3.59–3.66 (2H, m), 3.73–3.81 (3H, m), 3.94 (1H, d, $J = 9.8$ Hz), 4.16 (1H, d, $J = 17.6$ Hz), 4.21–4.30 (2H, m, involving 1H, d, $J = 17.6$ Hz, at δ 4.24), 4.53–4.59 (4H, m), 4.64 (2H, d, $J = 5.9$ Hz), 4.79 (1H, d, $J = 3.9$ Hz), 5.20–5.39 (8H, m), 5.86–5.99 (3H, m), 6.15 (1H, d, $J = 9.8$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{63}H_{119}NO_{12}PSi$ $(M + H)^+$: 1140.8239. Found: 1140.8242.

(Allyloxycarbonyl)methyl 4-*O*-Bis(allyloxy)phosphoryl-2-deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-2-[(*Z*)-7-tetradecenamido]-α-D-glucopyranoside (22). Compound **21** (0.52 g, 0.46 mmol) was treated as described in the formation of **14** from **13** to afford a crude product, which was purified by silica-gel column chromatography. Elution with EtOAc–hexane (3:7, and then 1:1) afforded **22** (0.38 g, 80%) as a gum. $[\alpha]_D$ +34.7 (c 0.6, $CHCl_3$). IR (neat) 3312, 1757, 1654, 1543, 1465 cm^{-1}. ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.88 (9H, t, $J = 6.8$ Hz), 1.22–1.55 (52H, m), 1.62–1.73 (4H, m), 1.97–2.05 (4H, m), 2.23 (2H, t, $J = 6.8$ Hz), 3.27–3.38 (3H, m), 3.59 (2H, t, $J = 9.8$ Hz), 3.67–3.76 (2H, m), 3.82 (1H, q, $J = 8.8$ Hz), 4.40 (1H, q, $J = 9.8$ Hz), 4.56 (2H, dd, $J = 4.9$, 7.8 Hz), 4.61–4.67 (4H, m), 4.82 (1H, d, $J = 3.9$ Hz), 5.24–5.41 (8H, m), 5.86–5.98 (3H, m), 6.18 (1H, d, $J = 8.8$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{57}H_{105}NO_{12}P$ $(M + H)^+$: 1026.7374. Found: 1026.7362.

(Allyloxycarbonyl)methyl 4-*O*-Bis(allyloxy)phosphoryl-2-deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-6-*O*-methyl-2-[(*Z*)-7-tetradecenamido]-α-D-glucopyranoside (23). Compound **22** (120 mg, 0.12 mmol) was treated as described in the formation of **15** from **14** to afford **23** (67 mg, 55%) as a colorless oil. $[\alpha]_D$ +34.4 (c 0.77, $CHCl_3$). IR (neat) 3310, 1756, 1653, 1542, 1465 cm^{-1}. ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.88 (9H, t, $J = 6.8$ Hz), 1.22–1.54 (52H, m), 1.62–1.76 (4H, m), 1.97–2.05 (4H, m), 2.23 (2H, t, $J = 7.8$ Hz), 3.26–3.32 (1H, m), 3.33 (2H, t, $J = 5.9$ Hz), 3.39 (3H, s), 3.63 (2H, t, $J = 8.8$ Hz), 3.66 (2H, d, $J = 3.9$ Hz), 3.74–3.81 (1H, m), 3.88 (1H, dt, $J = 3.9$, 10.7 Hz), 4.36 (1H, q, $J = 9.8$ Hz), 4.54–4.62 (4H, m), 4.67 (2H, d, $J = 5.9$ Hz), 4.82 (1H, d, $J = 3.9$ Hz), 5.23–5.41 (8H, m), 5.86–5.99 (3H, m), 6.15 (1H, d, $J = 8.8$ Hz). HRFABMS m/z (positive-ion); Calcd for $C_{58}H_{107}NO_{12}P$ $(M + H)^+$: 1040.7530. Found: 1040.7537.

