Received: 31 March 2016 | Revised: 11 August 2016 | Accepted: 16 August 2016

DOI 10.1002/jlcr.3443

WILEY Journal of Labelled Compounds and Radiopharmaceuticals

RESEARCH ARTICLE

Synthesis of ceramides NS and NP with perdeuterated and specifically ω deuterated *N*-acyl residues

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The synthesis of 12 deuterated ceramides with either a deuteration at the last carbon atom of the amide bound fatty acid or a perdeuterated fatty acid chain is described. The ceramides were prepared starting from sphingosine or phytosphingosine and ω deuterated or perdeuterated fatty acids with PyBOP® as activating agent in high yields. For the synthesis of the specifically deuterated fatty acids, dicarboxylic acids were transformed into ω deuterated alkyl bromide, which was chain elongated with blocked ω bromo alcohols by copper catalyzed Grignard coupling. Oxidation of regenerated alcohol function yields the ω deuterated fatty acids.

KEYWORDS

ceramides, Grignard coupling reactions, perdeuteration, PyBOP® coupling, stratum corneum, ω deuterated fatty acids synthesis

1 | INTRODUCTION

The skin is the largest organ of the human body and consists of 3 different layers, wherein the outermost, the epidermis, is again split into 4 layers. Stratum corneum (SC) as the outside part of the epidermis is necessary to protect the organism. Some significant functions are connected with this skin layer, for instance, the barrier function against various outside influences and the protection against water loss. On the other hand, the SC is the main barrier for dermally administered drugs. The SC consists of corneocytes embedded in a lipid matrix. The lipids in the SC are ceramides, cholesterol, and free fatty acids, the ceramides representing the main component. The distribution of the ceramides is not homogeneous in the lipid matrix. The most important representatives of this lipid class are the amides of sphingosine and phytosphingosine with normal fatty acids of varying chain lengths (CER [NS], CER [NP]). To understand the function of ceramides and manipulate dermal and transdermal drug delivery, it is necessary to understand the molecular superlattice of the lipid matrix. This can be achieved by neutron scattering investigations, especially the determination of the neutron scattering length density profiles of model SC preparations. However, the application of this method requires specifically deuterated ceramides. In literature the use of deuterated compounds in skin research is described rather marginally. Some authors used fully or partially deuterated fatty acids in the mixture with different ceramides and cholesterol or isopropyl myristate.[1–3] Also ceramides containing perdeuterated fatty acids as *N*-acyl residues have been described and investigated.[4–6] Only a few publications were found using specifically deuterated acyl chains. However, the sphingosine moiety was specifically deuterated in the head region by H-D exchange.[7] To the best of our knowledge, there was only 1 publication using ceramides comprising an ω deuterated fatty acid.[8]

The objective of this study was the synthesis of the SC model ceramides with 3 terminally deuterated fatty acids with chain lengths of 18, 22, and 24 carbon atoms for neutron scattering experiments. For this we used the copper catalyzed coupling of deuterated Grignard reagents with ω functionalized bromides. In addition, for the solid-state NMR investigations, ceramides with 3 perdeuterated fatty acids with 12, 18, and 24 carbon atoms were used, in which the tetracosanoic acid was prepared by hydrothermal Pd-catalyzed H/D exchange.[9] The synthesized fatty acids were efficiently bound to the amino group of sphingosine and phytosphingosine with PyBOP® as activating reagent.

In Figure 1, the synthesized ceramides with partially and fully deuterated fatty acids are shown. Whereas type **I** represents ceramides with sphingosine as amino base, the structure type **II** is constituted by ceramides with phytosphingosine.

2 | RESULTS AND DISCUSSION

The structures of different partially deuterated and perdeuterated synthesized CER [NS] and [NP] are shown in Figure 1, as already mentioned above. The syntheses of the precursors are shown in Schemes 1 to 4; those for the deuterated fatty acids and for the final ceramides are given in Scheme 5.

Perdeuterated lauric acid **15d** and stearic acid **15e** were commercially acquired, while perdeuterated lignoceric acid **15f** was prepared according to a modified procedure from the literature.[9] For that, the normal tetracosanoic acid **16** was heated in a hydrogenation apparatus under hydrothermal conditions at 195°C and 15 bar in D_2O for 100 hours. The crude perdeuterated fatty acid was then purified by column chromatography. The deuteration grade was determined to be 96% by high-resolution mass spectrometry (HR MS).[10] For the construction of the corresponding terminally deuterated fatty acids **15a** to **c** (Scheme 5), our synthesis strategy is composed of 2 separate structures, which were connected by a coupling reaction. These 2 structures are on the one hand THP blocked ω bromo alcohols **7a** to **c** and on the other hand terminally deuterated nonanylbromide **13**. The coupling of the protected deuterated fatty alcohols and the further transformation of these alcohols into deuterated fatty acids and the final ceramides **I** and **II** are shown in Scheme 5. The starting materials for the free acids were 2 dicarboxylic acids, or a derivative and a macrocyclic lactone. However, some intermediates in our synthetic process are commercially available, for instance, 9-bromononanol as precursor of **7a**. Because **5a** is not only a building block for **7a** but also for compound **13**, we decided to start from azelaic acid, which is inexpensive.

Following Scheme 1, azelaic acid was used in the synthesis of compounds **7a** and **13** as 2 intermediates for the coupling reactions to the terminally deuterated fatty alcohols or fatty acids. Dicarboxylic acid **1** was transformed into diester **2** using Dean-Stark apparatus followed by a monosaponification step yielding the monoester **3** according to a method described first by Ställberg-Stenhagen[11] with barium hydroxide. Afterwards, compound **3** was selectively reduced to the corresponding methyl 9-hydroxynonanoate (**4a**) by 2 methods. First, we used borane/DMS as reagent.[12] However, beside the displeasing odor of DMS, the reaction in our hands was not satisfying because of low yield and too many by-products. Therefore, we used another method according to Soai et al.[13] The free acid group of the monoester was transformed into the mixed anhydride with ethyl chloroformate. The obtained ω hydroxy ester **4a** was in turn protected by 3,4-dihydro-2*H*-pyran to yield compound **5a**. This substance is one suitable building block for the 2 compounds **7a** and **13** as already mentioned above. The transformation of **5a** into **7a** was realized by a high yield reduction with $LiAlH_4$ to the mono protected diol **6a**, which gives **7a** in 2 steps via the corresponding methanesulfonate and nucleophilic displacement of the mesylate residue against bromine by lithium bromide.

The synthesis of fatty acids with 22 carbon atoms requires compound **7b** (Scheme 2). **7b** is commercially available, however, at a high price. Because we needed larger amounts, we developed a short synthesis protocol starting from the inexpensive ethylene brassylate (**8**). This compound was reduced to diol **9** with $LiAlH_4$. After that, diol **9** was transformed with HBr using a Dean-Stark apparatus into bromotridecanol **10**. For the following Grignard reactions, the free hydroxyl group had to be blocked. This was achieved by a reaction with 3,4-dihydro-2*H*-pyran yielding the corresponding acetal **6b**. Any resultant dibromide and the output product were separated by column chromatography.

In Scheme 3, the preparation pathway of compound **7c** as non-deuterated building block for the coupling reaction to the fatty acid is described. Therefore, cyclopentadecanoate (**11**) was transformed into ω hydroxy ester **4b** by reaction with methanol according to the literature.[14] **4b** was converted to **7c** following the same procedure described for the preparation of **7a** starting from **4a**. All bromides **7a** to **c** were carefully purified by column chromatography before use in the Grignard reaction.

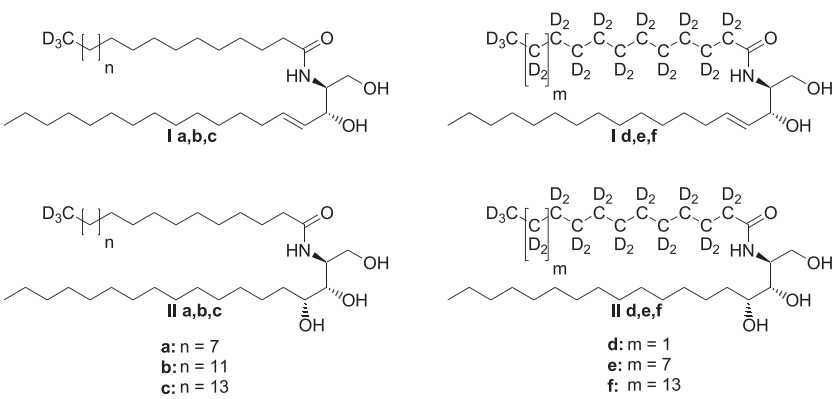
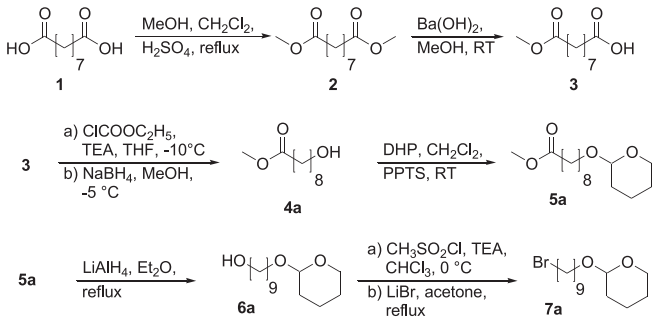
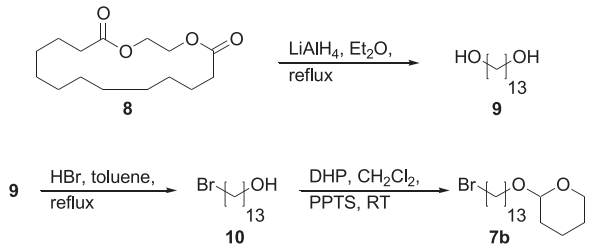


FIGURE 1 Synthesized ceramides with perdeuterated and terminally deuterated labelled *N*-acyl residues



