

SUPPORTING MATERIAL for**ACYL-CHAIN METHYL DISTRIBUTIONS OF LIQUID-ORDERED AND -
DISORDERED MEMBRANES**

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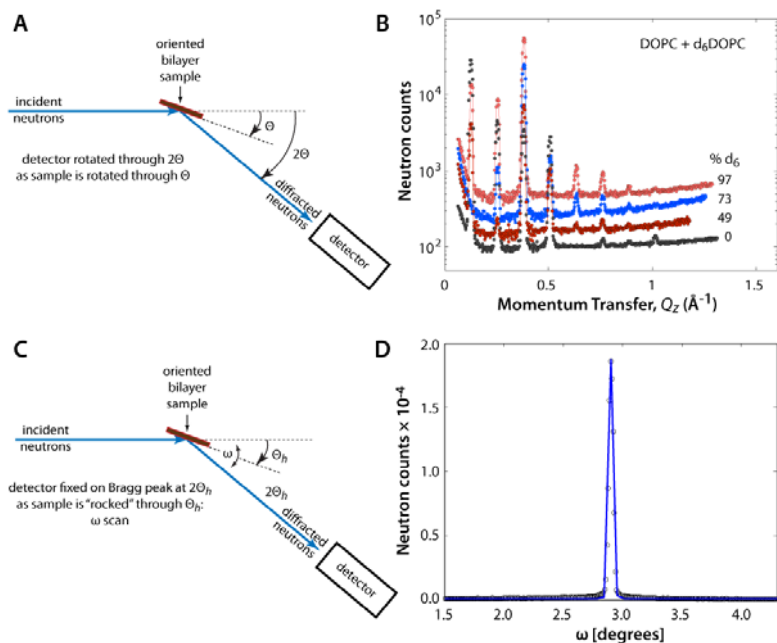


Figure S1. Diffraction measurements on oriented DOPC membranes. A. Schematic of the scattering geometry for the specular Θ - 2Θ mode of data collection. While sample is rotated through angle Θ relative to the incident neutron beam, the signal is collected at 2Θ . Bragg peaks occur at Θ_h . B. Diffraction data collected in the θ - 2θ mode for DOPC lipid multilayers containing various mole fractions (0%, 49%, 73% and 97%) of terminal-methyl deuterated DOPC (d_6 -DOPC). Data collected at 66% relative humidity (RH) and 21° C. For clarity, the upper three curves have been shifted from the original baselines. C. Schematic representation of the scattering geometry for determining the mosaic spread of the sample using rocking curves. The detection angle is fixed at a Bragg peak of Θ_h while sample is rotated in the beam through an angle ω . D. Rocking curve for a DOPC sample measured at Θ_1 at 66% RH (black circles). A Gaussian model for peak shape has been fitted to the data (blue line). The curve in D is presented as collected. No background was subtracted. Our samples are only 2000-3000 bilayers thick and we typically observe such flat backgrounds in rocking curves, combined with a low mosaic spread of less than 0.1 degrees.

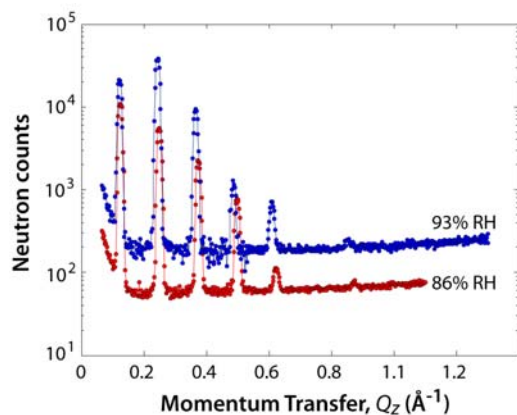


Figure S2. Diffracted intensities for DOPC multilayers measured at 21° C for 86% and 93% RH. Data were collected in the Θ -2 Θ mode.

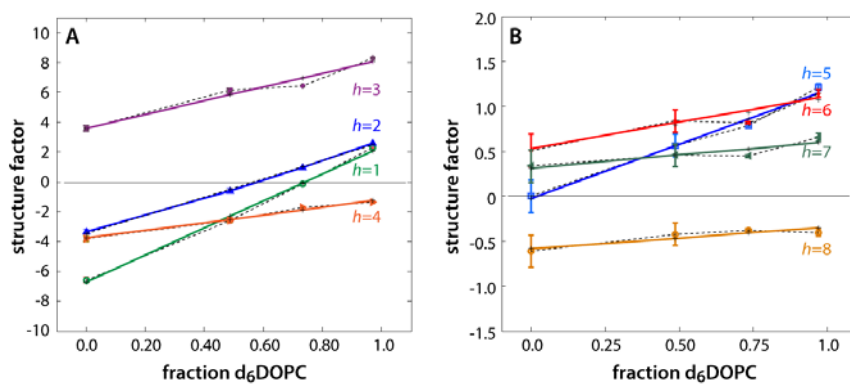


Figure S3. Neutron structure factors for DOPC bilayers as a function of mole fraction d_6 -DOPC. A. Diffraction orders $h = 1 - 4$. B. Diffraction orders $h = 5 - 8$. The structure factors for each sample composition were initially placed on the absolute per-lipid scale, based on the H_2O - D_2O exchange procedure. The scaled structure factors show a linear increase with the amount of deuterium (d_6 -DOPC), as required by symmetry. Average values of structure factors at each d_6 -DOPC fraction were determined by fitting straight lines to each data set (solid lines). These structure factors were used to construct the bilayer profiles. Only data determined at 66% RH are illustrated; similar analyses were performed for other hydrations (Table S2).

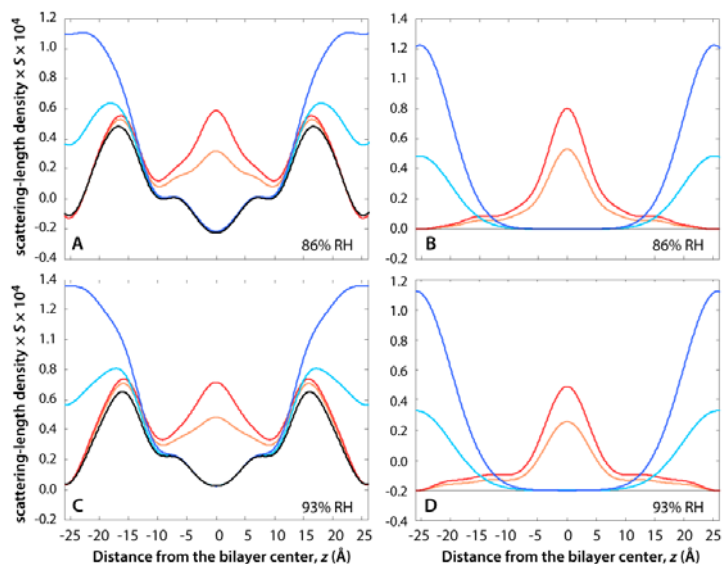


Figure S4. Transbilayer terminal methyl and water distributions for DOPC: d_6 DOPC bilayers at 86% and 93% RH. A. The bilayer scattering-length density distributions for several DOPC: d_6 -DOPC compositions at 86% RH. The terminal methyl distributions are shown in black (DOPC alone), orange (51:49 DOPC: d_6 -DOPC), and red (27:73 DOPC: d_6 DOPC). The water distributions are shown in cyan (80:20 H_2O : D_2O) and blue (50:50 H_2O : D_2O). B. Difference profiles for methyls and water at 86% RH. These are obtained by subtracting the DOPC profile (black) in panel A from the DOPC: d_6 -DOPC profiles. C. Same as in panel A, but at 93% RH. D. Same as in Panel B, but at 93% RH. The absolute per-lipid structure factors used to construct these profiles are shown in Table S2.