Carboxymethyl 2-Deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-4-*O*-phosphono-2-[(*Z*)-7-tetradecenamido]-α-D-glucopyranoside (24). Compound **22** (74 mg, 0.072 mmol) was treated as described in the formation of **16** from **14** to give **24** (26 mg, 40%) as a white powder. $[\alpha]_D$ +30.3 (c 0.3, CH_3OH). IR ($CHCl_3$) 3691, 3607, 1734, 1654, 1603, 1466 cm^{-1}. ^{1}H NMR (500 MHz, CD_3OD) δ 0.90 (9H, t, $J = 6.8$ Hz), 1.23–1.48 (51H, m), 1.50–1.57 (2H, m), 1.60–1.69 (2H, m), 1.70–1.77 (2H, m), 2.00–2.10 (4H, m), 2.25 (2H, dt, $J = 7.8$ Hz, 2.0 Hz), 3.35–3.42 (2H, m), 3.43–3.48 (1H, m), 3.62–3.69 (2H, m), 3.70–3.76 (1H, m), 3.76–3.84 (2H, m), 3.87–3.93 (1H, m), 4.07 (1H, dd, $J = 2.9$, 9.8 Hz), 4.10–4.18 (2H, m, containing 1H, d, $J = 16.6$ Hz, at δ 4.14), 4.25 (1H, d, $J = 16.6$ Hz), 4.80 (1H, d, $J = 2.9$ Hz), 5.31–5.42 (2H, m). HRFABMS m/z (positive-ion); Calcd for $C_{48}H_{92}NO_{12}PNa$ $(M + Na)^+$: 928.6255. Found: 928.6250.

Carboxymethyl 2-Deoxy-3-*O*-[(*R*)-3-(dodecyloxy)tetradecyl]-6-*O*-methyl-4-*O*-phosphono-2-[(*Z*)-7-tetradecenamido]-α-D-glucopyranoside (25). Compound **23** (65 mg, 0.062 mmol) was treated as described in the formation of **16** from **14** to give **25** (49 mg, 86%) as a white powder. $[\alpha]_D$ +34.5 (c 0.3, CH_3OH). IR ($CHCl_3$) 3690, 3438, 1752, 1676, 1604, 1511, 1466 cm^{-1}. ^{1}H NMR (500 MHz, CD_3OD) δ 0.90 (9H, t, $J = 6.8$ Hz), 1.24–1.50 (52H, m), 1.50–1.57 (2H, m), 1.60–1.68 (2H, m), 1.70–1.80 (2H, m), 2.00–2.08 (4H, m), 2.25 (2H, dt, $J = 8.8$, 2.0 Hz), 3.35–3.43 (5H, m, containing 3H, s, at δ 3.38), 3.45 (1H, dt, $J = 5.9$, 9.8 Hz), 3.59 (1H, dd, $J = 5.9$, 10.7 Hz), 3.63–3.69 (2H, m), 3.74 (1H, d, $J = 8.8$ Hz), 3.83–3.90 (2H, m), 4.07 (1H, dd, $J = 3.9$, 10.7 Hz), 4.11–4.17 (2H, m, containing 1H, d, $J = 16.6$ Hz, at δ 4.13), 4.25 (1H, d, $J = 16.6$ Hz), 4.78 (1H, d, $J = 2.9$ Hz), 5.31–5.41 (2H, m). HRFABMS m/z (positive-ion); Calcd for $C_{49}H_{94}NO_{12}PNa$ $(M + Na)^+$: 942.6411. Found: 942.6399.

2-Hydroxyethyl 2-Deoxy-4,6-*O*-isopropylidene-2-trifluoroacetamido-α-D-glucopyranoside (27). Compound **26** (355 mg, 1.00 mmol) was treated as described in the formation of **8** from **7** to give **27** (252 mg, 2 steps 70%) as a white solid. IR (KBr) 3435, 3084, 2995, 2941, 2923, 1719 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.44 (3H, s), 1.53 (3H, s), 1.84 (1H, brs, OH), 3.30 (1H, brs, OH), 3.56–3.90 (9H, m), 4.19 (1H, dt, $J = 3.7$, 9.5 Hz), 4.90 (1H, d, $J = 3.7$ Hz), 7.46 (1H, d, $J = 8.1$ Hz, NH). FABMS (positive-ion); m/z 382 $(M + Na)^+$, 360 $(M + H)^+$. FABMS (negative-ion) m/z 686 $(M - H)^-$. HRFABMS m/z (positive-ion); Calcd for $C_{13}H_{21}F_3NO_7$: 360.1270. Found: 360.1274. Anal. Calcd for $C_{13}H_{20}F_3NO_7$ (359.3): C, 43.46; H, 5.61; N, 3.90; F, 15.86%. Found: C, 43.38; H, 5.53; N, 3.85; F, 15.88%.