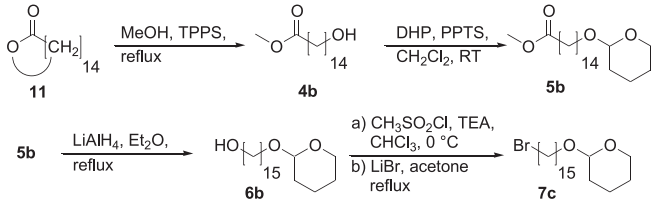
SCHEME 1 Synthesis of 2-[(9-bromononyl)oxy]tetrahydro-*2H*-pyran (**7a**)



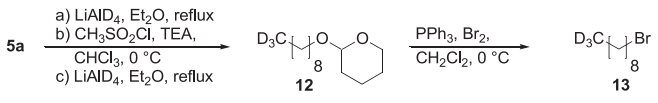
SCHEME 2 Synthesis of 2-[(13-bromotridecyl)oxy]tetrahydro-*2H*-pyran (**7b**)

As mentioned above, compound **5a** serves as starting material for the preparation of the terminally deuterated nonylbromide as coupling partner for compound **7a** to **c** (Scheme 4). For the introduction of the terminal CD_3-group in deuterated compounds, some methods have been described. Beside the reduction of a terminal trichloromethyl group using zinc,[15] the anodic coupling of deuterated acetic acid with mono esters of dicarboxylic acids is described.[16,17]

Terminally deuterated fatty acids were also prepared by alkylating the silver salts of carboxylic esters with a terminal triple bond with CD_3I followed by reduction of the triple bond.[15] Other authors used the reaction of acid chlorides with enamines (Hünig method) or with organocadmium reagents.[18] However, the most common method for the introduction of a terminal CD_3 group is the successive reduction of the ester group by lithium aluminum deuteride. Thus, ester **5a** is reduced with lithium aluminum deuteride followed by



SCHEME 3 Synthesis of 2-[(15-bromopentadecyl)oxy]tetrahydro-*2H*-pyran (**7c**)



SCHEME 4 Synthesis of 1-bromo-9,9,9-d_3-nonane (**13**)

mesylation of the originated alcohol function. The mesylate was not cleaned in between but immediately reacted further to give the deuterated methyl group with lithium aluminum deuteride.[19] For the conversion of compound **12** into nonylbromide **13**, the method of Meyer-Schwartz et al was used.[20]

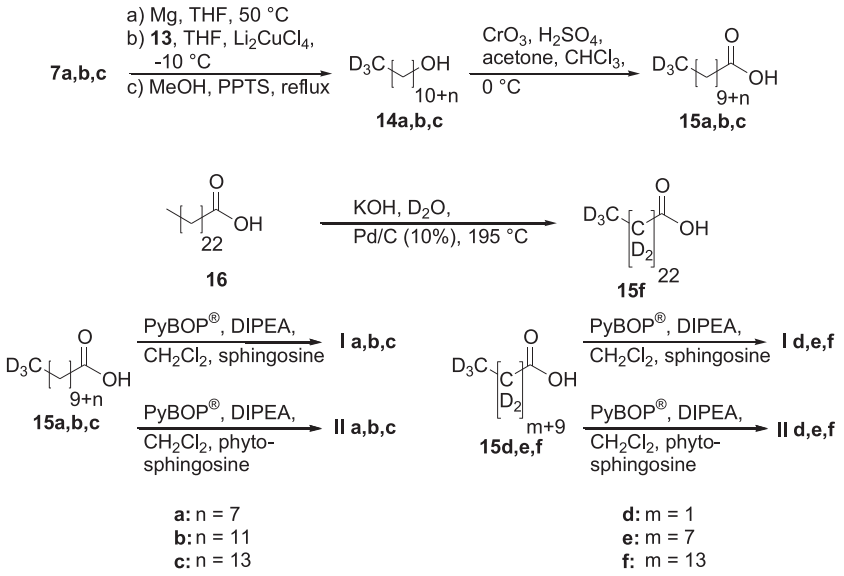
The terminally protected bromides **7a** to **c** were transformed into the Grignard-reagents in THF. Below −5°C these Grignard compounds were coupled with bromide **13** yielding 3 protected long chain alcohols with a triple deuteration at the terminal carbon atom.[21,22] To our knowledge, this coupling reaction was not used for the synthesis of ω deuterated fatty acids before. After removal of the protecting group, the resulting fatty alcohols **14a** to **c** have been oxidized without further purification to the deuterated fatty acids **15a** to **c** (Scheme 5). For that, the corresponding alcohols were oxidized in a different mode. The oxidation with pyridinium dichromate seems to be a very mild method, and therefore, we tested this reaction.[23] However, as a result of this procedure, we found a peak indicating the loss of 1 CH_2 group in the mass spectrum. This is a hint for α oxidation depending on the solvent used during oxidation. The side reaction was significant when chloroform was used in excess, but the use of chloroform beside DMF was necessary to avoid solvation problems. However, pure fatty acids were isolated with a modified jones oxidation procedure.

The further reactions to the ceramides **I** and **II** are shown in Scheme 5. While the perdeuterated fatty acids **15d,e** were acquired commercially, **15f** was synthesized from the corresponding acid **16** by reaction in D_2O at 195°C on Pd/C contact.[9] The perdeuterated as well as the partially deuterated fatty acid **15a** to **f** were then linked to sphingosine or phytoshpingosine. For the activation of the fatty acid, some methods have been reported in the literature. The most commonly used coupling agent is EEDQ.[24,25] Other authors have utilized EDC to form the anhydrides as reactive fatty acid derivative[4] or activated esters of the acids.[26,27] A conventional condensing agent in peptide synthesis is BOP® or PyBOP® as the non-toxic form.[28] Because ceramides contain also an amide bond, we used this reagent successfully and obtained high to very high yields of compounds **I** and **II**. To our knowledge, the use of PyBOP® in the ceramide synthesis has not been previously described. The ceramides of the sphingosines (**Ia-f**) were precipitated with heptane, while the ceramides from phytosphingosine (**IIa-f**) precipitated virtually completely during the reaction.

3 | EXPERIMENTAL

3.1 | General

Perdeuterated lauric acid-d_{23} and stearic acid-d_{35} were purchased from Sigma-Aldrich (St. Louis, MO, USA). Lithium aluminium deuteride was obtained from ARMAR



SCHEME 5 Synthesis of the deuterated fatty acids and ceramides NS and NP

(Europe) GmbH (Leipzig, Germany). PyBOP® was purchased from Carbolution Chemicals GmbH (Saarbrücken, Germany). Sphingosine and phytosphingosine were obtained from Evonik Goldschmidt GmbH (Essen, Germany) and were purified before use. All other chemicals and solvents were purchased from Sigma-Aldrich (St. Louis, MO, USA), and the solvents were dried before use. For column chromatography, silica gel 60 (0.063-0.200 mm) was obtained from Merck (Darmstadt, Germany). TLC plates were received from Macherey-Nagel (Düren, Germany) and were precoated aluminium sheets ALUGRAM® Xtra SIL G/UV$_{254}$. The TLC detection was obtained by using bromothymol blue solution. Melting points were determined on a Boetius apparatus and were not corrected. For mass spectrometry, a Finnigan MAT 710C (Thermoseparation Products, San Jose, CA, USA) for ESI MS, an LTQ-Orbitrap XL hybrid mass spectrometer (Thermo Fisher Scientific, Bremen, Germany) for HR MS, and an MS 5971 A (Hewlett-Packard) for GC–MS were used. ^{1}H NMR and ^{13}C NMR spectra were obtained on Varian Gemini 2000 and Varian Inova 500 instruments. $CDCl_3$, CD_3OD, THF-d_8, and mixtures thereof were used. On a CHNS-932 (Leco Corporation, St. Joseph, MI, USA) the elemental analyses were obtained. For HPLC an HP 1100 Agilent (Agilent Technologies, Waldbronn, Germany) was used with an ELSD 2000 Alltech (Grace Davison, Columbia, MD, USA) detector and a Nucleodur 100-5 125 × 2 column from Macherey-Nagel (Düren, Germany).

3.2 | Synthetic procedures

3.2.1 | Dimethyl azelate (2)

A mixture of azelaic acid (**1**) (106.7 g, 0.57 mol), methanol (85.2 mL, 2.1 mol, 4 eq.), CH_2Cl_2 (100 mL), and conc. sulfuric acid (5 g) was heated in a Dean-Stark apparatus to remove the produced water. When no more water was separated, most of the solvent was evaporated under reduced pressure. The crude diester was taken up in Et_2O (500 mL). Then, the organic layer was washed with water (100 mL), with 5% aqueous KOH (100 mL) and with brine (100 mL). The ether layer was dried over Na_2SO_4 and concentrated to dryness under reduced pressure. The crude residue was purified by column chromatography using gradient technique with heptane/Et_2O gradient to give diester **2** (116.6 g, 95%) as a colorless oil. R_f = 0.50 ($CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.28-1.32 (m, 6H, –$(CH_2)_2$(CH_2)$_3$(CH_2)$_2$–), 1.56-1.64 (m, 4H, 2× –CH_2CH_2COO–), 2.28 (t, J = 7.5 Hz, 4H, 2× –CH_2COO–), 3.65 (s, 6H, 2× –CH_3); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 25.0, 29.0, 29.1, 34.2, 51.6, 174.3; EI MS (*m/z*): 217 $[M]^+$, 185 $[M - OCH_3]^+$; elemental analysis calcd (%) for $C_{11}H_{20}O_4$ (216.27): C 61.09, H 9.32; found: C 60.93, H 9.26.