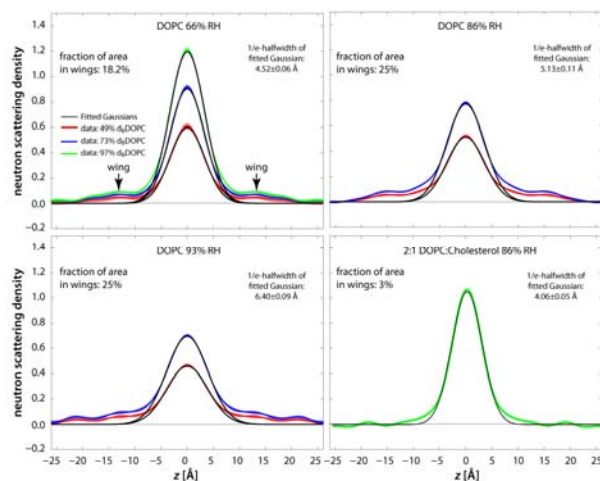


Figure S5. Determination of the fraction of methyl groups in contact with the hydrated headgroups. The fraction is taken as the area in the wings of the distribution compared to the total area. The areas of the wings were determined by subtracting the areas of Gaussians fitted to the central part of the distributions (black curves) from total areas. The DOPC: d_6 -DOPC ratios are indicated by the colors of the curves according to the key in upper left-hand panel. The hydrations and overall lipid compositions are indicated at the tops of the panels. The computed areas of the wings are indicated in each panel.

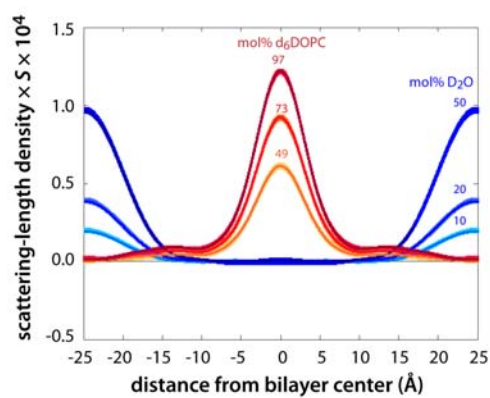


Figure S6. The same data as in Figure 1A replotted to indicate the error bands. The terminal methyl (red bands) and water distribution (blue bands), determined for different DOPC: d_6 -DOPC molar ratios (from bottom to top: 51:49, 27:73 and 3:97), and, respectively, DOPC measured in different H_2O/D_2O ratios (from bottom to top: 90:10, 80:20 and 50:50). Profiles shown with uncertainty bands calculated based on the uncertainties in the structure factors (Table S2).

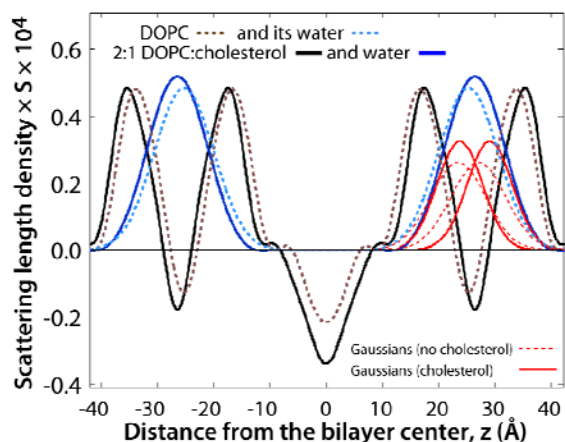


Figure S7: Bilayer profiles in the absence and presence of cholesterol. Scattering-length density profiles are shown for DOPC (dashed brown curve) and 2:1 DOPC:cholesterol (solid black curve) along with the associated water distributions (dashed blue curve for DOPC; solid blue curve for 2:1 DOPC:cholesterol). A significant increase in the Bragg spacing and an increase in the depth of the terminal-methyl trough are clearly observable for samples containing cholesterol. Although the overall width of the water distribution does not change much, Gaussian-model fitting and base width both show a narrower water distribution in the presence of cholesterol. The water distributions can best be described by two Gaussians overlapping in the space between two opposed bilayers (red curves), with widths and positions given in Table S1.

Table S1. Parameters characterizing the bilayer unit cells and the water distribution parameters.

RH (%)	waters/lipid	d [Å]	A_w [Å]	Z_w [Å]
66	5.4	49.6 ± 0.08	5.7 ± 0.6	22.7 ± 0.6
86	7.7	50.4 ± 0.11	6.9 ± 0.4	23.4 ± 0.5
86□(with cholesterol)	$7.9 \pm 0.2^*$	52.7 ± 0.20	5.7 ± 0.5	23.7 ± 0.5
93	9.4	51.6 ± 0.10	7.8 ± 0.7	23.7 ± 0.6

d is the repeat spacing, i.e. bilayer thickness including the water of hydration.

A_w is 1/e half-width of the water distribution

Z_w the mean positions of the water envelopes located at $\pm Z_w$.

♣ The number of water molecules per lipid was estimated, in this case, by using the six deuterium atoms in the terminal methyl as a calibration measure, for samples of known unit cell composition, and subsequent scaling of the water peak at that composition. The other values for the number of waters per lipid were taken from literature [1,2,3].

The water distribution parameters were determined by fitting the scaled structure factors (FD-FH) with a set of two overlapping Gaussians, for a best description of the water in the interbilayer space. The standard deviations of the parameters A_w and Z_w , were determined by sampling through a large set of scaled structure factors within their uncertainties [4].

Table S2. Scaled structure factors and uncertainties for DOPC:d₆-DOPC. The uncertainties in the scaled structure factors contain, conservatively, counting statistics and uncertainties associated with scaling and other structure factor corrections (Figure S3). n.d. means 'not determined'.

66% Relative Humidity					
<i>h</i>	DOPC (H ₂ O)	49% d ₆ -DOPC (H ₂ O)	73% d ₆ -DOPC (H ₂ O)	97% d ₆ -DOPC (H ₂ O)	DOPC 20:80 D ₂ O:H ₂ O
1	-6.733 ± 0.287	-2.302 ± 0.126	-0.050 ± 0.014	2.111 ± 0.039	-10.521 ± 0.629
2	-3.447 ± 0.147	-0.449 ± 0.049	1.075 ± 0.034	2.537 ± 0.045	-1.183 ± 0.068
3	3.576 ± 0.157	5.884 ± 0.297	7.057 ± 0.203	8.183 ± 0.137	2.616 ± 0.158
4	-3.772 ± 0.166	-2.521 ± 0.125	-1.885 ± 0.058	-1.275 ± 0.027	-3.483 ± 0.219
5	-0.024 ± 0.171	0.573 ± 0.054	0.877 ± 0.030	1.168 ± 0.033	0.000 ± 0.120
6	0.501 ± 0.041	0.798 ± 0.071	0.949 ± 0.038	1.093 ± 0.037	0.501 ± 0.042
7	0.313 ± 0.053	0.461 ± 0.071	0.536 ± 0.059	0.608 ± 0.046	0.313 ± 0.068
8	-0.445 ± 0.042	-0.361 ± 0.111	-0.318 ± 0.069	-0.278 ± 0.066	-0.445 ± 0.050
86% Relative Humidity					
1	-4.340 ± 0.206	-0.058 ± 0.026	2.119 ± 0.023	n.d.	-9.506 ± 0.103
2	-4.579 ± 0.204	-2.262 ± 0.061	-1.084 ± 0.023	n.d.	-1.879 ± 0.103
3	3.638 ± 0.159	5.432 ± 0.147	6.344 ± 0.076	n.d.	2.722 ± 0.081
4	-2.369 ± 0.107	-1.437 ± 0.043	-0.964 ± 0.050	n.d.	-2.167 ± 0.055
5	-0.754 ± 0.037	-0.274 ± 0.129	-0.000 ± 0.088	n.d.	-0.783 ± 0.026
6	0.035 ± 0.112	0.356 ± 0.097	0.519 ± 0.036	n.d.	0.000 ± 0.051
7	0.342 ± 0.062	0.517 ± 0.076	0.606 ± 0.045	n.d.	0.342 ± 0.052
8	-0.000 ± 0.093	-0.000 ± 0.229	-0.000 ± 0.065	n.d.	0.000 ± 0.158