2-(4-Methoxybenzyloxy)ethyl 2-Deoxy-4,6-*O*-isopropylidene-2-trifluoroacetamido-α-D-glucopyranoside (28). To a solution of **27** (1.80 g, 5.01 mmol) in DMF (20 mL) was gradually added NaH (60% oil dispersion, 245 mg, 6.13 mmol) at 0 °C with stirring. After 15 min, 4-methoxybenzyl chloride (0.73 mL, 5.24 mmol) was added to this solution, which was stirred for 5 h at room temperature. The reaction mixture was quenched with cold water, extracted with EtOAc, washed with water and brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to give a mixture that was chromatographed on a silica-gel column. Elution with hexane–EtOAc (2:3) gave **28** (2.08 g, 87%). IR ($CHCl_3$) 3600, 3426, 2938, 2917, 2882, 1731, 1612 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.43 (3H, s), 1.51 (3H, s), 3.63–3.85 (8H, m, containing 3H, s, at δ 3.81), 4.18 (1H, td, $J = 3.7$, 5.9 Hz), 4.46, 4.50 (2H, AB-q, $J = 11.7$ Hz), 4.86 (1H, d, $J = 3.7$ Hz), 6.89 (2H, d, $J = 8.8$ Hz), 7.25 (2H, d, $J = 8.8$ Hz). FABMS (positive-ion); m/z 502 $(M + Na)^+$, 480 $(M + H)^+$. HRFABMS m/z (positive-ion); Calcd for $C_{21}H_{28}F_3NO_8Na$: 502.1665. Found: 502.1677.

2-(4-Methoxybenzyloxy)ethyl 2-Deoxy-4,6-*O*-isopropylidene-3-*O*-[(*R*)-3-[(*Z*)-tetradec-7-enyloxy]tetradecyl]-2-trifluoroacetamido-α-D-glucopyranoside (29). To a solution of **28** (1.35 g, 2.82 mmol) in DMF (10 mL) was gradually added NaH (60% oil dispersion, 285 mg, 7.13 mmol) at 0 °C with stirring. After 15 min, **6** (1.43 g, 2.84 mmol) was added to this solution, which was stirred for 3 h at room temperature. The reaction mixture was quenched with water, extracted with EtOAc, washed with water and brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to give a mixture that was chromatographed on a silica-gel column. Elution with hexane–EtOAc (3:1) gave **29** (2.12 g, 85%). IR ($CHCl_3$) 3428, 2929, 2856, 1733, 1613 cm^{-1}. ^1H NMR (400 MHz, $CDCl_3$) δ 0.88 (6H, t, $J = 6.6$–7.3 Hz), 1.26–1.32 (36H, m), 1.41 (3H, s), 1.50 (3H, s), 1.61–1.71 (2H, m), 1.97–2.02 (4H, m), 3.27–3.41 (3H, m), 3.49–3.86 (14H, m, containing 3H, s, at δ 3.81), 4.17 (1H, dt, $J = 3.7$, 9.5 Hz), 4.46, 4.49 (2H, AB-q, $J = 11.7$ Hz), 4.86 (1H, d, $J = 3.7$ Hz), 5.31–5.38 (2H, m), 6.83 (1H, d, $J = 9.5$ Hz, NH), 6.89 (2H, d, $J = 8.8$ Hz), 7.25 (2H, d, $J = 8.8$ Hz). FABMS (positive-ion); m/z 908 $(M + Na)^+$, 884 $(M - H)^+$. HRFABMS m/z (positive-ion); Calcd for $C_{49}H_{82}F_3NO_9Na$: 908.5839. Found: 908.5836. Anal. Calcd for $C_{49}H_{82}F_3NO_9$ (886.2): C, 66.41; H, 9.33; N, 1.58; F, 6.43%. Found: C, 65.19; H, 9.10; N, 1.71; F, 6.08%.