3.2.2 | 9-Methoxy-9-oxononanoic acid (3)

Dimethyl azelate (**2**) (116.6 g, 0.54 mol) was dissolved in methanol (300 mL). Under stirring a solution of $Ba(OH)_2$ (85 g, 0.27 mol, 0.5 eq.) in methanol (500 mL) was added. The mixture was allowed to stand for 12 hours at room temperature. Then, the formed precipitate was filtered by suction and washed 3 times with diethyl ether (150 mL). The precipitate was taken up with a solution of diluted HCl (20%) (500 mL). The water layer was extracted 3 times with diethyl ether (250 mL). The combined ether layers were washed twice with 5% aqueous K_2CO_3 solution (150 mL), with water (150 mL), with saturated NH_4Cl, dried over Na_2SO_4, and concentrated under reduced pressure. The crude residue was purified by column chromatography using gradient technique with $CHCl_3$/Et_2O gradient to give monoester **3** (64.4 g, 59%) as a colorless oil. R_f = 0.38 ($CHCl_3$/Et_2O 8/2); ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.27-1.34 (m, 6H, –$(CH_2)_2$(CH_2)$_3$(CH_2)$_2$–), 1.56-1.64 (m, 4H, 2× –CH_2CH_2COO–), 2.26-2.33 (m, 4H, 2× –CH_2COO–), 3.64 (s, 3H, –CH_3); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 24.7, 24.9, 28.9-29.0, 34.1, 51.6, 174.4,

180.1; ESI MS (*m*/*z*): 201.2 [*M* − H]$^-$; elemental analysis calcd (%) for $C_{10}H_{18}O_4$ (202.24): C 59.38, H 8.97; found: C 59.27, H 8.98.

3.2.3 | Methyl 9-hydroxynonanoate (4a)

3.2.3.1 | Method A

Compound **3** (22.6 g, 0.11 mol) was dissolved in abs. THF (60 mL). The solution was cooled to −20°C, and a solution of borane/dimethylsulfide (10.4 mL, 0.11 mol, 1 eq.) in abs. THF (100 mL) was added dropwise over 30 minutes under stirring. The stirring was continued at room temperature for 4 hours. For working up, water (200 mL) was added over 10 minutes. K_2CO_3 (26.2 g, 0.19 mol) and Et_2O (200 mL) were added. The organic layer was separated, and the water phase was extracted twice with Et_2O (150 mL). The combined organic layers were washed with brine (200 mL), dried over Na_2SO_4, and concentrated to dryness under reduced pressure. The crude residue was purified by column chromatography using gradient technique with $CHCl_3$/Et_2O gradient to give the methyl 9-hydroxynonanoate **4a** (10.1 g, 48%) as a colorless oil.

3.2.3.2 | Method b

Compound **3** (20.73 g, 0.1 mol) and TEA (14.3 mL, 0.1 mol, 1 eq.) were dissolved in abs. THF (150 mL). The solution was cooled to −10°C, and ethyl chloroformate (9.8 mL, 0.1 mol, 1 eq.) was added dropwise over 45 minutes. The stirring was continued at −10°C for additional 30 minutes. The precipitate was removed by inert filtration. The precipitate was washed twice with abs. THF (30 mL). The combined THF phases were cooled down to −5°C, and $NaBH_4$ (14.73 g, 0.39 mol, 3.8 eq.) was added. Over a period of 2 hours, methanol (61.5 mL, 1.5 mol) was added dropwise. The stirring was continued at −5°C for 1 hour and then at room temperature for 12 hours. For working up, 5 N HCl (100 mL) was added dropwise, followed by adding water (100 mL). The water layer was extracted 3 times with $CHCl_3$ (200 mL). The combined organic layers were washed with 5% aqueous KOH (100 mL), aqua (100 mL), brine (100 mL), dried over Na_2SO_4, and concentrated to dryness under reduced pressure. The crude residue was purified by column chromatography using gradient technique with $CHCl_3$/Et_2O gradient to give the methyl 9-hydroxynonanoate **4a** (15.7 g, 81%) as a colorless oil. R_f = 0.32 ($CHCl_3$/Et_2O 8/2); ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.21-1.32 (m, 8H, HO(CH$_2$)$_2$(C***H***$_2$)$_4$(CH$_2$)$_2$–), 1.48-1.62 (m, 4H, HOCH$_2$C***H***$_2$–, –C***H***$_2$CH$_2$COO–), 2.26 (t, *J* = 7.5 Hz, 2 H, –C***H***$_2$COO–), 3.58 (t, *J* = 6.6 Hz, 2H, HOC***H***$_2$–), 3.63 (s, 3H, –C***H***$_3$); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 25.0, 25.7, 29.1-29.3, 32.8, 34.1, 51.5, 62.9, 174.4; ESI MS (*m*/*z*): 189.0 [*M* + H]$^+$; elemental analysis calcd (%) for $C_{10}H_{20}O_3$ (188.26): C 63.80, H 10.71; found: C 63.59, H 10.72.

3.2.4 | Methyl 15-hydroxypentadecanoate (4b)

Compound **4b** was prepared from the 15-pentadecanolide **11** in accordance with literature.[14] The methyl 15-hydroxypentadecanoate **4b** was used without further characterization and purification. ESI MS (*m*/*z*): 273.0 [*M* − H]$^-$.

3.2.5 | Methyl 9-[(tetrahydro-*2H*-pyran-2-yl)oxy]nonanoate (5a)[29]

A solution of methyl 9-hydroxynonanoate (**4a**) (24.5 g, 130 mmol), DHP (21.2 mL, 234 mmol, 1.8 eq.), and catalytic amounts of PPTS in CH_2Cl_2 (300 mL) was stirred at room temperature. For working up, water (150 mL) was added, and the organic layer was separated. The water phase was extracted twice with CH_2Cl_2 (50 mL). The combined organic layers were washed with brine (150 mL), dried over Na_2SO_4, and concentrated to dryness under reduced pressure. The crude residue was purified by column chromatography using gradient technique with heptane/Et_2O and TEA (10 drops per 100 mL) eluent to give the methyl 9-[(tetrahydro-*2H*-pyran-2-yl)oxy]nonanoate (**5a**) (35.1 g, 99%) as a colorless oil. R_f = 0.41 ($CHCl_3$/Et_2O 8/2); ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.26-1.36 (m, 8H, –(C***H***$_2$)$_4$(CH$_2$)$_2$COO–), 1.48-1.63 (m, 8H, –(C***H***$_2$)$_2$CH$_2$CH–OCH$_2$C***H***$_2$(CH$_2$)$_6$–, –C***H***$_2$CH$_2$COO–), 1.68-1.74 (m, 1H, –OCH–C***H***H′–), 1.78-1.88 (m, 1H, –OCH–CH***H***′–), 2.29 (t, *J* = 7.5 Hz, 2 H, –C***H***$_2$COO–), 3.37 (dt, *J* = 9.6 Hz, *J* = 6.7 Hz, 1H, –OC***H***H′(CH$_2$)$_6$–), 3.46-3.52 (m, 1H, –OCH–(CH$_2$)$_3$C***H***H′–), 3.66 (s, 3H, –C***H***$_3$), 3.71 (dt, *J* = 9.6 Hz, *J* = 6.9 Hz, 1H, –OCH***H***′(CH$_2$)$_6$–), 3.83-3.89 (m, 1H, –OCH–(CH$_2$)$_3$CH***H***′–), 4.55-4.57 (m, 1H, –OC***H***–O–); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 19.9, 25.1, 25.7, 26.3, 29.2-29.9, 30.9, 34.2, 51.6, 62.5, 67.8, 99.0, 174.4; ESI MS (*m*/*z*): 295.1 [*M* + Na]$^+$; elemental analysis calcd (%) for $C_{15}H_{28}O_4$ (272.37): C 66.14, H 10.36; found: C 66.14, H 10.64.

3.2.6 | Methyl 15-[(tetrahydro-*2H*-pyran-2-yl)oxy] pentadecanoate (5b)[29]

Compound **5b** was obtained following the procedure for compound **5a**. Methyl 15-hydroxypentadecanoate (**4b**) (81.4 g, 0.30 mol) was converted into methyl 15-[(tetrahydro-*2H*-pyran-2-yl)oxy]pentadecanoate (**5b**) (103.6 g, 97%) as a colorless oil. R_f = 0.43 ($CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.23-1.34 (m, 20H, –(C***H***$_2$)$_{10}$(CH$_2$)$_2$COO–), 1.48-1.63 (m, 8H, –(C***H***$_2$)$_2$CH$_2$CH–OCH$_2$C***H***$_2$(CH$_2$)$_{12}$–, –C***H***$_2$CH$_2$COO–), 1.66-1.72 (m, 1H, –OCH–C***H***H′–), 1.77-1.85 (m, 1H, –OCH–CH***H***′–), 2.28 (t, *J* = 7.5 Hz, 2H, –C***H***$_2$COO–), 3.36 (dt, *J* = 9.4 Hz, *J* = 6.7 Hz, 1H, –OC***H***H′(CH$_2$)$_{13}$–), 3.45-3.50 (m, 1H, –OCH–(CH$_2$)$_3$C***H***H′–), 3.66 (s, 3H, –C***H***$_3$), 3.71 (dt, *J* = 9.6 Hz, *J* = 6.9 Hz, 1H, –OCH***H***′(CH$_2$)$_{13}$–), 3.82-3.87 (m, 1H, –OCH–(CH$_2$)$_3$CH***H***′–), 4.54-4.56 (m, 1H, –OC***H***–O–); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 19.8, 25.1, 25.6, 26.4, 29.3-29.9, 30.9, 34.2, 51.5, 62.4, 67.8, 98.9,

174.4; ESI MS (*m/z*): 379.2 [*M* + Na]$^+$; elemental analysis calcd (%) for $C_{21}H_{40}O_4$ (356.53): C 70.74, H 11.31; found: C 70.78, H 11.43.