Table S2, cont'd

93% Relative Humidity					
1	-2.927 ± 0.182	0.787 ± 0.028	2.674 ± 0.062	n.d.	-8.968 ± 0.242
2	-5.450 ± 0.341	-3.409 ± 0.126	-2.372 ± 0.051	n.d.	-2.673 ± 0.454
3	3.408 ± 0.209	5.151 ± 0.191	6.036 ± 0.129	n.d.	2.648 ± 0.278
4	-1.290 ± 0.084	-0.499 ± 0.045	-0.000 ± 0.091	n.d.	-1.166 ± 0.107
5	-1.064 ± 0.069	-0.651 ± 0.049	-0.440 ± 0.033	n.d.	-1.077 ± 0.094
6	0.000 ± 0.223	0.000 ± 0.159	0.000 ± 0.105	n.d.	0.000 ± 0.066
7	0.458 ± 0.052	0.498 ± 0.051	0.519 ± 0.071	n.d.	0.458 ± 0.056
8	-0.000 ± 0.145	-0.000 ± 0.188	-0.000 ± 0.102	n.d.	0.000 ± 0.073

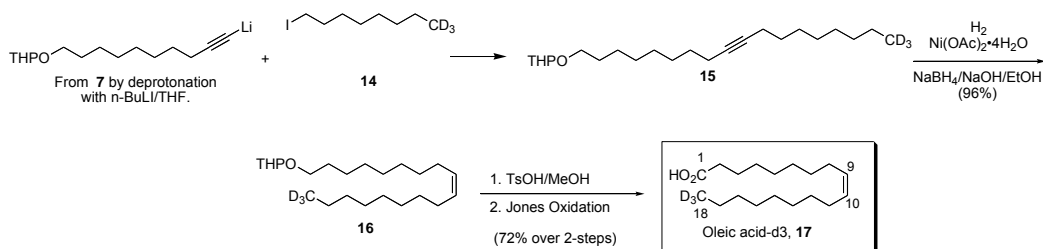
Table S3. Scaled Structure Factors for DOPC/cholesterol bilayers. The uncertainties in the scaled structure factors contain, conservatively, counting statistics and uncertainties associated with scaling and other structure factor corrections (Figure S3). All measurements were made at 86% RH.

<i>h</i>	2:1 DOPC:cholesterol	2:1 d ₆ -DOPC:d ₆ -cholesterol	2:1 DOPC:cholesterol
	H ₂ O	H ₂ O	20:80 D ₂ O:H ₂ O
1	-5.568 ± 0.323	0.514 ± 0.028	-11.154 ± 0.495
2	-5.291 ± 0.307	-2.320 ± 0.096	-1.942 ± 0.112
3	3.421 ± 0.199	10.256 ± 0.411	2.154 ± 0.101
4	-3.048 ± 0.178	0.000 ± 0.186	-2.825 ± 0.144
5	-0.381 ± 0.085	0.000 ± 0.149	-0.382 ± 0.215
6	0.345 ± 0.070	2.107 ± 0.092	0.345 ± 0.093
7	0.403 ± 0.110	0.952 ± 0.077	0.403 ± 0.100
8	-0.584 ± 0.075	-0.912 ± 0.093	-0.610 ± 0.187

Synthesis of Specifically Deuterated Oleic Acid

Oleic acid deuterated exclusively on the terminal methyl group was synthesized on a gram scale by coupling two readily available components: an unlabeled ten carbon lithioalkyne, (Scheme 1) containing both the eventual C1-carboxyl and C9,10-alkene groups of oleic acid in latent form, and a methyl-deuterated 1-iodo-*n*-octane, **14**, which carries the deuterium labeled methyl group destined to become C18 in oleic acid. These two partners react smoothly to give the full-length alkyne **15**, which is then to be reduced to the *cis* (Z) alkene present in the target. This venerable reaction, catalytic semi-hydrogenation of an alkyne, produced unacceptably large amounts (often 5% or more) of the undesired *trans* (E) alkene under most conventional conditions; however, the use of Ni(OAc)₂/H₂/NaBH₄ poisoned with ethylenediamine consistently gave high yields of the Z-alkene contaminated with ~ 0.5% of the E-isomer, **16** (by GC analysis of the corresponding methyl esters). Hydrolysis of the THP ether and Jones oxidation gave 18,18,18-trideuterio-oleic acid; analysis by mass spectrometry and 500Mz proton NMR verified that the terminal (C18) methyl group was fully deuterated (98 ± 1%) and that the alkene was 99.5% Z-isomer.

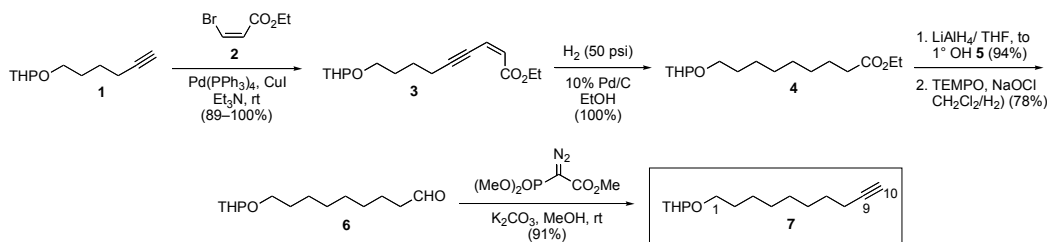
Scheme 1. Synthesis of C18 (terminal methyl)-deuterated oleic acid-d3, **17.**



There are a large number of potential sources of the two coupling precursors, **7** and **14**, employed in this route. We found that the obvious commercially available starting materials, for example the 9-hydroxynonanoic acid parent of **4** (Scheme 2), tend to be expensive and/or difficult to purify while still requiring several protection steps. For both the unlabeled precursor **7** (Scheme 2) and the labeled coupling partner **14** (Scheme 3), we found that the most expeditious route to the respective 10- and 8-carbon building blocks was a somewhat unconventional Pd-catalyzed three carbon homologation of the readily available alkynes **1** and **8**, as shown, followed by hydrogenation of the resultant eneyne. In the former case, the resultant protected nonanoate ester derivative **4** undergoes a further one-carbon homologation via conversion into the aldehyde **6** and direct conversion into the 10-carbon unlabeled alkyne **7**.