2-(4-Methoxybenzyloxy)ethyl 2-Deoxy-2-(2,2-difluorotetradecanamido)-4,6-*O*-isopropylidene-3-*O*-[(*R*)-3-[(*Z*)-tetradec-7-enyloxy]tetradecyl]-α-D-glucopyranoside (30). A solution of **29** (1.90 g, 2.14 mmol) in EtOH (10 mL) and aq 1 M NaOH (10 mL) was stirred at 60 °C for 4 h. The solution was concentrated in vacuo, diluted with EtOAc, washed with water and brine, dried over Na_2SO_4, filtered, and the filtrate was concentrated in vacuo to give an amine, which was dissolved in CH_2Cl_2 (15 mL). 2,2-Difluorotetradecanoic acid (681 mg, 2.57 mmol), DCC (535 mg, 2.59 mmol), and DMAP (316 mg, 2.58 mmol) were added to this solution, which was stirred for 1 h at room temperature, filtered, and concentrated in vacuo to give a mixture. The mixture was chromatographed on silica-gel column. Elution with hexane–EtOAc (4:1) gave **30** (2.13 g, 96%) as a gum. IR ($CHCl_3$) 3438, 2928, 2856, 1707, 1613 cm^{-1}. ^1H NMR (400 MHz, $CDCl_3$) δ 0.88 (9H, t, $J = 6.6$ Hz), 1.26–1.32 (56H, m), 1.41 (3H, s), 1.50 (3H, s), 1.63–1.73 (2H, m), 2.01–2.11 (6H, m), 3.29–3.41 (3H, m), 3.48–3.90 (14H, m, containing 3H, s, at δ 3.81), 4.17 (1H, dt, $J = 3.7$, 9.5 Hz), 4.46–4.50 (2H, AB-q, $J = 11.7$ Hz), 4.83 (1H, d, $J = 3.7$ Hz), 5.31–5.38 (2H, m), 6.63 (1H, d, $J = 9.5$ Hz, NH), 6.88 (2H, d, $J = 8.8$ Hz), 7.26 (2H, d, $J = 8.8$ Hz). FABMS (positive-ion); m/z 1058 $(M + Na)^+$. HRFABMS m/z (positive-ion); Calcd for $C_{61}H_{107}F_2NO_9Na$: 1058.7812. Found: 1058.7805. Anal. Calcd for $C_{61}H_{107}F_2NO_9$ (1036.5): C, 70.69; H, 10.41; N, 1.35; F, 3.67%. Found: C, 69.92; H, 10.18; N, 1.26; F, 3.59%.

2-Hydroxyethyl 2-Deoxy-2-(2,2-difluorotetradecanamido)-4,6-*O*-isopropylidene-3-*O*-[(*R*)-3-[(*Z*)-tetradec-7-enyloxy]tetradecyl]-α-D-glucopyranoside (31). To a solution of **30** (1.65 g, 1.59 mmol) in CH_2Cl_2 (10 mL) and H_2O (1 mL) was added DDQ (435 mg, 1.92 mmol) at room temperature. After stirring for 2 h, the reaction mixture was diluted with EtOAc, washed with sat. aq $NaHCO_3$ and brine, dried over $MgSO_4$, filtered, and concentrated in vacuo to give a crude product, which was chromatographed on a silica-gel column. Elution with hexane–EtOAc (3:1) gave **31** (1.22 g, 84%) as a gum. IR ($CHCl_3$) 3626, 3440, 2928, 2856, 1707 cm^{-1}. ^1H NMR (400 MHz, $CDCl_3$) δ 0.88 (9H, t, $J = 6.6$ Hz), 1.41 (3H, s), 1.50 (3H, s), 1.26–1.54 (56H, m), 1.59–1.73 (2H, m), 1.99–2.14 (6H, m), 3.29–3.41 (3H, m), 3.49–3.65 (3H, m), 3.68–3.86 (8H, m), 4.14 (1H, dt, $J = 3.7$, 9.5 Hz), 4.91 (1H, d, $J = 3.7$ Hz), 5.31–5.39 (2H, m), 6.62 (1H, d, $J = 8.8$ Hz, NH). FABMS (positive-ion); m/z 938 $(M + Na)^+$, 916 $(M + H)^+$. HRFABMS m/z (positive-ion); Calcd for $C_{53}H_{99}F_2NO_8Na$: 938.7236. Found: 938.7238.