3.2.7 | 9-[(Tetrahydro-*2H*-pyran-2-yl)oxy]nonan-1-ol (6a)

A solution of compound **6a** (13.62 g, 0.05 mol) in Et_2O (150 mL) was added dropwise to a suspension of $LiAlH_4$ (1.85 g, 0.05 mol, 1 eq.) in Et_2O (100 mL). The reaction was heated under reflux until complete reduction. For quenching of the reaction, water (150 mL) was added dropwise under cooling. The mixture was stirred for further 2 hours. The organic layer was separated, and the water phase was extracted twice with Et_2O (100 mL). The combined ether layers were washed with brine (150 mL), dried over Na_2SO_4, and concentrated to dryness under reduced pressure. The crude residue was purified by column chromatography using gradient technique with heptane/Et_2O and TEA (10 drops per 100 mL) eluent to give the 9-[(tetrahydro-2*H*-pyran-2-yl) oxy]nonan-1-ol (**6a**) (11.2 g, 92%) as a colorless oil. $R_f = 0.36$ ($CHCl_3$/Et_2O 8/2); ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.30-1.35 (m, 10H, $HO(CH_2)_2(C\boldsymbol{H}_2)_5$–), 1.48-1.61 (m, 8H, $HOCH_2C\boldsymbol{H}_2$–, –$C\boldsymbol{H}_2CH_2OCH$–$CH_2(C\boldsymbol{H}_2)_2$–), 1.67-1.73 (m, 1H, –OCH–C***H***H′–), 1.77-1.88 (m, 1H, –OCH–CH***H***′–), 3.37 (dt, *J* = 9.6 Hz, *J* = 6.7 Hz, 1H, –$(CH_2)_8C\boldsymbol{H}$H′O–), 3.46-3.52 (m, 1H, –OCH–$(CH_2)_3C\boldsymbol{H}$H′–), 3.61 (t, *J* = 6.7 Hz, 2H, $HOC\boldsymbol{H}_2$–), 3.71 (dt, *J* = 9.6 Hz, *J* = 6.9 Hz, 1H, –$(CH_2)_8CH\boldsymbol{H}'$O–), 3.83-3.88 (m, 1H, –OCH–$(CH_2)_3CH\boldsymbol{H}'$–), 4.55-4.57 (m, 1H, –O***C***H–O–); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 19.8, 25.6, 25.9, 26.3, 29.5-29.9, 30.9, 32.9, 62.5, 63.1, 67.8, 99.0; ESI MS (*m/z*): 267.1 [*M* + Na]$^+$; elemental analysis calcd (%) for $C_{14}H_{28}O_3$ (244.36): C 68.81, H 11.55; found: C 68.69, H 11.56.

3.2.8 | 15-[(Tetrahydro-*2H*-pyran-2-yl)oxy]pentadecan-1-ol (6b)

Compound **6b** was obtained following the procedure for compound **6a**. Methyl 15-[(tetrahydro-2*H*-pyran-2-yl)oxy] pentadecanoate (**5b**) (17.83 g, 0.05 mol) was converted into 15-[(tetrahydro-2*H*-pyran-2-yl)oxy]pentadecan-1-ol (**6b**) (14.46 g, 88%) as a white waxy solid. $R_f = 0.38$ ($CHCl_3$/Et_2O 8/2); mp 29°C; ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.26-1.38 (m, 22H, $HO(CH_2)_2(C\boldsymbol{H}_2)_{11}$–), 1.48-1.62 (m, 8H, $HOCH_2C\boldsymbol{H}_2$–, –$C\boldsymbol{H}_2CH_2OCH$–$CH_2(C\boldsymbol{H}_2)_2$–), 1.68-1.74 (m, 1H, –OCH–C***H***H′–), 1.80-1.87 (m, 1H, –OCH–CH***H***′–), 3.38 (dt, *J* = 9.6 Hz, *J* = 6.7 Hz, 1H, –$(CH_2)_{14}C\boldsymbol{H}$H′O–), 3.47-3.52 (m, 1H, –OCH–$(CH_2)_3C\boldsymbol{H}$H′–), 3.64 (t, *J* = 6.7 Hz, 2H, $HOC\boldsymbol{H}_2$–), 3.72 (dt, *J* = 9.6 Hz, *J* = 6.9 Hz, 1H, –$(CH_2)_{14}CH\boldsymbol{H}'$O–), 3.85-3.89 (m, 1H, –OCH–$(CH_2)_3CH\boldsymbol{H}'$–), 4.57-4.58 (m, 1H, –O***C***H–O–); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 19.9, 25.7, 25.9, 26.4, 29.6-29.9, 31.0, 33.0, 62.5, 63.3, 67.9, 99.0; ESI MS (*m/z*): 351.2 [*M* + Na]$^+$; elemental analysis calcd (%) for $C_{20}H_{40}O_3$ (328.52): C 73.12, H 11.27; found: C 73.11, H 11.54.

3.2.9 | Tridecane-1,13-diol (9)

Compound **9** was prepared from ethylene brassylate (**8**) by reduction with $LiAlH_4$. The tridecane-1,13-diol (**9**) was used without further characterization and purification.

3.2.10 | 13-Bromotridecan-1-ol (10)

Compound **10** was prepared from tridecane-1,13-diol (**9**) in accordance with the literature.[30] 13-Bromotridecan-1-ol (**10**) was used without further characterization and purification.

3.2.11 | 2-[(9-Bromononyl)oxy]tetrahydro-*2H*-pyran (7a)[31]

Compound **6a** (10.51 g, 0.043 mol) and TEA (6.26 mL, 0.045 mol, 1.05 eq.) were dissolved in $CHCl_3$ (100 mL). The mixture was cooled to 0°C, and a solution of methanesulfonyl chloride (3.5 mL, 0.045 mol, 1.05 eq.) in $CHCl_3$ (25 mL) was added dropwise under strong stirring. The mixture was warmed to room temperature and was stirred for further 12 hours. For working up, TEA (6.26 mL, 0.045 mol) was added before ice (100 g) was added. The mixture was stirred for further 2 hours. The organic layer was separated, and the water phase was extracted 3 times with $CHCl_3$ (50 mL). The combined organic layers were washed with brine (150 mL), dried over Na_2SO_4, concentrated to dryness under reduced pressure, and were dried over P_2O_5. The crude residue was dissolved in acetone (80 mL). Dry LiBr (9.34 g, 0.108 mol) was added, and the mixture was heated under reflux for 6 hours. For working up, most of the solvent was evaporated. The residue was poured into ice. The precipitate was extracted 3 times with Et_2O (100 mL). The combined ether phases were washed twice with brine (100 mL), dried over Na_2SO_4, and concentrated to dryness under reduced pressure. The crude residue was purified by column chromatography using gradient technique with heptane/Et_2O and TEA (10 drops per 100 mL) eluent to give the 2-[(9-bromononyl)oxy] tetrahydro-2*H*-pyran (**7a**) (10.6 g, 80%) as a colorless oil. $R_f = 0.47$ ($CHCl_3$/heptane 6/4); ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.30-1.43 (m, 10H, $Br(CH_2)_2(C\boldsymbol{H}_2)_5$–), 1.50-1.61 (m, 6H, –$C\boldsymbol{H}_2CH_2OCH$–$CH_2(C\boldsymbol{H}_2)_2$–), 1.68-1.74 (m, 1H, –OCH–C***H***H′–), 1.78-1.88 (m, 3H, $BrCH_2C\boldsymbol{H}_2$–, –OCH–CH***H***′–), 3.35-3.41 (m, 3H, $BrC\boldsymbol{H}_2(CH_2)_7C\boldsymbol{H}$H′–), 3.47-3.52 (m, 1H, –OCH–$(CH_2)_3C\boldsymbol{H}$H′–), 3.72 (dt, *J* = 9.5 Hz, *J* = 6.9 Hz, 1H, $Br(CH_2)_8CH\boldsymbol{H}'$–), 3.84-3.89 (m, 1H, –OCH–$(CH_2)_3CH\boldsymbol{H}'$–), 4.56-4.57 (m, 1H, –O***C***H–O–); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 19.9, 25.7, 26.3, 28.3, 28.3, 29.5, 29.9, 30.9, 33.0, 34.1, 62.5, 67.8, 99.0; EI-MS (*m/z*): 305/307 [*M* − H]$^+$; elemental analysis calcd (%) for $C_{14}H_{27}BrO_2$ (307.25): C 54.72, H 8.86; found: C 54.91, H 9.06.

3.2.12 | 2-[(13-Bromotridecyl)oxy]tetrahydro-*2H*-pyran (**7b**)[29]

Compound **7b** was prepared from 13-bromotridecan-1-ol (**10**) in accordance with literature. The analytical data of 2-[(13-bromotridecyl)oxy]tetrahydro-*2H*-pyran (**7b**) are in accordance with the reported in the literature.[32] 2-[(13-Bromotridecyl)oxy]tetrahydro-*2H*-pyran (**7b**) was used without further characterization. ESI MS (*m/z*): 385.0/387.1 $[M + Na]^+$.

3.2.13 | 2-[(15-Bromopentadecyl)oxy]tetrahydro-*2H*-pyran (**7c**).[31]

Compound **7c** was obtained following the procedure for compound **7a**. 15-[(Tetrahydro-*2H*-pyran-2-yl)oxy] pentadecan-1-ol (**6b**) (13.80 g, 0.042 mol) was converted into 2-[(15-bromopentadecyl)oxy]tetrahydro-*2H*-pyran (**7c**) (12.5 g, 76%) as a colorless oil. The analytical data of compound **7c** are in accordance with the reported in the literature.[33] ESI MS (*m/z*): 413.1/415.1 $[M + Na]^+$.