Comment [1]:
Stephen H. White Aug 5,
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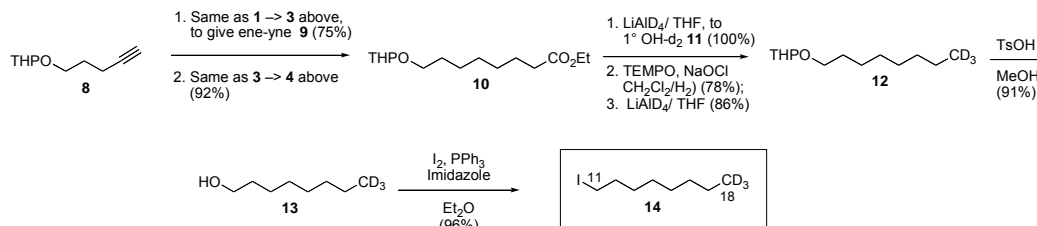
Scheme 2. Preparation of unlabeled C1-C10 precursor, **7.**



Preparation of the coupling partner bearing the deuterium label was carried out by a similar route (Scheme 3) that produced the octanoate ester **10**. The deuterium label was then introduced very efficiently by sequential reduction of the ethyl ester and derived primary alcohol (as the mesylate) with lithium aluminum deuteride, resulting in the efficient conversion of the ester carbonyl carbon into the desired $-CD_3$ that will become the terminal C18 methyl group in oleic acid-d3. This 8-carbon fragment, **12**, containing carbons 11-18 of the target oleic acid, was converted into the iodide **14** as shown in preparation for coupling as described above. Although the minimum theoretical level of deuterium incorporation into this fragment was as low as 94% based on the $LiAlH_4$ label claim of $> 98\%$ D, MS analysis of the final oleic acid product showed that it was in fact higher, at $98 \pm 1\%$.

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Scheme 3. Preparation of labeled C11-C18 precursor **14.**



Experimental Procedures and Compound Characterization

General Experimental Methods. 1H NMR spectra were either recorded at ambient temperature or elevated temperatures at 400 and 500 MHz. ^{13}C NMR spectra were recorded at ambient temperature at 100 and 125 MHz. For 1H NMR spectra acquired in $CDCl_3$, chemical shifts are reported as δ values in ppm and are calibrated according to internal $CHCl_3$ (7.26 ppm). For ^{13}C NMR spectra, chemical shifts are reported as δ values in ppm relative to chloroform. The 1H NMR data are reported as follows: chemical shift in ppm on the δ scale, multiplicity (app = apparent, br = broad, s = singlet, d = doublet, t = triplet, q = quartet, quin = quintet, m = multiplet, coupling constants (Hz), and integration. Infrared spectra (IR) were obtained on an FTIR spectrophotometer and are reported in wavenumbers (cm^{-1}). High-resolution mass spectra were acquired on analytical chemical ionization (CI), electron ionization (EI), or electrospray ionization (ESI) spectrometer and were obtained by peak matching. Analytical thin-layer chromatography (TLC) was performed using 0.25 mm precoated silica gel plates. Liquid chromatography was performed using forced flow (flash chromatography) of the indicated solvent system on silica gel (SiO_2) 60 (200-400) mesh.

All reactions were carried out using flame-dried or oven-dried glassware and inert atmosphere operations were conducted under N_2 (g) or Ar (g) passed through a Drierite drying tube. Anhydrous tetrahydrofuran (THF), methylene chloride (CH_2Cl_2), triethylamine (Et_3N), acetonitrile (CH_3CN), and diethyl ether (Et_2O) were filtered through two columns of activated basic alumina and transferred under Ar (g) according to the method described by Grubbs.⁵ Hexamethylphosphoramide (HMPA) was distilled over calcium hydride (under reduce pressure) and stored over activated 4 Å molecular sieves.⁶ All other commercial reagents were used as received, unless noted otherwise.

6-(Tetrahydro-2H-pyran-2-yloxy)-1-hexyne, 1. To a solution of 5-hexyn-1-ol (10.4 g, 106 mmol) in CH₂Cl₂ (80 mL) was introduced dihydropyran (15.0 mL, 164 mmol), followed by pyridinium *p*toluenesulfonic acid (0.560 g, 2.23 mmol). After 2 h, the reaction was poured into saturated aqueous NaHCO₃ (50 mL), extracted with CH₂Cl₂ (3 x 100 mL), dried over MgSO₄, concentrated *in vacuo*. The resultant yellow oil was purified by flash chromatography (10:90 EtOAc:hexanes) to provide the alkyne **1** as a colorless oil (17.8 g, 93%): ¹H NMR (500 MHz, CDCl₃) δ 4.57 (app. t, *J* = 3.5, 1H), 3.87–3.82 (m, 1H), 3.75 (dt, *J* = 9.6, 6.9, 1H), 3.50–3.45 (m, 1H), 3.41 (dt, *J* = 9.6, 6.0, 1H), 2.22 (td, *J* = 7.1, 2.6, 2H), 1.94 (t, *J* = 2.6, 1H), 1.85–1.79 (m, 1H), 1.73–1.67 (m, 3H), 1.65–1.50 (m, 6H); ¹³C NMR (125 MHz, CDCl₃) δ 99.2, 84.2, 68.8, 67.3, 62.8, 31.2, 29.2, 25.9, 25.8, 20.0, 18.7; IR (thin film) 3296, 2942, 2869, 2117, 1137, 1121 cm⁻¹; HRMS (ESI/MeOH) *m/z* calculated for C₁₁H₁₈O₂ (M+Na)⁺ 205.1205, found 205.1207.

(Z)-Ethyl-3-bromoacrylate, 2.⁷ To anhydrous LiBr (12.4 g, 144 mmol) was added CH₃CN (180 mL) and glacial AcOH (8.20 mL, 143 mmol). Ethyl propiolate (11.0 mL, 108 mmol) was introduced and the reaction mixture was heated to reflux. After 1.5 h, the mixture was cooled to room temperature and diluted with H₂O (30 mL). Solid K₂CO₃ was added in portions until a pH of 7 was obtained. The reaction mixture was extracted with Et₂O (3 x 75 mL), dried over MgSO₄, concentrated *in vacuo* to provide a light yellow oil. The resultant oil was purified by distillation (30 mmHg, 82–85 °C) to yield the exclusively the *Z*-vinylbromide **2** as a colorless oil (15.9 g, 80%): ¹H NMR (500 MHz, CDCl₃) δ 6.99 (d, *J* = 8.3, 1H), 6.60 (d, *J* = 8.3, 1H), 4.24 (q, *J* = 7.1, 2H), 1.31 (q, *J* = 7.1, 3H); ¹³C NMR (125 MHz, CDCl₃) δ 164.4, 125.0, 121.5, 61.2, 14.6; IR (thin film) 3078, 2983, 1731, 1615 cm⁻¹; HRMS (ESI/MeOH) *m/z* calcd for C₅H₇BrO₂ (M+Na)⁺ 200.9527, found 200.9536.

(Z)-ethyl 9-(tetrahydro-2H-pyran-2-yloxy)non-2-en-4-ynoate, 3. To Pd(PPh₃)₄ (0.648 g, 0.561 mmol) and Cul (0.215 g, 1.13 mmol) was added Et₃N (150 mL) under an argon atmosphere. The resultant suspension was stirred vigorously for 2 min or until a homogenous reaction mixture was attained, followed by slow addition of vinyl bromide **2** (10.5 g, 58.4 mmol) in Et₃N (25 mL). After 5 min, a solution of alkyne **1** (10.5 g, 57.4 mmol) in Et₃N (25 mL) was added dropwise. Reaction mixture was aged for 24 h, filtered over a pad of Celite, and the filter cake was washed with hexanes (5 x 100 mL). The filtrate was stirred over saturated aqueous NH₄Cl (50 mL), extracted with CH₂Cl₂ (3 x 500 mL), and concentrated *in vacuo* to yield a thick brown oil. The resultant oil was purified by flash chromatography (8:92 EtOAc:hexanes) to yield enyne **3** as a yellow oil (15.9 g, 99%): ¹H NMR (500 MHz, CDCl₃) δ 6.14 (dt, *J* = 11.4, 2.4, 1H), 6.01 (d, *J* = 11.4, 1H), 4.57 (dd, *J* = 4.3, 2.9, 1H), 4.21 (q, *J* = 7.1, 2H), 3.87 (ddd, *J* = 11.2, 7.9, 3.3, 1H), 3.75 (dt, *J* = 9.6, 6.9, 1H), 3.51–3.46 (m, 1H), 3.41 (dt, *J* = 9.8, 6.1, 1H), 2.48 (td, *J* = 6.8, 2.4, 2H), 1.85–1.64 (m, 6H), 1.60–1.49 (m, 4H), 1.29 (t, *J* = 7.1, 3H); ¹³C NMR (125 MHz, CDCl₃) δ 165.3, 128.0, 124.3, 104.1, 99.2, 78.3, 67.3, 62.7, 60.7, 31.2, 29.4, 25.9, 25.7, 20.4, 20.1, 14.7; IR (thin film) 2941, 2869, 2248, 2207, 1725, 1708, 1609, 1183 cm⁻¹; HRMS (ESI/MeOH) *m/z* calcd for C₁₆H₂₄O₄ (M+Na)⁺ 303.1372, found 303.1562.