(Allyloxycarbonyl)methyl 2-Deoxy-2-(2,2-difluorotetradecanamido)-4,6-*O*-isopropylidene-3-*O*-[(*R*)-3-[(*Z*)-tetradec-7-enyloxy]tetradecyl]-α-D-glucopyranoside (32). Compound **31** (974 mg, 1.06 mmol) was treated as described in the formation of **11** from **10** to give **32** (731 mg, 71%, 3 steps) as a gum. IR ($CHCl_3$) 3435, 2928, 2856, 1755, 1708 cm^{-1}. ^1H NMR (400 MHz, $CDCl_3$) δ 0.88 (9H, t, $J = 6.6$ Hz), 1.26–1.32 (56H, m), 1.40 (3H, s), 1.50 (3H, s), 1.60–1.72 (2H, m), 2.01–2.16 (6H, m), 3.28–3.41 (3H, m), 3.51–3.59 (2H, m), 3.68–3.84 (5H, m), 4.20 (2H, s), 4.21 (1H, dt, $J = 3.7$, 9.5 Hz), 4.64–4.66 (2H, m), 4.84 (1H, d, $J = 3.7$ Hz), 5.26–5.38 (4H, m), 5.90 (1H, m), 6.83 (1H, d, $J = 8.8$ Hz, NH). FABMS (positive-ion); m/z 992 $(M + Na)^+$, 970 $(M + H)^+$. HRFABMS m/z (positive-ion); Calcd for $C_{56}H_{101}F_2NO_9Na$: 992.7342. Found: 992.7349. Anal. Calcd for $C_{56}H_{101}F_2NO_9$ (970.4): C, 69.31; H, 10.49; N, 1.44; F, 3.92%. Found: C, 69.59; H, 10.50; N, 1.52; F, 3.78%.

(Allyloxycarbonyl)methyl 2-Deoxy-2-(2,2-difluorotetradecanamido)-3-*O*-[(*R*)-3-[(*Z*)-tetradec-7-enyloxy]tetradecyl]-α-D-glucopyranoside (33). A solution of **32** (673 mg, 0.694 mmol) in 80% AcOH aq (10 mL) was stirred at 60 °C for 3 h. The solution was diluted with EtOAc, washed with sat. aq $NaHCO_3$ and brine, dried over $MgSO_4$, filtered, concentrated in vacuo, and chromatographed on a silica-gel column. Elution with hexane–EtOAc (1:1) gave **33** (600 mg, 93%) as a white powder. IR ($CHCl_3$) 3605, 3431, 2928, 2856, 1754, 1707 cm^{-1}. ^1H NMR (400 MHz, $CDCl_3$) δ 0.88 (9H, t, $J = 6.6$ Hz), 1.26–1.73 (58H, m), 2.01–2.17 (7H, m, containing OH), 3.34–3.42 (3H, m), 3.55 (1H, t, $J = 8.8$, 10.3 Hz), 3.62–3.68 (2H, m), 3.74–3.88 (5H, m, containing OH), 4.20 (1H, dt, $J = 3.7$, 9.5 Hz), 4.23 (2H, s), 4.64–4.66 (2H, m), 4.84 (1H, d, $J = 3.7$ Hz), 5.26–5.39 (4H, m), 5.90 (1H, m), 6.92 (1H, d, $J = 9.5$ Hz, NH). FABMS (positive-ion); m/z 952 $(M + Na)^+$, 930 $(M + H)^+$. HRFABMS m/z (positive-ion); Calcd for $C_{53}H_{97}F_2NO_9Na$: 952.7029. Found: 952.7070. Anal. Calcd for $C_{53}H_{97}F_2NO_9$ (930.3): C, 68.42; H, 10.51; N, 1.51; F, 4.08%. Found: C, 68.22; H, 10.36; N, 1.54; F, 4.29%.