3.2.14 | 2-(9,9,9-d_3-Nonyloxy)tetrahydro-*2H*-pyran (**12**)[19]

A solution of compound **5a** (15.62 g, 0.057 mol) in Et_2O (65 mL) was added dropwise to a suspension of $LiAlD_4$ (1.44 g, 0.034 mol, 0.6 eq.) in Et_2O (50 mL). The mixture was heated until complete conversion. For working up, D_2O (3.5 mL) was added dropwise under ice cooling. The mixture was stirred for 2 hours. The precipitate was removed via filtering by suction. The filter was washed 3 times with Et_2O (10 mL). The combined ether phases were washed with water (70 mL), brine (70 mL), dried over Na_2SO_4, concentrated to dryness under reduced pressure, and dried over P_2O_5. The crude residue of the alcohol and TEA (8.6 mL, 0.062 mol) were dissolved in $CHCl_3$ (100 mL). The mixture was cooled to 0°C, and a solution of methanesulfonyl chloride (4.8 mL, 0.062 mol) in $CHCl_3$ (50 mL) was added dropwise under stirring. The mixture was warmed to room temperature and was stirred for further 12 hours. For working up, TEA (8.6 mL, 0.062 mol) was added before ice water (50 mL) was added. The mixture was stirred for further 2 hours. The organic layer was separated, and the water phase was extracted 3 times with $CHCl_3$ (50 mL). The combined organic layers were washed with brine (150 mL), dried over Na_2SO_4, concentrated to dryness under reduced pressure, and dried over P_2O_5. A solution of the dried mesylate in Et_2O (75 mL) was dropped into a suspension of $LiAlD_4$ (1.34 g, 32 mmol) in Et_2O (25 mL). The mixture was heated until complete conversion. For working up, the reaction was cooled to 0°C. Water (3.5 mL) was added dropwise, and the mixture was stirred for further 2 hours. The precipitate was removed via filtering by suction. The filter was washed 3 times with Et_2O (10 mL). The combined ether phases were washed with water (50 mL), brine (50 mL), dried over Na_2SO_4, and concentrated to dryness under reduced pressure. The crude residue was purified by column chromatography using gradient technique with heptane/Et_2O and TEA (10 drops per 100 mL) eluent to give the 2-(9,9,9-d_3-nonyloxy) tetrahydro-*2H*-pyran (**12**) (8.83 g, 64%) as a colorless oil. R_f = 0.61 ($CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.25-1.36 (m, 12H, $D_3C(CH_2)_6$–), 1.48-1.62 (m, 6H, –CH_2CH_2OCH–$CH_2(CH_2)_2$–), 1.67-1.74 (m, 1H, –OCH–CHH'–), 1.78-1.86 (m, 1H, –OCH–CHH'–), 3.37 (dt, J = 9.6 Hz, J = 6.7 Hz, 1H, $D_3C(CH_2)_7CHH'$–), 3.46-3.54 (m, 1H, –OCH–$(CH_2)_3CHH'$–), 3.72 (dt, J = 9.6 Hz, J = 6.9 Hz, 1H, $D_3C(CH_2)_7CHH'$–), 3.83-3.89 (m, 1H, –OCH–$(CH_2)_3CHH'$–), 4.55-4.57 (m, 1H, –OC*H*–O–); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 19.8, 22.5, 25.7, 26.4, 29.4, 29.6, 29.7, 29.9, 30.9, 31.9, 62.4, 67.8, 99.0; ESI MS (*m/z*): 254.2 $[M + Na]^+$; GC-MS: t_R = 12.2 minutes, *purity* > 99.5%, (*m/z*) 230 [10%, $M - H^+$].

3.2.15 | 1-Bromo-9,9,9-d_3-nonane (**13**)[20]

To a stirred solution of triphenylphosphine (21.35 g, 0.082 mol) in CH_2Cl_2 (200 mL), a solution of bromine (4.2 mL, 0.082 mol) in CH_2Cl_2 (40 mL) was added at a temperature of 0°C. The white colored mixture was stirred for further 30 minutes before a solution of compound **12** (8.83 g, 0.038 mol) was added. The reaction was stirred for a 12 hours at room temperature. The CH_2Cl_2 phase was washed twice with water (50 mL), dried over Na_2SO_4, and concentrated to dryness under reduced pressure. The residue was absorbed on silica gel before purifying by column chromatography with heptane eluent to give the 1-bromo-9,9,9-d_3-nonane (**13**) (8.02 g, 89%) as a colorless oil. R_f = 0.60 ($CHCl_3$/heptane 2/8); ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.27-1.32 (m, 10H, $D_3C(CH_2)_5$–), 1.39-1.44 (m, 2H, –$CH_2(CH_2)_2Br$), 1.82-1.89 (m, 2H, –CH_2CH_2Br), 3.40 (t, J = 6.9 Hz, 2H, –CH_2Br); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 22.5, 28.3, 28.9, 29.4, 29.6, 31.9, 33.0, 34.2; GC-MS: t_R = 7.5 minutes, *purity* > 99.5%, (*m/z*) 209/211 [2%, M^+], 135/137 [100%, $Br(CH_2)_4^+$].

3.3 | General procedure for the synthesis of the ω,ω,ω-d_3-alkan-1-ol (**14a,b,c**).[21,22]

A solution of compound **7a,b,c** (8 mmol) in abs. THF (20 mL) was dropped to magnesium turnings (0.40 g, 16.4 mmol) under argon atmosphere. The mixture was warmed at 50°C for 3 hours. The magnesium excess was removed. The Grignard solution was cooled to −5°C before a solution of compound **13** (1.47 g, 7 mmol) in abs THF (50 mL) and a solution of freshly prepared Li_2CuCl_4 in abs THF (1.4 mL, 0.1M) were added under stirring. The mixture was stirred for 3 hours at −10°C. For working up, a cold saturated NH_4Cl solution (50 mL) was added, and the mixture was extracted 3 times with ether (50 mL). The combined ether phases were washed with brine, dried over Na_2SO_4, and concentrated to dryness under reduced pressure. The residue was dissolved in 30 mL MeOH. Catalytic amounts of PPTS were added. The mixture was heated under reflux

for 3 hours. Afterwards, the mixture was cooled to −20°C for full crystallization. The formed precipitated was filtered off to give the ω,ω,ω-d_3-alkan-1-ols (**14a,b,c**).

3.3.1 | 18,18,18-d_3-Octadecan-1-ol (14a)

Yield: (1.05 g, 55%); white crystalline solid; R_f = 0.27 ($CHCl_3$); mp 54°C; ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.26-1.43 (m, 30H, $D_3C(CH_2)_{15}$–), 1.53-1.60 (m, 2H, –CH_2CH_2OH), 3.44 (t, J = 6.6 Hz, 2H, –CH_2OH); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 22.6, 25.9, 29.5-29.9, 32.0, 33.0, 63.3; GC-MS: t_R = 17.24 minutes, *purity* > 99.5%, (*m/z*) 273 [1%, M^+], 227 [5%, M–$C_3H_4D_3^+$].

3.3.2 | 22,22,22-d_3-Docosan-1-ol (14b)

Yield: (1.11 g, 48%); white crystalline solid; R_f = 0.37 ($CHCl_3$); mp 68°C; ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.26-1.45 (m, 38H, $D_3C(CH_2)_{19}$–), 1.53-1.60 (m, 2H, –CH_2CH_2OH), 3.64 (t, J = 6.6 Hz, 2H, –CH_2OH); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 22.6, 25.9, 29.5-29.9, 32.0, 33.0, 63.3; GC–MS: t_R = 17.03 minutes, *purity* > 95%, (*m/z*) 328 [2%, $M - H^+$].

3.3.3 | 24,24,24-d_3-Tetracosan-1-ol (14c)

Yield: (1.28 g, 51%); white crystalline solid; R_f = 0.29 ($CHCl_3$); mp 73°C; ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.26-1.43 (m, 42H, $D_3C(CH_2)_{21}$–), 1.53-1.60 (m, 2H, –CH_2CH_2OH), 3.64 (t, J = 6.6 Hz, 2H, –CH_2OH); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 22.6, 25.9, 29.5-29.9, 32.0, 33.0, 63.3; ESI MS (*m/z*): 735.7 $[2M + Na]^+$;

3.4 | General procedure for the synthesis of the ω,ω,ω-d_3-alkanoic acid (15a,b,c)

CrO_3 (4.4 mmol) was suspended in 10 N H_2SO_4 (2.22 mL). The mixture was cooled to 0°C. A solution of the ω,ω,ω-d_3-alkan-1-ols (14a,b,c) (2 mmol) in a mixture of acetone (50 mL) and $CHCl_3$ (30 mL) was added dropwise over a period of 2 hours. Afterwards, the mixture was stirred for an additional hour under cooling. For working up, water (20 mL) and brine (20 mL) were added, and the organic phase was separated. The aqueous phase was extracted 3 times with $CHCl_3$ (50 mL). The combined organic phases were dried over Na_2SO_4 and concentrated to dryness under reduced pressure. The crude residue was purified by column chromatography using gradient technique with heptane/$CHCl_3$/Et_2O eluent to give the ω,ω,ω-d_3-alkanoic acids (**15a,b,c**).

3.4.1 | 18,18,18-d_3-Octadecanoic acid (15a)

Yield: (0.47 g, 81%); white crystalline solid; R_f = 0.34 ($CHCl_3$/Et_2O 8/2); mp 67°C; ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.26-1.35 (m, 28H, $D_3C(CH_2)_{14}$–), 1.60-1.67 (m, 2H, –CH_2CH_2COOH), 2.35 (t, J = 7.5 Hz, 2H, –CH_2COOH); ^{13}C NMR (100 MHz, $CDCl_3$): δ (ppm) 22.6, 24.9, 29.2-29.9, 32.0, 34.1, 179.6; ESI MS (*m/z*): 286.6 $[M - H]^-$, 573.3 $[2M - H]^-$, 595.7 $[2M - 2H - Na]^-$.

3.4.2 | 22,22,22-d_3-Docosanoic acid (15b)

Yield: (0.54 g, 79%); white crystalline solid; R_f = 0.51 ($CHCl_3$/Et_2O 8/2); mp 76°C; ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.26-1.35 (m, 36H, $D_3C(CH_2)_{18}$–), 1.61-1.66 (m, 2H, –CH_2CH_2COOH), 2.35 (t, J = 7.5 Hz, 2H, –CH_2COOH); ^{13}C NMR (100 MHz, THF-d_8): δ (ppm) 23.5, 26.0, 30.3-30.8, 33.0, 34.4, 174.6; ESI MS (*m/z*): 685.4 $[2M - H]^-$.