Ethyl 9-(tetrahydro-2H-pyran-2-yloxy)nonanoate, 4. To the enyne **3** (10.1 g, 36.0 mmol) was added anhydrous EtOH (30 mL) in a heavy walled Parr bottle. Under a constant atmosphere of N₂ (g), Pd/C (10% Pd content, 0.567 g, 0.533 mmol) was added to the reaction mixture and the suspension was subjected to three cycles of evacuation (30 sec) followed by purging with N₂ (g). The Parr bottle was transferred to a Parr hydrogenator and H₂ (50 atm) was introduced for 15 min. The reaction was purged under N₂, filtered over Celite, the filter cake was washed with CH₂Cl₂ (3 x 50 mL), and concentrated *in vacuo* to provide the saturated ester **4** as a colorless oil (10.3 g, 100%): ¹H NMR (500 MHz, CDCl₃) δ 4.56 (dd, *J* = 4.3, 2.8, 1H), 4.12 (q, *J* = 7.1, 2H), 3.87 (ddd, *J* = 11.1, 7.7, 3.3, 1H), 3.72 (dt, *J* = 9.6, 6.9, 1H), 3.51–3.47 (m, 1H), 3.37 (dt, *J* = 9.6, 6.7, 1H), 2.28 (app. t, *J* = 7.6, 2H), 1.85–1.77 (m, 1H), 1.73–1.68 (m, 1H), 1.63–1.49 (m, 8H), 1.35–1.29 (m, 8H), 1.24 (t, *J* = 7.1, 3H); ¹³C NMR (125 MHz, CDCl₃) δ 173.8,

98.8, 67.5, 62.3, 60.1, 34.3, 30.7, 29.6, 29.2, 29.1, 29.0, 26.1, 25.4, 24.9, 19.6, 14.2; IR (thin film) 2935, 2856, 1736 cm^{-1} ; HRMS (ESI/MeOH) m/z calcd for $\text{C}_{16}\text{H}_{30}\text{O}_4$ ($\text{M}+\text{Na}$)⁺ 309.2042, found 309.2029.

9-(Tetrahydro-2H-pyran-2-yloxy)nonan-1-ol, 5. To a cooled (0 °C) suspension of LiAlH_4 (0.570 g, 15.0 mmol) in THF (10 mL) was added a solution of ethyl ester **4** (4.00 g, 14.0 mmol) in THF (10 mL) over 10 min. Extra THF (2 x 5 mL) was used to aid in transfer of the ester. The reaction suspension was allowed to warm to ambient temperature. After 24 h, the reaction suspension was cooled (0 °C), EtOAc (5 mL) was added, followed by slow addition of 10% KOH (aq) (9 mL) over 15 min (*Caution: vigorous evolution of H_2 gas*). The resultant white suspension was warm to ambient temperatures (over 1 h), filtered over a pad of Celite, washed with hot EtOAc (3 x 20 mL), and concentrated *in vacuo* to a clear oil. The oil was purified by flash chromatography (30:70 EtOAc:hexanes) to the yield alcohol **5** as a clear oil (8.50 g, 100 %): ^1H NMR (500 MHz, CDCl_3) δ 4.57 (dd, $J = 4.3, 2.8, 1\text{H}$), 3.87 (ddd, $J = 11.2, 7.7, 3.3, 1\text{H}$), 3.72 (dt, $J = 9.6, 6.9, 1\text{H}$), 3.63 (t, $J = 6.7, 2\text{H}$), 3.52–3.47 (m, 1H), 3.37 (dt, $J = 9.6, 6.7, 1\text{H}$), 1.85–1.80 (m, 1H), 1.73–1.69 (m, 1H), 1.63–1.46 (m, 9H), 1.38–1.29 (m, 10H); ^{13}C NMR (125 MHz, CDCl_3) δ 99.3, 68.1, 63.5, 62.8, 33.2, 31.2, 30.2, 30.0, 29.9, 29.8, 26.6, 26.1, 25.9, 20.1; IR (thin film) 3406, 2929, 2855, 1032 cm^{-1} ; HRMS (ESI/MeOH) m/z calcd for $\text{C}_{14}\text{H}_{28}\text{O}_3$ ($\text{M}+\text{Na}$)⁺ 267.1936, found 267.1927.

9-(Tetrahydro-2H-pyran-2-yloxy)nonanal, 6. To a cooled (–10 °C) solution of alcohol **5** (3.21 g, 13.1 mmol) in CH_2Cl_2 (20 mL) was added TEMPO (0.023 g, 0.147 mmol) and KBr (0.124 g, 1.04 mmol). The mixture was stirred vigorously and a solution of NaHCO_3 (4.16 g, 49.5 mmol) and Chlorox® (21 mL, 14.9 mmol, 0.71 M in NaOCl) diluted in H_2O (15 mL) was added dropwise over 20 min. The reaction temperature was maintained between –10 °C to 0 °C during the course of the reaction. The reaction mixture was extracted with CH_2Cl_2 (3 x 20 mL). The combined organic phase was successively washed with 10% aqueous potassium iodide (10 mL), saturated sodium thiosulfate (10 mL), brine (20 mL), dried over MgSO_4 , and concentrated *in vacuo* to give a pink oil. The resultant oil was purified by flash chromatography (10:70 EtOAc:hexanes) to yield the aldehyde **6** as a clear oil (2.46 g, 78%): ^1H NMR (500 MHz, CDCl_3) δ 9.75 (t, $J = 1.8, 1\text{H}$), 4.57 (dd, $J = 4.3, 2.8, 1\text{H}$), 3.87 (ddd, $J = 11.1, 7.8, 3.2, 1\text{H}$), 3.72 (dt, $J = 9.6, 6.9, 1\text{H}$), 3.51–3.47 (m, 1H), 3.37 (dt, $J = 9.6, 6.7, 1\text{H}$), 2.41 (td, $J = 7.3, 1.8, 2\text{H}$), 1.85–1.80 (m, 2H), 1.73–1.68 (m, 2H), 1.63–1.50 (m, 6H), 1.35–1.30 (m, 10H); ^{13}C NMR (125 MHz, CDCl_3) δ 203.4, 99.3, 68.1, 62.8, 44.3, 31.2, 30.1, 29.7 (2), 29.5, 26.6, 25.9, 22.5, 20.2; IR (thin film) 2934, 2856, 1735, 1709, 1023 cm^{-1} ; HRMS (ESI/MeOH) m/z calcd for $\text{C}_{14}\text{H}_{28}\text{O}_3$ ($\text{M}+\text{Na}$)⁺ 265.1780, found 265.1773.