(Allyloxycarbonyl)methyl 4-*O*-Bis(allyloxy)phosphoryl-6-*O*-(*t*-butyldimethylsilyl)-2-deoxy-2-(2,2-difluorotetradecanamido)-3-*O*-[(*R*)-3-[(*Z*)-tetradec-7-enyloxy]tetradecyl]-α-D-glucopyranoside (34). To a solution of **33** (553 mg, 0.594 mmol) in CH_2Cl_2 (10 mL) were added DMAP (96 mg, 0.784 mmol) and *t*-butyldimethylsilyl chloride (108 mg, 0.717 mmol). After stirring for 3 h at room temperature, the mixture was diluted with CH_2Cl_2, washed with sat. aq $NaHCO_3$ and brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated in vacuo to give 6-*O* silylated product, which was dissolved in THF (10 mL). To this solution were added 1*H*-tetrazole (65 mg, 0.925 mmol) and diallyl diisopropylphosphoramidite (221 mg, 0.901 mmol). After stirring for 3 h at room temperature, 30% H_2O_2 (3 mL) was added to the reaction mixture at 0 °C. After stirring for 1 h at room temperature, the mixture was quenched with sat. aq $Na_2S_2O_3$, extracted with EtOAc, washed with water, sat. aq $NaHCO_3$ and brine, dried over $MgSO_4$, filtered, and concentrated in vacuo to give a crude product, which was chromatographed on a silica-gel column. Elution with hexane–EtOAc (4:1) gave **34** (600 mg, 84%) as a gum. IR ($CHCl_3$) 3431, 2928, 2856, 1754, 1709 cm^{-1}. ^1H NMR (500 MHz, $CDCl_3$) δ 0.06 (6H, s), 0.86–

0.89 (18H, m), 1.19–1.53 (56H, m), 1.67–1.79 (2H, m), 1.99–2.16 (6H, m), 3.23 (1H, m), 3.33 (2H, t, $J = 6.7$ Hz), 3.60 (1H, m), 3.69 (1H, dd, $J = 9.5$, 9.9 Hz), 3.77–3.84 (3H, m), 3.95 (1H, m), 4.18–4.24 (3H, m, containing 2H, s, at δ 4.21), 4.29 (1H, m), 4.53–4.62 (4H, m), 4.65 (2H, d, $J = 5.9$ Hz), 4.84 (1H, d, $J = 3.6$ Hz), 5.23–5.38 (8H, m), 5.86–5.97 (3H, m), 6.85 (1H, d, $J = 9.4$ Hz, NH). FABMS (positive-ion); m/z 1226 $(M + Na)^+$, 1204 $(M + H)^+$. HRFABMS m/z (positive-ion); Calcd for $C_{65}H_{120}F_2NO_{12}PSiNa$: 1226.8183. Found: 1226.8236. Anal. Calcd for $C_{65}H_{120}F_2NO_{12}PSi$ (1204.7): C, 64.80; H, 10.04; N, 1.16; F, 3.15; P, 2.57%. Found: C, 63.96; H, 9.89; N, 1.24; F, 3.05; P, 2.44%.

(Allyloxycarbonyl)methyl 4-*O*-Bis(allyloxy)phosphoryl-2-deoxy-2-(2,2-difluorotetradecanamido)-3-*O*-[(*R*)-3-[(*Z*)-tetradec-7-enyloxy]tetradecyl]-α-D-glucopyranoside (35). Compound **34** (542 mg, 0.450 mmol) was treated as described in the formation of **14** from **13** to give **35** (448 mg, 91%) as a gum. IR ($CHCl_3$) 3432, 2927, 2855, 1754, 1711 cm^{-1}. ^1H NMR (400 MHz, $CDCl_3$) δ 0.88 (9H, t, $J = 6.6$ Hz), 1.16–1.51 (56H, m), 1.67–1.77 (2H, m), 2.01–2.16 (6H, m), 3.22 (1H, m), 3.27–3.38 (2H, m), 3.48–3.78 (4H, m), 3.84 (1H, m), 3.96–4.12 (2H, m, containing OH), 4.19, 4.24 (2H, AB-q, $J = 16.8$ Hz), 4.25 (1H, td, $J = 3.7$, 9.5 Hz), 4.42 (1H, q, $J = 9.5$ Hz), 4.54–4.59 (2H, m), 4.63–4.67 (4H, m), 4.86 (1H, d, $J = 3.7$ Hz), 5.26–5.42 (8H, m), 5.83–5.99 (3H, m), 6.87 (1H, d, $J = 9.5$ Hz, NH). FABMS (positive-ion); m/z 1112 $(M + Na)^+$, 1090 $(M + H)^+$. HRFABMS m/z (positive-ion); Calcd. for $C_{59}H_{106}F_2NO_{12}PNa$: 1112.7318. Found: 1112.7338.