3.4.3 | 24,24,24-d_3-Tetracosanoic acid (15c)

Yield: (0.60 g, 84%); white crystalline solid; R_f = 0.51 ($CHCl_3$/Et_2O 8/2); mp 80.5°C; ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 1.26-1.36 (m, 40H, $D_3C(CH_2)_{20}$–), 1.60-1.67 (m, 2H, –CH_2CH_2COOH), 2.35 (t, J = 7.5 Hz, 2H, –CH_2COOH); ^{13}C NMR (100 MHz, THF-d_8): δ (ppm) 23.5, 26.0, 30.3-30.8, 33.0, 34.4, 174.6; ESI MS (*m/z*): 370.8 $[M - H]^-$, 763.9 $[2M + Na - 2H]^-$.

3.4.4 | Tetracosanoic acid-d_{47} (15f)[9]

A mixture of tetracosanoic acid **16** (3 g, 8.1 mmol), KOH (530 mg, 9.4 mmol), D_2O (100 mL), and Pd/C (10%) (300 mg) was stirred under argon atmosphere at 195°C and 15 bar for 100 hours. For working up, the mixture was acidified with conc. DCl (1 mL), and $CHCl_3$ (200 mL) was added. The mixture was filtrated to remove Pd/C. Afterwards, the organic layer was separated, and then the aqueous phase was extracted twice with $CHCl_3$ (100 mL). The combined organic layers were dried over Na_2SO_4 and concentrated to dryness under reduced pressure. The crude residue was purified by column chromatography using $CHCl_3$/MeOH eluent to give the tetracosanoic acid-d_{47} (**15d**) (2.81 g, 83%) as a white, crystalline solid.

R_f = 0.48 ($CHCl_3$/Et_2O 8/2); mp 75°C; HR MS (*m/z*) $[M - H]^-$ $C_{24}H_1D_{46}O_2$: calculated 413.6469; found 413.6483.

3.5 | General procedure for coupling of the fatty acids and sphingoid bases to the ceramides[28]

The fatty acid (**15a-f**) (1 eq.) and PyBOP® (1.1 eq.) were given to CH_2Cl_2 (5 mL/0.5 mmol acid). DIPEA (2 eq.) was added under stirring to the mixture. After 15 minutes, sphingosine or phytosphingosine (1.1 eq.) was added to the clear solution. The mixture was stirred for 12 hours.

For working up of the sphingosine containing ceramides (**Ia-f**), heptane (10 mL/0.5 mmol acid) was added to the suspension for a full precipitation before filtration. The precipitate was washed twice with heptane. The separated residue was purified by column chromatography using gradient technique with $CHCl_3$/MeOH and NH_3 (0.5%) eluent.

For working up of the phytosphingosine containing ceramides (**IIa-f**), the mixtures were evaporated to dryness.

The crude residues were absorbed on silica gel for purifying by column chromatography using gradient technique with $CHCl_3$/MeOH and NH_3 (0.5%) eluent.

3.5.1 | *N*-[(2*S*,3*R*,4*E*)-1,3-Dihydroxyoctadec-4-en-2-yl]-18,18,18-d_3-octadecanamid (Ia)

Compound **15a** (200 mg, 0.7 mmol) was converted into the product **Ia** (0.32 g, 81%); white waxy solid; R_f = 0.27 ($CHCl_3$/MeOH/NH_3 95/5/1); mp 98°C; ^{1}H NMR (500 MHz, $CDCl_3$): δ (ppm) 0.88 (t, *J* = 6.9 Hz, 3H, –C***H***$_3$), 1.26-1.39 (m, 50H, $D_3C(CH_2)_{14}$–, $H_3C(CH_2)_{11}$–), 1.61-1.67 (m, 2H, –C***H***$_2$CH$_2$CONH–), 2.04-2.08 (m, 2H, –C***H***$_2$CH=CH–), 2.23 (t, *J* = 7.6 Hz, 2H, –C***H***$_2$CONH–), 3.71 (dd, *J* = 11.3 Hz, *J* = 3.3 Hz, 1H, –C***H***H′OH), 3.89-3.93 (m, 1H, –C***H***–CH$_2$OH), 3.96 (dd, *J* = 11.2 Hz, *J* = 3.8 Hz, 1H, –CH***H***′OH), 4.31-4.33 (m, 1H, –CH=CHC***H***OH–), 5.53 (dd, *J* = 15.4 Hz, *J* = 6.4 Hz, 1H, –CH=C***H***CHOH–), 5.76-5.82 (m, 1H, –C***H***=CHCHOH–), 6.22 (d, *J* = 7.3 Hz, 1H, –CON***H***–); ^{13}C NMR (125 MHz, $CDCl_3$): δ (ppm) 14.3, 22.6, 25.9, 29.3-29.9, 32.0, 32.1, 32.4, 37.0, 54.7, 62.7, 74.9, 129.0, 134.5, 174.0; ESI MS (*m*/*z*): 591.6 [*M* + Na]$^+$, 1159.4 [2*M* + Na]$^+$, 567.7 [*M* − H]$^-$; HR MS (*m*/*z*) [M + H]$^+$ $C_{36}H_{69}D_3NO_3$: calculated 569.5695; found 569.5696; HPLC ELSD purity >99.5%.

3.5.2 | *N*-[(2*S*,3*R*,4*E*)-1,3-Dihydroxyoctadec-4-en-2-yl]-22,22,22-d_3-docosanamid (Ib)

Compound **15b** (200 mg, 0.58 mmol) was converted into the product **Ib** (0.30 g, 83%); white waxy solid; R_f = 0.31 ($CHCl_3$/MeOH/NH_3 95/5/1); mp 97°C; ^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) 0.88 (t, *J* = 6.8 Hz, 3H, –C***H***$_3$), 1.10-1.39 (m, 58H, $D_3C(CH_2)_{18}$–, $H_3C(CH_2)_{11}$–), 1.60-1.68 (m, 2H, –C***H***$_2$CH$_2$CONH–), 2.03-2.09 (m, 2H, –C***H***$_2$CH=CH–), 2.23 (t, *J* = 7.6 Hz, 2H, –C***H***$_2$CONH–), 3.71 (dd, *J* = 11.1 Hz, *J* = 3.1 Hz, 1H, –C***H***H′OH), 3.89-3.93 (m, 1H, –C***H***–CH$_2$OH), 3.96 (dd, *J* = 11.2 Hz, *J* = 3.8 Hz, 1H, –CH***H***′OH), 4.31-4.34 (m, 1H, –CH=CHC***H***OH–), 5.53 (dd, *J* = 15.4 Hz, *J* = 6.4 Hz, 1H, –CH=C***H***CHOH–), 5.75-5.82 (m, 1H, –C***H***=CHCHOH–), 6.22 (d, *J* = 7.1 Hz, 1H, –CON***H***–); ^{13}C NMR (125 MHz, $CDCl_3$, 40°C): δ (ppm) 14.2, 22.6, 22.8, 25.9, 29.3-29.9, 32.0, 32.1, 32.4, 37.0, 54.8, 62.7, 74.8, 129.1, 134.5, 174.0; ESI MS (*m*/*z*): 623.8 [*M* − H]$^-$; HR MS (*m*/*z*) [M + H]$^+$ $C_{40}H_{77}D_3NO_3$: calculated 625.6321; found 625.6353; HPLC ELSD purity >99.5%.

3.5.3 | *N*-[(2*S*,3*R*,4*E*)-1,3-Dihydroxyoctadec-4-en-2-yl]-24,24,24-d_3-tetracosanamid (Ic)

Compound **15c** (200 mg, 0.54 mmol) was converted into the product **Ic** (0.27 g, 77%); white waxy solid; R_f = 0.34 ($CHCl_3$/MeOH/NH_3 95/5/1); mp 95.5°C; ^{1}H NMR (500 MHz, $CDCl_3$): δ (ppm) 0.88 (t, *J* = 7.0 Hz, 3H, –C***H***$_3$), 1.26-1.39 (m, 60H, $D_3C(CH_2)_{20}$–, $H_3C(CH_2)_{11}$–), 1.61-1.67 (m, 2H, –C***H***$_2$CH$_2$CONH–), 2.04-2.08 (m, 2H, –C***H***$_2$CH=CH–), 2.23 (t, *J* = 7.6 Hz, 2H, –C***H***$_2$CONH–), 3.71 (dd, *J* = 11.2 Hz, *J* = 3.4 Hz, 1H, –C***H***H′OH), 3.89-3.93 (m, 1H, –C***H***–CH$_2$OH), 3.96 (dd, *J* = 11.3 Hz, *J* = 3.8 Hz, 1H, –CH***H***′OH), 4.31-4.34 (m, 1H, –CH=CHC***H***OH–), 5.53 (dd, *J* = 15.4 Hz, *J* = 6.5 Hz, 1H, –CH=C***H***CHOH–), 5.76-5.82 (m, 1H, –C***H***=CHCHOH–), 6.20 (d, *J* = 7.1 Hz, 1H, –CON***H***–); ^{13}C NMR (125 MHz, $CDCl_3$, 40°C): δ (ppm) 14.2, 22.6, 22.8, 25.9, 29.3-29.9, 32.0, 32.1, 32.4, 37.0, 54.8, 62.7, 74.9, 129.1, 134.5, 174.0; ESI MS (*m*/*z*): 651.8 [*M* − H]$^-$; HR MS (*m*/*z*) [M + H]$^+$ $C_{42}H_{81}D_3NO_3$: calculated 653.6634; found 653.6644; HPLC ELSD purity >98%.