10-(Tetrahydro-2H-pyran-2-yloxy)-1-decyne, 7. To aldehyde **6** (2.27 g, 9.37 mmol) in MeOH (45 mL) was added K_2CO_3 (2.68 g, 19.4 mmol) followed by dropwise addition of the Ohira reagent (2.04 g, 10.6 mmol) over 5 min. After 3 h, H_2O (20 mL) and Et_2O (10 mL) was added and stirred vigorously for 15 min. The reaction mixture was subsequently extracted with Et_2O (2 x 20 mL). The combined organic layer was washed with H_2O (3 x 50 mL), brine (50 mL), dried over MgSO_4 , and concentrated *in vacuo* to give an orange oil. The resultant oil was purified by flash chromatography (20:80 EtOAc:hexanes) to provide the alkyne **7** as a clear oil (2.03 g, 91%): ^1H NMR (500 MHz, CDCl_3) δ 4.57 (dd, $J = 4.4, 2.7, 1\text{H}$), 3.87 (ddd, $J = 11.1, 7.6, 3.3, 1\text{H}$), 3.72 (dt, $J = 9.6, 6.9, 1\text{H}$), 3.51–3.47 (m, 1H), 3.37 (dt, $J = 9.6, 6.7, 1\text{H}$), 2.17 (td, $J = 7.1, 2.7, 2\text{H}$), 1.93 (t, $J = 2.6, 1\text{H}$), 1.85–1.80 (m, 1H), 1.73–1.68 (m, 1H), 1.63–1.48 (m, 6H), 1.41–1.29 (m, 10H); ^{13}C NMR (125 MHz, CDCl_3) δ 99.3, 85.2, 68.5, 68.1, 62.8, 31.2, 30.2, 29.8, 29.5, 29.1, 38.9, 26.6, 25.9, 20.2, 18.8; IR (thin film) 3310, 2935, 2856, 2117, 1033 cm^{-1} ; HRMS (ESI/MeOH) m/z calcd for $\text{C}_{15}\text{H}_{26}\text{O}_2$ ($\text{M}+\text{Na}$)⁺ 261.1830, found 261.1833.

5-(Tetrahydro-2H-pyran-2-yloxy)-1-pentyne, 8. To a solution of 4-pentyn-1-ol (10.4 g, 124 mmol) in CH_2Cl_2 (120 mL) was introduced dihydropyran (17.0 mL, 186 mmol), followed by pyridinium *p*-toluenesulfonic acid (0.680 g, 2.71 mmol). After 2 h, the reaction was poured in to

saturated aqueous NaHCO₃ (50 mL), extracted with CH₂Cl₂ (3 x 100 mL), dried over MgSO₄, concentrated *in vacuo*. The resultant yellow oil was purified by flash chromatography (10:90 EtOAc:hexanes) to provide the alkyne **8** as a colorless oil (11.8 g, 56%): ¹H NMR (500 MHz, CDCl₃) δ 4.56 (dd, *J* = 4.2, 2.9, 1H), 3.88–3.79 (m, 1H and dt, *J* = 9.8, 6.3, 1H), 3.52–3.45 (m, 1H and dt, *J* = 9.8, 6.2, 1H), 2.30 (tdd, *J* = 7.0, 2.6, 1.4, 2H), 1.93 (t, *J* = 2.7, 1H), 1.84–1.75 (m, 3H), 1.70 (m, 1H), 1.60–1.50 (m, 4H); ¹³C NMR (125 MHz, CDCl₃) δ 99.0, 84.1, 68.7, 66.0, 62.4, 30.9, 28.9, 25.7, 19.7, 15.6; IR (thin film) 3295, 2942, 2872, 2117, 1137, 1121 cm⁻¹; HRMS (ESI/MeOH) *m/z* calcd for C₁₀H₁₆O₂ (M+Na)⁺ 191.1048, found 191.1040.

To Pd(PPh₃)₄ (0.343 g, 0.297 mmol) and CuI (0.120 g, 0.630 mmol) was added Et₃N (150 mL) under an argon atmosphere. The resultant suspension was stirred vigorously for 2 min or until a homogenous reaction mixture was attained, followed by slow addition of vinyl bromide **2** (10.6 g, 59.3 mmol, >3:1 mixture of *cis:trans-isomers*) in Et₃N (25 mL). After 5 min, a solution of alkyne **8** (10.0 g, 59.4 mmol) in Et₃N (25 mL) was added dropwise. Reaction mixture was aged for 24 h, filtered over a pad of Celite, and the filter cake was washed with hexanes (5 x 30 mL). The filtrate was stirred over saturated aqueous NH₄Cl (50 mL), extracted with CH₂Cl₂ (3 x 500 mL), and concentrated *in vacuo* to yield a thick brown oil. The resultant oil was purified by flash chromatography (8:92 EtOAc:hexanes) to yield enyne **9** a yellow oil (11.8 g, 75%, as an inconsequential mixture of *cis:trans-isomers*); the predominant *Z*-isomer that was produced in a smaller scale reaction was purified for analysis: ¹H NMR (500 MHz, CDCl₃) δ 6.12 (dt, *J* = 11.4, 2.3, 1H), 6.02 (d, *J* = 11.4, 1H), 4.61–4.59 (m, 1H), 4.21 (q, *J* = 7.1, 2H), 3.93–3.81 (m, 2H), 3.53–3.47 (m, 2H), 2.60–2.53 (m, 2H), 1.86 (app. quin, *J* = 6.6, 2H), 1.83–1.78 (m, 1H), 1.73–1.65 (m, 1H), 1.58–1.50 (m, 4H), 1.29 (t, *J* = 7.1, 3H); ¹³C NMR (125 MHz, CDCl₃) δ 165.1, 127.9, 124.0, 103.5, 99.0, 78.1, 66.2, 62.4, 60.5, 30.9, 28.9, 25.7, 19.7, 17.3, 14.5; IR (thin film) 2941, 2871, 2209, 1725, 1609, 1182 cm⁻¹; HRMS (ESI/MeOH) *m/z* calcd for C₁₅H₂₂O₄ (M+Na)⁺ 289.1416, found 289.1422.

Ethyl 8-(tetrahydro-2H-pyran-2-yloxy)octanoate, 10. To the unsaturated ester **9** (10.0 g, 37.7 mmol) was added anhydrous EtOH (45 mL) in a heavy walled Parr bottle. Under a constant atmosphere of N₂ (g), Pd/C (10% Pd content, 0.390 g, 0.367 mmol) was added and the suspension was subjected to three cycles of evacuation (30 sec) followed by purging with N₂ (g). The Parr bottle was transferred to the Parr hydrogenator and H₂ (50 atm) was introduced for 20 min. The reaction was purged under N₂, filtered over Celite, the filter cake was washed with CH₂Cl₂ (3 x 75 mL), and concentrated *in vacuo* to provide a yellow oil. The resultant oil was purified by flash chromatography (20:80 EtOAc:hexanes) to yield saturated ester **10** as a colorless oil (9.51 g, 92%): ¹H NMR (400 MHz, CDCl₃) δ 4.56 (dd, *J* = 4.3, 2.8, 1H), 4.11 (q, *J* = 7.1, 2H), 3.86 (ddd, *J* = 11.0, 8.0, 3.4, 1H), 3.72 (dt, *J* = 9.6, 6.9, 1H), 3.50–3.47 (m, 1H), 3.37 (dt, *J* = 9.6, 6.7, 1H), 2.28 (app. t, *J* = 7.5, 2H), 1.85–1.80 (m, 1H), 1.73–1.68 (m, 1H), 1.63–1.49 (m, 7H), 1.35–1.29 (m, 7H), 1.25 (t, *J* = 7.1, 3H); ¹³C NMR (100 MHz, CDCl₃) δ 174.3, 99.3, 68.0, 62.8, 60.6, 34.8, 31.2, 30.1, 29.5, 29.5, 26.5, 25.9, 25.4, 20.1, 14.7; IR (thin film) 2937, 2858, 1737 cm⁻¹; HRMS (ESI/MeOH) *m/z* calcd for C₁₅H₂₈O₄ (M+Na)⁺ 295.1885, found 295.1881.