(Allyloxycarbonyl)methyl 4-*O*-Bis(allyloxy)phosphoryl-2-deoxy-2-(2,2-difluorotetradecanamido)-6-*O*-methyl-3-*O*-[(*R*)-3-[(*Z*)-tetradec-7-enyloxy]tetradecyl]-α-D-glucopyranoside (36). Compound **35** (304 mg, 0.279 mmol) was treated as described in the formation of **15** from **14** to give **36** (237 mg, 77%). IR ($CHCl_3$) 3432, 2928, 2856, 1753, 1711 cm^{-1}. ^1H NMR (400 MHz, $CDCl_3$) δ 0.88 (9H, t, $J = 6.6$ Hz), 1.25–1.51 (56H, m), 1.68–1.75 (2H, m), 1.99–2.17 (6H, m), 3.23 (1H, m), 3.33 (2H, t, $J = 6.6$ Hz), 3.40 (3H, s), 3.60 (1H, m), 3.66–3.70 (3H, m), 3.81 (1H, m), 3.89–3.93 (1H, m), 4.23 (2H, s), 4.25 (1H, td, $J = 3.7$, 9.5 Hz), 4.39 (1H, q, $J = 9.5$ Hz), 4.55–4.65 (6H, m), 4.87 (1H, d, $J = 3.7$ Hz), 5.24–5.40 (8H, m), 5.83–6.00 (3H, m), 6.84 (1H, d, $J = 9.5$ Hz, NH). FABMS (positive-ion); m/z 1126 $(M + Na)^+$, 1104 $(M + H)^+$. HRFABMS m/z (positive-ion); Calcd for $C_{60}H_{108}F_2NO_{12}PNa$: 1126.7475. Found: 1126.7517.

Carboxymethyl 2-Deoxy-2-(2,2-difluorotetradecanamido)-4-*O*-phosphono-3-*O*-[(*R*)-3-[(*Z*)-tetradec-7-enyloxy]tetradecyl]-α-D-glucopyranoside (37). Compound **35** (101 mg, 0.093 mmol) was treated as described in the formation of **16** from **14** to give **37** (76.5 mg, 85%) as a white powder. $[\alpha]_D^{24}$ +26.5 (c 0.7, $CHCl_3$). IR (KBr) 3314, 3129, 2924, 2853, 1686 cm^{-1}. ^1H NMR (500 MHz, CD_3OD) δ 0.90 (9H, t, $J = 6.8$ Hz), 1.29–1.53 (56H, m), 1.71–1.79 (2H, m), 2.03–2.15 (6H, m), 3.36–3.40 (2H, m), 3.47 (1H, m), 3.62–3.65 (3H, m), 3.78 (1H, t, $J = 9.8$ Hz), 4.00–4.08 (4H, m, containing 1H, d, $J = 15.6$ Hz, at δ 4.04), 4.13 (1H, q, $J = 9.8$ Hz), 4.22 (1H, d, $J = 15.6$ Hz), 4.82 (1H, d, $J = 3.9$ Hz), 5.31–5.38 (2H, m). FABMS (positive-ion); m/z 992 $(M + Na)^+$, 970 $(M + H)^+$. HRFABMS m/z (positive-ion); Calcd for $C_{50}H_{94}F_2NO_{12}PNa$: 992.6379. Found: 992.6381.