3.5.4 | *N*-[(2*S*,3*R*,4*E*)-1,3-Dihydroxyoctadec-4-en-2-yl]-d_{23}-dodecanamid (Id)

Lauric acid-d_{23} (**15d**) (100 mg, 0.45 mmol) was converted into the product **Id** (0.17 g, 71%); white waxy solid; R_f = 0.44 ($CHCl_3$/MeOH/NH_3 95/5/1); mp 49°C; ^{1}H NMR (500 MHz, $CDCl_3$): δ (ppm) 0.87 (t, *J* = 6.7 Hz, 3H, –C***H***$_3$), 1.19-1.37 (m, 22H, $H_3C(CH_2)_{11}$–), 2.01-2.05 (m, 2H, –C***H***$_2$CH=CH–), 3.67 (dd, *J* = 8.6 Hz, *J* = 2.4 Hz, 1H, –C***H***H′OH), 3.86-3.93 (m, 2H, –C***H***–CH***H***′OH), 4.25-4.28 (m, 1H, –CH=CHC***H***OH–), 5.51 (dd, *J* = 15.4 Hz, *J* = 6.4 Hz, 1H, –CH=C***H***CHOH–), 5.72-5.78 (m, 1H, –C***H***=CHCHOH–), 6.36-6.40 (m, 1H, –CON***H***–); ^{13}C NMR (125 MHz, $CDCl_3$): δ (ppm) 14.2, 22.8, 29.3-29.8, 32.0, 32.5, 54.8, 62.5, 74.4, 129.1, 134.1, 174.2; ESI MS (*m*/*z*): 527.6 [*M* + Na]$^+$; 503.7 [*M* − H]$^-$, HR MS (*m*/*z*) [M + H]$^+$ $C_{30}H_{37}D_{23}NO_3$: calculated 505.6011; found 505.6012; HPLC ELSD purity >99.5%.

3.5.5 | *N*-[(2*S*,3*R*,4*E*)-1,3-Dihydroxyoctadec-4-en-2-yl]-d_{35}-octadecanamid (Ie)

Stearic acid-d_{35} (**15d**) (100 mg, 0.31 mmol) was converted into product **Ie** (0.15 g, 79%); white waxy solid; R_f = 0.46 ($CHCl_3$/MeOH/NH_3 95/5/1); mp 94°C; ^{1}H NMR (500 MHz, $CDCl_3$): δ (ppm) 0.89 (t, *J* = 6.7 Hz, 3H, –C***H***$_3$), 1.24-1.41 (m, 22H, $H_3C(CH_2)_{11}$–), 2.02-2.10 (m, 2H, –C***H***$_2$CH=CH–), 3.72 (dd, *J* = 11.0 Hz, *J* = 2.8 Hz, 1H, –C***H***H′OH), 3.89-3.93 (m, 1H, –C***H***–CH$_2$OH), 3.96 (dd, *J* = 11.2 Hz, *J* = 3.8 Hz, 1H, –CH***H***′OH), 4.31-4.34 (m, 1H, –CH=CHC***H***OH–), 5.54 (dd, *J* = 15.4 Hz, *J* = 6.4 Hz, 1H, –CH=C***H***CHOH–), 5.75-5.83 (m, 1H, –C***H***=CHCHOH–), 6.19 (d, *J* = 6.5 Hz, 1H, –CON***H***–); ^{13}C NMR (125 MHz, $CDCl_3$): δ (ppm) 14.1, 22.7, 28.0-29.7, 31.9, 32.3, 54.5, 62.5, 74.6, 128.8, 134.3, 174.0; ESI MS (*m*/*z*): 623.6 [*M* + Na]$^+$; 1223.8 [2*M* − Na]$^+$; 599.2 [*M* − H]$^-$; HR MS (*m*/*z*) [M + Na]$^+$ $C_{36}H_{36}D_{35}NO_3Na$: calculated 623.7523; found 623.7522; HPLC ELSD purity >99.5%.

3.5.6 | *N*-[(2*S*,3*R*,4*E*)-1,3-Dihydroxyoctadec-4-en-2-yl]-D_{47}-tetracosanamid (If)

Tetracosanoic acid-d_{47} (**15f**) (100 mg, 0.24 mmol) was converted into the product **If** (0.14 g, 81%); white waxy solid; R_f = 0.56 ($CHCl_3$/MeOH/NH_3 95/5/1); mp 93°C; ^{1}H NMR (500 MHz, $CDCl_3$): δ (ppm) 0.89 (t, *J* = 6.7 Hz, 3H, –C***H***$_3$), 1.20-1.40 (m, 22H, $H_3C(CH_2)_{11}$–), 2.04-2.09 (m, 2H, –C***H***$_2$CH=CH–), 3.71 (dd, *J* = 10.9 Hz, *J* = 2.8 Hz,

1H, –C*H*H′OH), 3.89-3.92 (m, 1H, –C*H*–CH_2OH), 3.95 (dd, J = 11.1 Hz, J = 3.8 Hz, 1H, –CH*H*′OH), 4.31-4.33 (m, 1H, –CH=CHC*H*OH–), 5.54 (dd, J = 15.4 Hz, J = 6.4 Hz, 1H, –CH=C*H*CHOH–), 5.76-5.83 (m, 1H, –C*H*=CHCHOH–), 6.18 (d, J = 7.2 Hz, 1H, –CON*H*–); ^{13}C NMR (125 MHz, $CDCl_3$, 40°C): δ (ppm) 14.2, 22.8, 29.3-29.9, 32.1, 32.4, 54.8, 62.7, 74.9, 129.1, 134.5, 173.9; ESI MS (*m/z*): 718.8 [$C_{42}H_{37}D_{46}NO_3$ + Na]$^+$; 695.0 [$C_{42}H_{37}D_{46}NO_3$ − H]$^-$; HR MS (*m/z*) [M + Na]$^+$ $C_{42}H_{37}D_{46}NO_3Na$: calculated 718.9152; found 718.9144; HPLC ELSD purity >99.5%.

3.5.7 | *N*-[(2*S*,3*S*,4*R*)-1,3,4-Trihydroxyoctadec-2-yl]-18,18,18-d_3-octadecanamid (IIa)

Compound **15a** (200 mg, 0.7 mmol) was converted into the product **IIa**(0.35 g, 85%); white waxy solid. R_f = 0.13 ($CHCl_3$/MeOH/NH_3 95/5/1); mp 125.5°C; ^{1}H NMR (500 MHz, $CDCl_3$, 40°C): δ (ppm) 0.89 (t, J = 6.9 Hz, 3H, –C*H*$_3$), 1.14-1.32 (m, 52H, $D_3C(CH_2)_{14}$–, $H_3C(CH_2)_{12}$–), 1.45-1.52 (m, 1H, –C*H*H′CHOH–), 1.61-1.67 (m, 2H, –C*H*$_2$CH$_2$CONH–), 1.76-1.82 (m, 1H, –CH*H*′CHOH–, overlaid by H_2O), 2.24 (t, J = 7.6 Hz, 2H, –C*H*$_2$CONH–), 3.58 (dd, J = 6.7 Hz, J = 2.9 Hz, 1H, –CH_2CHOHC*H*OH–), 3.61-3.65 (m, 1H, –CH_2C*H*OH–), 3.75 (dd, J = 11.6 Hz, J = 5.3 Hz, 1H, –C*H*H′OH), 3.94 (dd, J = 11.5 Hz, J = 2.4 Hz, 1H, –CH*H*′OH), 4.12-4.16 (m, 1H, –C*H*–CH_2OH), 6.30 (d, J = 7.2 Hz, 1H, –CON*H*–); ^{13}C NMR (125 MHz, $CDCl_3$/CD_3OD, 45°C): δ (ppm) 13.6, 22.1, 22.4, 25.6, 29.1-29.4, 31.6, 31.7, 32.8, 36.3, 51.9, 61.1, 72.4, 75.5, 174.5; ESI MS (*m/z*): 609.5 [*M* + Na]$^+$, 585.6 [*M* − H]$^-$; HR MS (*m/z*) [M + H]$^+$ $C_{36}H_{71}D_3NO_4$: calculated 587.5801; found 587.5790; [M + Na]$^+$ $C_{36}H_{70}D_3NO_4Na$: calculated 609.5620; found 609.5606; HPLC ELSD purity >99.5%.

3.5.8 | *N*-[(2*S*,3*S*,4*R*)-1,3,4-Trihydroxyoctadec-2-yl]-22,22,22-d_3-docosanamid (IIb)

Compound **15b** (200 mg, 0.58 mmol) was converted into the product **IIb** (0.34 g, 91%); white waxy solid; R_f = 0.14 ($CHCl_3$/MeOH/NH_3 95/5/1); mp 120°C; ^{1}H NMR (400 MHz, $CDCl_3$, 40°C): δ (ppm) 0.89 (t, J = 6.7 Hz, 3H, –C*H*$_3$), 1.27-1.35 (m, 60H, $D_3C(CH_2)_{18}$–, $H_3C(CH_2)_{12}$–), 1.48-1.53 (m, 1H, –C*H*H′CHOH–), 1.61-1.67 (m, 2H, –C*H*$_2$CH$_2$CONH–), 1.75-1.871 (m, 1H, –CH*H*′CHOH–, overlaid by H_2O), 2.24 (t, J = 7.6 Hz, 2H, –C*H*$_2$CONH–), 3.59 (dd, J = 6.8 Hz, J = 2.8 Hz, 1H, –CH_2CHOHC*H*OH–), 3.62-3.66 (m, 1H, –CH_2C*H*OH–), 3.76 (dd, J = 11.5 Hz, J = 4.9 Hz, 1H, –C*H*H′OH), 3.94 (dd, J = 11.4 Hz, J = 2.0 Hz, 1H, –CH*H*′OH), 4.12-4.16 (m, 1H, –C*H*–CH_2OH), 6.32 (d, J = 5.9 Hz, 1H, –CON*H*–); ^{13}C NMR (100 MHz, $CDCl_3$/CD_3OD, 40°C): δ (ppm) 13.8, 22.3, 22.6, 25.7, 25.8, 29.1-29.4, 31.8, 31.9, 33.0, 36.5, 52.0, 61.2, 72.5, 75.7, 174.6; ESI MS (*m/z*): 665.6 [*M* + Na]$^+$, 641.7 [*M* − H]$^-$; HR MS (*m/z*) [M + H]$^+$ $C_{40}H_{79}D_3NO_4$: calculated 643.6427; found 643.6401; HPLC ELSD purity >99.5%.

