JOC*Article*

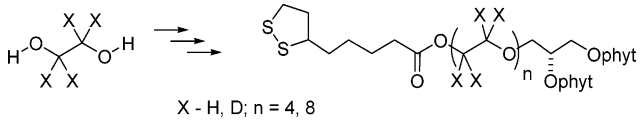
New Deuterated Oligo(ethylene glycol) Building Blocks and Their Use in the Preparation of Surface Active Lipids Possessing Labeled Hydrophilic Tethers

Robert J. Faragher and Adrian L. Schwan*

Department of Chemistry, University of Guelph, Guelph, Ontario, Canada N1G 2W1

schwan@uoguelph.ca

Received September 13, 2007



X - H, D; n = 4, 8

For the introduction of additional analysis protocols of tethered molecules, a method is presented to prepare functionalized, deuterated oligo(ethylene glycols) from ethylene glycol-d_4. Partial oligomerization of ethylene glycol-d_4 and conversion to ditosylates is accompanied by coupling reactions to prepare doubly benzyl protected oligo(ethylene glycols) with two to five repeating units. The tetramer bearing 16 deuteria was elaborated at both ends to eventually prepare 2,3-di-*O*-phytanyl-*sn*-glycerol-1-tetraethylene glycol-D,L-α-lipoic acid ester (DPTL), which bears a fully deuterated tetra(ethylene glycol) spacer group. Through linking of functionalized components, an analogue of DPTL possessing an octa(ethylene glycol) spacer group was prepared, both in deuterated and unlabeled form.

Introduction

Linker technology[1−3] has found a home in many fields including organic synthesis,[4] drug delivery,[5,6] and interfacial chemistry[7−10] including molecule immobilization techniques.[7,11−14] Ethylene glycol-based polymers and oligomers have proved very popular for these purposes because of water solubility, nontoxicity, and low cost.[2,6]

Poly(ethylene glycol) (PEG) and oligo(ethylene glycol) (OEG) linkers are particularly useful in the area of surface modification and analysis.[9,10,15] The binding of tethered substrates to surfaces has facilitated the evolution of tethered bilayer lipid membranes (tBLMs)[16−18] which serve as viable and popular models for the biological membrane, enabling the study of membrane proteins[18−21] and ion transport mechanisms.[12,17] The concept calls for a molecule(s) that tethers the lipid to a metal surface, yet incorporates a spacer region between the bilayer and the metal substrate. Optimally, the tether is hydrophilic and can counteract the hydrophobic nature of the metal surface. The substrate for study is then introduced and self-assembles through interaction with the immobilized lipid.

* Address correspondence to this author. Fax: (519)766-1499. Phone: (519)-824-4120, ext. 53061.

(1) Rodwell, J. D.; McKearn, T. J. *Bio/Technology* **1985**, *3*, 889−894.

(2) Hinds, K. D. In *Biomaterials for Delivery and Targeting of Proteins and Nucleic Acids*; Mahato, R. I., Ed.; CRC Press LLC: Boca Raton, FL, 2005; pp 119−185.

(3) Shoseyov, O.; Shani, Z.; Levy, I. *Microbiol. Mol. Biol. Rev.* **2006**, *70*, 283−295.

(4) (a) Beythien, J.; Barthelemy, S.; Schneeberger, P.; White, P. D. *Tetrahedron Lett.* **2006**, *47*, 3009−3012. (b) de Paz, J. L.; Noti, C.; Seeberger, P. H. *J. Am. Chem. Soc.* **2006**, *128*, 2766−2767. (c) Subramanian, T.; Wang, Z.; Troutman, J. M.; Andres, D. A.; Spielmann, H. P. *Org. Lett.* **2005**, *7*, 2109−2112. (d) Flickinger, S. T.; Patel, M.; Binkowski, B. F.; Lowe, A. M.; Li, M.-H.; Kim, C.; Cerrina, F.; Belshaw, P. J. *Org. Lett.* **2006**, *8*, 2357−2360. (e) Liu, X.-L.; Sheng, S.-R.; Wang, Q.-Y.; Sun, W.-K.; Xin, Q.; Ao, H.-Y. *J. Chem. Res.* **2006**, 118−120.

(5) (a) Pasut, G.; Morpurgo, M.; Veronese, F. M. In *Delivery of Protein and Peptide Drugs in Cancer*; Torchilin, V. P., Ed.; Imperial College Press: London, UK, 2006; pp 53−84. (b) Veronese, F. M.; Pasut, G. *Drug Discovery Today* **2005**, *10*, 1451−1458. (c) Gonzalo, T.; Talman, E. G.; Van de Ven, A.; Temming, K.; Greupink, R.; Beljaars, L.; Reker-Smit, C.; Meijer, D. K. F.; Molema, G.; Poelstra, K.; Kok, R. J. *J. Controlled Release* **2006**, *111*, 193−203.