Carboxymethyl 2-Deoxy-2-(2,2-difluorotetradecanamido)-6-*O*-methyl-4-*O*-phosphono-3-*O*-[(*R*)-3-[(*Z*)-tetradec-7-enyloxy]tetradecyl]-α-D-glucopyranoside (38). Compound **36** (163 mg, 0.148 mmol) was treated as described in the formation of **16** from **14** to give **38** (134 mg, 92%) as a white powder. $[\alpha]_D^{24}$ +44.7 (c 0.5, $CHCl_3$). IR (KBr) 3314, 2925, 2853, 1758, 1685 cm^{-1}. ^1H NMR (500 MHz, CD_3OD) δ 0.90 (9H, t, $J = 6.9$ Hz), 1.29–1.54 (56H, m), 1.71–1.80 (2H, m), 1.99–2.12 (6H, m), 3.33–3.40 (5H, m, containing 3H, s, at δ 3.38), 3.45 (1H, m), 3.60 (1H, dd, $J = 5.9$, 10.8 Hz), 3.66 (1H, m), 3.75 (1H, m), 3.79–3.92 (3H, m), 4.08–4.19 (3H, m, containing 1H, d, $J = 16.6$ Hz, at δ 4.12), 4.28 (1H, d, $J = 16.6$ Hz), 4.84 (1H, d, $J = 3.5$ Hz), 5.31–5.39 (2H, m). FABMS (positive-ion); m/z 1006 $(M + Na)^+$, 984 $(M + H)^+$. HRFABMS m/z (positive-ion); Calcd for $C_{51}H_{96}F_2NO_{12}PNa$: 1006.6536. Found: 1006.6558.

Methods for Measurement of Biological Activity. The sources of the materials used in the study are as follows: lipopolysaccharide (LPS) from *E. coli* serotype 026:B6 and 12-*O*-tetradecanoylphorbol acetate (TPA) were from Sigma, St. Louis, MO; RPMI-1640 medium, fetal bovine serum (FBS), and newborn calf serum (NBCS) were from Gibco, Grand Island, NY; and human TNFα ELISA kit and mouse TNFα ELISA kit were from Genzyme, Techne, Mineapolis, MN.

Cell Culture: Human monoblastic U937 cells were maintained in RPMI-1640 medium supplemented with 10% FBS, 100 U/mL of penicillin and 100 μg/mL of streptomycin (growth medium).

Production of TNFα by U937 cells: U937 cells (1×10^4/200 μL/well) were plated in 96-well plates (Corning, Cambridge, MA), and were cultured in the presence of TPA (30 ng/mL) for 72 h at 37 °C. After removing the supernatant, the cells were incubated in 200 μL of fresh RPMI-1640 medium containing 10% NBCS, in the absence or the presence of 30 ng/mL of LPS with graded concentrations of the compounds in the humidified atmosphere of 5% CO_2 for 4.5 h at 37 °C. After incubation, the amount of TNFα produced in the culture supernatants was determined using the TNFα ELISA kits. As a control, the amount of TNFα produced by U937 cells, which were stimulated with 30 ng/mL of LPS in the absence of compounds, was used. The concentrations (nM) of the compounds required to inhibit the LPS-induced TNFα production by U937 cells by 50% (IC_{50}) was calculated from the control amount. All experiments were carried out at least twice, showing that the data are reproducible.

Production of TNFα by mouse peritoneal macrophage: C57BL/6 female mice (6–7 wk old) were obtained from Charls River Japan, Inc., Yokohama, Japan. Peritoneal resident macrophages were collected by peritoneal lavage with ice-cold saline. After washing, cells were resuspended in RPMI-1640 medium supplemented with 10% NBCS, 100 U/mL of penicillin and 100 μg/mL of streptomycin, and were plated in 96-well plates (5×10^4/100 μL/well). After incubation overnight at 37 °C, nonadherent cells were removed by washing three times with RPMI-1640 medium containing 10% NBCS, and adherent cells were incubated in 100 μL of the same medium, in the absence or presence of 10 ng/mL of LPS with graded concentrations of the compounds in the humidified atmosphere of 5% CO_2 for 4.5 h at 37 °C. After incubation, the amount of TNFα produced in the culture supernatants was determined using the mouse TNFα ELISA kits. IC_{50} of the compounds was calculated as described above.

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