3.5.9 | *N*-[(2*S*,3*S*,4*R*)-1,3,4-Trihydroxyoctadec-2-yl]-24,24,24-d_3-tetracosanamid (IIc)

Compound **15c** (200 mg, 0.54 mmol) was converted into the product **IIc** (0.32 g, 89%); white waxy solid, R_f = 0.15 ($CHCl_3$/MeOH/NH_3 95/5/1); mp 114°C; ^{1}H NMR (500 MHz, $CDCl_3$, 40°C): δ (ppm) 0.89 (t, J = 6.9 Hz, 3H, –C*H*$_3$), 1.14-1.32 (m, 64H, $D_3C(CH_2)_{20}$–, $H_3C(CH_2)_{12}$–), 1.44-1.56 (m, 1H, –C*H*H′CHOH–), 1.61-1.67 (m, 2H, –C*H*$_2$CH$_2$CONH–), 1.76-1.82 (m, 1H, –CH*H*′CHOH–), 2.34 (t, J = 7.4 Hz, 2H, –C*H*$_2$CONH–), 3.58 (dd, J = 7.0 Hz, J = 2.6 Hz, 1H, –CH_2CHOHC*H*OH–), 3.61-3.65 (m, 1H, –CH_2C*H*OH–), 3.76 (dd, J = 11.5 Hz, J = 5.3 Hz, 1H, –C*H*H′OH), 3.95 (dd, J = 11.6 Hz, J = 2.5 Hz, 1H, –CH*H*′OH), 4.12-4.16 (m, 1H, –C*H*–CH_2OH), 6.29 (d, J = 5.7 Hz, 1H, –CON*H*–); ^{13}C NMR (125 MHz, $CDCl_3$/CD_3OD, 40°C): δ (ppm) 13.6, 22.2, 22.5, 25.6, 25.7 29.1-29.6, 31.6, 31.7, 32.7, 36.3, 51.9, 61.1, 72.4, 75.4, 174.6; ESI MS (*m/z*): 693.7 [*M* + Na]$^+$, 669.8 [*M* − H]$^-$; HR MS (*m/z*) [M + H]$^+$ $C_{42}H_{83}D_3NO_4$: calculated 671.6740; found 671.6733; HPLC ELSD purity >99.5%.

3.5.10 | *N*-[(2*S*,3*S*,4*R*)-1,3,4-Trihydroxyoctadec-2-yl]-d_{23}-dodecanamid (IId)

Lauric acid-d_{23} (**15d**) (100 mg, 0.34 mmol) was converted into the product **IId** (0.19 g, 82%); white waxy solid; R_f = 0.20 ($CHCl_3$/MeOH/NH_3 95/5/1); mp 117°C; ^{1}H NMR (500 MHz, $CDCl_3$, 40°C): δ (ppm) 0.89 (t, J = 6.8 Hz, 3H, –C*H*$_3$), 1.20-1.37 (m, 24H, $H_3C(CH_2)_{12}$–), 1.44-1.55 (m, 1H, –C*H*H′CHOH–), 1.73-1.80 (m, 1H, –CH*H*′CHOH–), 3.58 (dd, J = 6.7 Hz, J = 2.9 Hz, 1H, –CH_2CHOHC*H*OH–), 3.60-3.67 (m, 1H, –CH_2C*H*OH–), 3.75 (dd, J = 11.5 Hz, J = 5.3 Hz, 1H, –C*H*H′OH), 3.94 (dd, J = 11.5 Hz, J = 2.5 Hz, 1H, –CH*H*′OH), 4.10-4.14 (m, 1H, –C*H*–CH_2OH), 6.29 (d, J = 7.0 Hz, 1H, –CON*H*–); ^{13}C NMR (125 MHz, $CDCl_3$, 27°C): δ (ppm) 14.3, 23.1, 26.3, 29.8-30.2, 32.4, 33.2, 52.5, 61.6, 72.9, 75.9, 175.4; ESI MS (*m/z*): 521.7 [*M* − H]$^-$, 1043.8 [2*M* − H]$^-$; HR MS (*m/z*) [M + H]$^+$ $C_{30}H_{39}D_{23}NO_4$: calculated 523.6117; found 523.6118; [M + Na]$^+$ $C_{30}H_{38}D_{23}NO_4Na$: calculated 545.5937; found 545.5936; HPLC ELSD purity >99.5%.

3.5.11 | *N*-[(2*S*,3*S*,4*R*)-1,3,4-Trihydroxyoctadec-2-yl]-d_{35}-octadecanamid (IIe)

Stearic acid-d_{35} (**15e**) (100 mg, 0.31 mmol) was converted into the product **IIe** (0.16 g, 81%); white waxy solid; R_f = 0.25 ($CHCl_3$/MeOH/NH_3 95/5/1); mp 121°C; ^{1}H NMR (500 MHz, $CDCl_3$, 40°C): δ (ppm) 0.89 (t, J = 6.8 Hz, 3H, –C*H*$_3$), 1.26-1.34 (m, 24H, $H_3C(CH_2)_{12}$–), 1.46-1.53 (m, 1H, –C*H*H′CHOH–), 1.73-1.80 (m, 1H, –CH*H*′CHOH–, overlaid by H_2O), 3.58 (dd, J = 6.7 Hz, J = 2.9 Hz, 1H, –CH_2CHOHC*H*OH–), 3.61-3.66 (m, 1H, –CH_2C*H*OH–), 3.75 (dd, J = 11.5 Hz, J = 5.2 Hz, 1H, –C*H*H′OH), 3.93 (dd, J = 11.7 Hz, J = 1.9 Hz, 1H, –CH*H*′OH), 4.11-4.15 (m, 1H, –C*H*–CH_2OH), 6.41 (d, J = 5.7 Hz, 1H, –CON*H*–);^{13}C

NMR (125 MHz, THF-d_8, 40°C): δ (ppm) 13.4, 22.5, 25.8, 29.3-29.8, 31.8, 33.2, 52.8, 61.5, 72.2, 76.2, 172.3; ESI MS (*m/z*): 641.7 $[M + Na]^+$, 1259.8 $[2M + Na]^+$, 617.8 $[M - H]^-$; HR MS (m/z) $[M + H]^+$ $C_{36}H_{29}D_{35}NO_5$: calculated 619.7809; found 619.7790; HPLC ELSD purity >98%.

3.5.12 | *N*-[(2*S*,3*S*,4*R*)-1,3,4-Trihydroxyoctadec-2-yl]-d_{47}-tetracosanamid (IIf)

Tetracosanoic acid-d_{47} (**15f**) (100 mg, 0.24 mmol) was converted into the product **IIf** (0.15 g, 84%); white waxy solid; R_f = 0.29 ($CHCl_3$/MeOH/NH_3 95/5/1); mp 114°C; ^{1}H NMR (500 MHz, $CDCl_3$, 40°C): δ (ppm) 0.89 (t, *J* = 6.7 Hz, 3H, –C***H***$_3$), 1.20-1.35 (m, 24H, $H_3C(CH_2)_{12}$–), 1.42-1.59 (m, 1H, –C***H***H′CHOH–), 1.75-1.81 (m, 1H, –CH***H***′CHOH–), 3.58 (dd, *J* = 6.6 Hz, *J* = 2.9 Hz, 1H, –CH_2CHOHC***H***OH–), 3.60-3.66 (m, 1H, –CH_2C***H***OH–), 3.76 (dd, *J* = 11.5 Hz, *J* = 5.3 Hz, 1H, –C***H***H′OH), 3.95 (dd, *J* = 11.4 Hz, *J* = 2.2 Hz, 1H, –CH***H***′OH), 4.11-4.16 (m, 1H, –C***H***–CH_2OH), 6.28 (d, *J* = 8.0 Hz, 1 H, –CON***H***–); ^{13}C NMR (125 MHz, THF-d_8, 35°C): δ (ppm) 14.6, 23.7, 27.0, 29.1-31.0, 33.0, 34.5, 54.0, 62.7, 73.4, 77.4, 173.5; ESI MS (*m/z*): 736.8 $[C_{42}H_{40}D_{46}NO_4 + Na]^+$, 712.9 $[C_{42}H_{40}D_{46}NO_4 - H]^-$; HR MS (*m/z*) $[M + H]^+$ $C_{42}H_{40}D_{46}NO_4$: calculated 714.9439; found 714.9431; HPLC ELSD purity >99.5%.

4 | CONCLUSION

Six ceramides [NS] and six ceramides [NP] with perdeuterated and specifically deuterated fatty acids in the ω position were synthesized. While for the syntheses of the perdeuterated fatty acids literature methods were used, the specifically deuterated acids were prepared for the first time by a copper-catalyzed Grignard coupling of 2 suitably functionalized bromides. For the reaction of the deuterated fatty acids with the sphingoid bases, PyBOP® was successfully used as the activator, and the corresponding ceramides could be isolated in high yields. The specifically deuterated ceramides will be used in neutron scattering experiments and the perdeuterated specimen in solid-state NMR investigations to get insight in ceramide arrangement in the SC.

ACKNOWLEDGEMENTS

The study was supported by the Deutsche Forschungsgemeinschaft (DFG DO 463/6-1and NE 427/30-1) and Evonik Industries, AG. The authors thank Dr. Christian Ihling and Prof. Dr. Andrea Sinz (Institute of Pharmacy, Martin-Luther-University, Halle (Saale), Germany) for obtaining the HRMS data and Manuela Woigk (Institute of Pharmacy, Martin-Luther-University, Halle (Saale), Germany) for the HPLC data.

CONFLICT OF INTEREST

There is no conflict of interest.

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How to cite this article: Sonnenberger, S., Lange, S., Langner, A., Neubert, R. H. H., and Dobner, B. (2016) Synthesis of ceramides NS and NP with perdeuterated and specifically ω deuterated *N*-acyl residues. *J. Label Compd. Radiopharm*, doi: 10.1002/jlcr.3443