1,1-dideuterio-8-(tetrahydro-2H-pyran-2-yloxy)octan-1-ol, 11. To a cooled (0 °C) suspension of LiAlD₄ (1.46 g, 34.9 mmol, +98% atom D) in THF (40 mL) was added a solution of ethyl ester **10** (9.51 g, 34.7 mmol) in THF (20 mL) over 10 min. Extra THF (2 x 10 mL) was used to aid in transfer of the ester. Reaction suspension was allowed to warm to ambient temperatures over 1 h period and stirred for an additional hour. The reaction suspension was cooled (0 °C) and 10% KOH (aq) (4 mL) was added slowly over 15 min (*Caution: vigorous evolution of H₂ gas*). The resultant reaction mixture was warmed to ambient temperatures over 1 h, and filtered over a pad of Celite. The filter cake was washed with hot EtOAc (3 x 20 mL), and the filtrate was concentrated *in vacuo* to the yield deuterated alcohol **11** as a clear oil (8.50 g, 100 %): ¹H NMR (500 MHz, CDCl₃) δ 4.57 (dd, *J* = 4.3, 2.8, 1H), 3.86 (ddd, *J* = 11.1, 7.6, 3.4,

1H), 3.73 (dt, $J = 9.6, 6.9$, 1H), 3.52–3.47 (m, 1H), 3.37 (dt, $J = 9.6, 6.7$, 1H), 1.85–1.80 (m, 1H), 1.73–1.68 (m, 1H), 1.60–1.46 (m, 9H), 1.38–1.29 (m, 8H); ^{13}C NMR (125 MHz, CDCl_3) δ 99.3, 68.1, 62.8, 33.0, 31.2, 30.2, 29.9, 29.8, 26.6, 26.0, 25.9, 20.1; IR (thin film) 3421, 2929, 2855, 2192, 2087 cm^{-1} ; HRMS (ESI/MeOH) m/z calcd for $\text{C}_{13}\text{H}_{24}\text{D}_2\text{O}_3$ ($\text{M}+\text{Na}$) $^+$ 255.1905, found 255.1909.

8,8,8-trideuterio-1-(tetrahydro-2H-pyran-2-yloxy)octane, 12. To a cooled (0 °C) solution of deuterated alcohol **11** (8.11 g, 34.9 mmol) and Et_3N (6.00 mL, 43.0 mmol) in CH_2Cl_2 (57 mL) was slowly added methanesulfonyl chloride (2.70 mL, 34.9 mmol) over 5 min. After stirring for 15 min, saturated NaHCO_3 (aq) (20 mL) was added, followed by warming the reaction mixture to room temperature. The bi-phasic mixture was extracted with CH_2Cl_2 (3 x 40 mL), dried over MgSO_4 , and concentrated *in vacuo* to yield the corresponding mesylate as yellow oil. The resultant mesylate was processed through the next synthetic sequence without further purification.

The unpurified oil was immediately diluted with THF (30 mL) and added over 10 min to a cooled (0 °C) suspension of LiAlD_4 (1.72 g, 41.0 mmol, +98% atom D) in THF (10 mL). Extra THF (2 x 10 mL) was used to aid in transfer of the mesylate. Reaction suspension was allowed to gradually warm to room temperature and heated at reflux for 24 h. The suspension was cooled (0 °C) and quenched by slow addition of aqueous solution of 10% KOH (aq) (4 mL). The resultant white suspension was warmed to ambient temperatures and filtered over a pad of Celite. The filter cake was washed with hot EtOAc (3 x 20 mL) and concentrated *in vacuo* to provide a yellow oil. The oil was purified by flash chromatography (10:90 EtOAc :hexanes) to give the deuterated ether **12** as a clear oil (6.53 g, 86 %): ^1H NMR (500 MHz, CDCl_3) δ 4.57 (dd, $J = 4.3, 2.9$, 1H), 3.87 (ddd, $J = 11.0, 7.6, 3.4$, 1H), 3.72 (dt, $J = 9.6, 6.9$, 1H), 3.51–3.47 (m, 1H), 3.38 (dt, $J = 9.6, 6.7$, 1H), 1.86–1.79 (m, 1H), 1.74–1.68 (m, 1H), 1.61–1.48 (m, 6H), 1.36–1.26 (m, 10H); ^{13}C NMR (125 MHz, CDCl_3) δ 99.1, 67.9, 62.6, 32.0, 31.0, 30.0, 29.7, 29.5, 26.5, 25.8, 22.6, 19.9; IR (thin film) 2924, 2856, 2211, 2123, 2074, 1139, 1123, 1079, 1034 cm^{-1} ; HRMS (ESI/MeOH) m/z calcd for $\text{C}_{13}\text{H}_{23}\text{D}_3\text{O}_2$ ($\text{M}+\text{Na}$) $^+$ 240.2019, found 240.2012.

8,8,8-trideuterio-1-iodooctane, 14. To a solution of deuterated THP-ether **12** (6.53 g, 30.0 mmol) in MeOH (50 mL) was added *p*- $\text{TsOH}\cdot\text{H}_2\text{O}$ (0.583 g, 3.06 mmol). After 4 h, saturated aqueous NaHCO_3 (10 mL) was added. The mixture was diluted with H_2O (50 mL) and Et_2O (100 mL). The organic phase was separated and the aqueous layer was extracted with Et_2O (3 x 50 mL). The combined organic phases was washed with H_2O (50 mL), dried (MgSO_4), and concentrated *in vacuo* to give a yellow oil. The oil was purified by flash chromatography (20:80 – 30:70 EtOAc :hexanes) to yield the alcohol **13** as a clear oil (3.62 g, 91%). The resultant alcohol **13** was processed to the next step without further purification.

To a cooled (0 °C) solution of PPh_3 (8.50 g, 32.4 mmol) and imidazole (2.86 g, 42.0 mmol) in Et_2O (50 mL) was introduced I_2 (9.04 g, 35.6 mmol) in portions. After 1 h, a solution of the alcohol **13** (3.62 g, 27.0 mmol) in Et_2O (20 mL) was added over 5 min. Extra Et_2O (2 x 10 mL) was used to aid in complete transfer of the alcohol. After 30 min, CH_2Cl_2 (50 mL) was added to solubilize the heterogeneous reaction mixture and warmed to room temperature. After an additional 30 min, saturated aqueous $\text{Na}_2\text{S}_2\text{O}_3$ (50 mL) was added and stirred vigorously for 15 min. The reaction mixture was extracted with Et_2O (3 x 50 mL), the combined organic layers were washed with H_2O (50 mL), dried (MgSO_4), and concentrated *in vacuo* to give white solids. The solids were suspended in pentanes (50 mL), allowed to stand for 2 h, and filtered over a pad of Celite. The Celite cake was washed with pentanes (5 x 50 mL) and the filtrate was concentrated *in vacuo* to yield the iodide **14** as a clear oil (6.30 g, 96%): ^1H NMR (500 MHz, CDCl_3) δ 3.19 (t, $J = 7.1$, 2H), 1.82 (quin, $J = 7.1$, 2H), 1.40–1.35 (m, 2H), 1.30–1.26 (m, 8H); ^{13}C NMR (125 MHz, CDCl_3) δ 34.0, 32.1, 31.0, 29.6, 29.0, 22.8, 7.85; IR (thin film) 2923, 2854, 2212, 2121, 2074 cm^{-1} .

1-(Tetrahydro-2H-pyran-2-yloxy)-18,18,18-trideuteriooctadec-9-yne, 15. To a cooled ($-42\text{ }^{\circ}\text{C}$) solution of alkyne **7** (1.81 g, 7.59 mmol) in THF/HMPA (3:1 mixture, 20 mL) under an argon atmosphere was added *n*-BuLi (4.0 mL, 8.48 mmol, 2.1 M solution in hexanes). The bright red colored reaction mixture was maintained between $-50\text{ }^{\circ}\text{C}$ to $-40\text{ }^{\circ}\text{C}$. After 1 h, a solution of iodide **14** (2.07 g, 8.51 mmol) in THF (10 mL) was added slowly over 10 min. Extra THF (2 x 5 mL) was used to aid in transfer of the iodide. The reaction mixture was warmed to room temperature over 6 h and allowed to stir for an additional 12 h. The mixture was diluted with H_2O (20 mL) and extracted with Et_2O (3 x 20 mL). The combined organic phase was washed with H_2O (3 x 30 mL), dried over MgSO_4 , and concentrated *in vacuo* to give an orange oil. The oil was purified by flash chromatography to yield alkyne **15** as a clear oil (2.20 g, 82%): ^1H NMR (500 MHz, CDCl_3) δ 4.57 (dd, $J = 4.3, 2.8, 1\text{H}$), 3.87 (ddd, $J = 11.2, 7.7, 3.4, 1\text{H}$), 3.72 (dt, $J = 9.6, 6.9, 1\text{H}$), 3.52–3.48 (m, 1H), 3.37 (dt, $J = 9.6, 6.7, 1\text{H}$), 2.17–2.12 (m, 4H), 1.86–1.80 (m, 1H), 1.73–1.68 (m, 1H), 1.60–1.59 (m, 4H), 1.55–1.50 (m, 3H), 1.48–1.44 (m, 4H), 1.38–1.26 (m, 17H); ^{13}C NMR (125 MHz, CDCl_3) δ 99.1, 80.5, 80.4, 67.9, 62.6, 32.0, 31.0, 30.0, 29.6, 29.5, 29.4, 29.4, 29.3, 29.1, 29.0, 26.4, 25.7, 22.6, 19.3, 19.0; HRMS (ESI/MeOH) *m/z* calcd for $\text{C}_{23}\text{H}_{39}\text{D}_3\text{O}_2$ ($\text{M}+\text{Na}$) $^+$ 376.3271, found 376.362.

(Z)-1-(Tetrahydro-2H-pyran-2-yloxy)-18,18,18-trideuteriooctadec-9-ene, 16. To a round bottomed flask containing $\text{Ni}(\text{OAc})_2 \cdot 4\text{H}_2\text{O}$ (0.447 g, 1.80 mmol) was added EtOH (7 mL), and the resultant green solution was subjected to three cycles of evacuation (30 sec) followed by purging with argon. To this vigorously stirred solution was added NaBH_4 (69.7 mg, 1.76 mmol) dissolved in EtOH (5 mL) and 1.0 M NaOH (1 mL). The reaction mixture instantly separated into two phases, with black precipitate, and evolution of H_2 (g) was observed. The reaction vessel was fitted with a balloon of H_2 (g) and stirred under an atmosphere of H_2 (g) (1 atm). After 10 min, ethylene diamine (0.25 mL) was introduced, followed by alkyne **15** (2.10 g, 5.94 mmol) in EtOH (5 mL). Extra EtOH (2 x 5 mL) was used to aid in transfer of the alkyne. After 4 h, the reaction mixture was purged with Ar (g) and filtered over a pad of Celite, and the filter cake was washed with CH_2Cl_2 (3 x 30 mL). The filtrate was successively washed with H_2O (50 mL), brine (20 mL), dried over MgSO_4 , and concentrated *in vacuo* to give the crude product, which was purified by flash chromatography (10:90 EtOAc:hexanes) to yield the alkene **16** as a clear oil (2.02 g, 96%): ^1H NMR (500 MHz, CDCl_3) δ 5.37–5.31 (m, 2H), 4.57 (dd, $J = 4.3, 2.8, 1\text{H}$), 3.87 (dd, $J = 11.1, 7.7, 3.4, 1\text{H}$), 3.72 (dt, $J = 9.6, 6.9, 1\text{H}$), 3.52–3.48 (m, 1H), 3.37 (dt, $J = 9.6, 6.7, 1\text{H}$), 2.03–1.97 (m, 4H), 1.86–1.80 (m, 1H), 1.74–1.68 (m, 1H), 1.62–1.44 (m, 6H), 1.33–1.26 (m, 22H); ^{13}C NMR (125 MHz, CDCl_3) δ 130.4, 130.3, 99.3, 68.1, 62.8, 32.3, 31.2, 30.2, 30.2, 30.0, 30.0, 29.9, 29.9, 29.8, 29.7, 27.6, 27.6, 26.7, 26.0, 22.8, 20.1; HRMS (ESI/MeOH) *m/z* calcd for $\text{C}_{23}\text{H}_{41}\text{D}_3\text{O}_2$ ($\text{M}+\text{Na}$) $^+$ 378.3427, found 378.3419.

(Z)-18,18,18-trideuteriooctadec-9-enoic acid (oleic acid-d3), 17. To the THP-ether **16** (2.00 g, 5.62 mmol) dissolved in MeOH (25 mL) was added *p*TsOH $\cdot\text{H}_2\text{O}$ (0.152 g, 0.799 mmol). After 30 min, saturated aqueous sodium bicarbonate (10 mL) was added, and the mixture was diluted with H_2O (20 mL) and extracted with Et_2O (3 x 50 mL). The combined organic layers were washed with H_2O (2 x 50 mL), dried over MgSO_4 , and concentrated *in vacuo* to give the deprotected primary alcohol as a clear colorless oil that was used without further purification.

To a cooled ($0\text{ }^{\circ}\text{C}$) solution of the alcohol in acetone (25 mL) was added Jones reagent (4.6 mL, 12.4 mmol, 2.7 M CrO_3 in 4 N H_2SO_4) over 10 min. The reaction mixture was maintained between $0\text{ }^{\circ}\text{C}$ – $10\text{ }^{\circ}\text{C}$, and after 4 h, *i*-PrOH (10 mL) was added and stirred for 30 min. The green suspension was filtered through a pad of Celite, the filter cake was washed with acetone (3 x 20 mL) followed by EtOAc (3 x 50 mL), and the filtrate was concentrated to 30% of its original volume. The organic phase was diluted with H_2O (20 mL), extracted with EtOAc (5 x 50 mL), washed with brine (20 mL), dried over MgSO_4 , and concentrated *in vacuo* to give dark green oil. This crude product was diluted with EtOAc (50 mL), stirred over Darco charcoal for 15 min, filtered over Celite, and the filter cake was washed with EtOAc (3 x 50 mL). The filtrate was

concentrated *in vacuo* to give light green oil that was purified by flash chromatography (10:90 EtOAc:hexanes) to yield the acid **17** as a clear oil (1.15 g, 72%): ^1H NMR (500 MHz, CDCl_3) δ 5.38–5.30 (m, 2H), 2.35 (t, $J = 7.5\text{H}$, 2H), 2.05–1.90 (m, 4H), 1.63 (app. q, $J = 7.5\text{H}$, 2H), 1.32–1.28 (m, 20H); ^{13}C NMR (125 MHz, CDCl_3) δ 179.4, 130.3, 130.0, 34.1, 32.1, 30.0, 29.9, 29.8, 29.6, 29.4, 29.3, 29.3, 27.5, 27.4, 24.9, 22.6; HRMS (ESI/MeOH) m/z calcd for $\text{C}_{18}\text{H}_{31}\text{D}_3\text{O}_2$ (M-H) $^{-1}$ 284.2669, found 284.2675.

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