(6) Pasut, G.; Guiotto, A.; Veronese, F. M. *Expert Opin. Ther. Pat.* **2004**, *14*, 859−894.

(7) (a) Blomberg, E.; Claesson, P. M.; Konradsson, P.; Liedberg, B. *Langmuir* **2006**, *22*, 10038−10046. (b) Miyamae, T.; Yokoyama, H.; Han, S.; Ishizone, T. *e-J. Surf. Sci. Nanotechnol.* **2006**, *4*, 515−520. (c) Tang, Z.; Padmawar, P. A.; Canteenwala, T.; Gao, Y.; Watkins, E.; Majewski, J.; Chiang, L. Y.; Wang, H.-L. *Langmuir* **2006**, *22*, 5366−5373.

(8) (a) Hoffmann, C.; Tovar, G. E. M. *J. Colloid Interface Sci.* **2006**, *295*, 427−435. (b) Oestblom, M.; Valiokas, R.; Konradsson, P.; Svensson, S. C. T.; Liedberg, B.; Garrett, M.; Allara, D. L. *J. Phys. Chem. B* **2006**, *110*, 1830−1836. (c) Balamurugan, S.; Ista, L. K.; Yan, J.; Lopez, G. P.; Fick, J.; Himmelhaus, M.; Grunze, M. *J. Am. Chem. Soc.* **2005**, *127*, 14548−14549. (d) Malysheva, L.; Onipko, A.; Valiokas, R.; Liedberg, B. *Appl. Surf. Sci.* **2005**, *246*, 372−376. (e) Li, L.; Chen, S.; Zheng, J.; Ratner, B. D.; Jiang, S. *J. Phys. Chem. B* **2005**, *109*, 2934−2941.

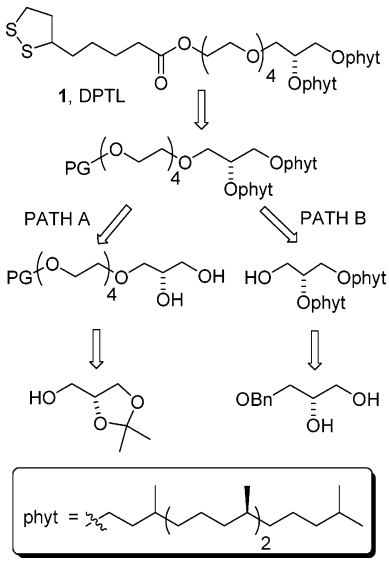
(9) (a) Boozer, C.; Ladd, J.; Chen, S.; Yu, Q.; Homola, J.; Jiang, S. *Anal. Chem.* **2004**, *76*, 6967−6972. (b) Zheng, J.; Li, L.; Chen, S.; Jiang, S. *Langmuir* **2004**, *20*, 8931−8938. (c) Zwahlen, M.; Herrwerth, S.; Eck, W.; Grunze, M.; Haehner, G. *Langmuir* **2003**, *19*, 9305−9310. (d) Chan, Y.-H. M.; Schweiss, R.; Werner, C.; Grunze, M. *Langmuir* **2003**, *19*, 7380−7385.

10.1021/jo701979z CCC: $40.75
Published on Web 01/17/2008

2,3-Di-O-phytanyl-*sn*-glycerol-1-tetraethylene glycol-D,L-α-lipoic acid ester (DPTL, **1**) represents a popular substrate for the studies noted above.[12,20,22−26] Not only does DPTL feature a tetra(ethylene glycol) unit as the requisite hydrophilic tether, but it also advertises nonhydrolyzable ether linkages throughout and two phytanyl chains which embrace guest molecules. Until now, DPTL has been prepared and utilized without concern for isotope incorporation.[25,26] If DPTL were to feature a deuterated tether, one could call on neutron reflectivity measurements[27] to assess the amount of water that resides in the tethered section of the membrane.[28] Moreover the development of isotopically labeled oligo(ethylene glycol) linkers in general would facilitate structural analysis by other techniques including IR,[29] NMR,[30] and mass spectrometry.[31,32]

The previous preparation of DPTL, which parallels retrosynthetic path A of Scheme 1,[25] does not mesh well with any reasonable plan for deuterium incorporation principally because 20 equiv of tetra(ethylene glycol) (**EG4**) were employed to incorporate the tether. We also sought to improve other steps, such as the double phytanylation reaction, which occurred in 49% previously.[25] Finally, we held the view that alternative path B should be evaluated as it introduces the labeled component at a later stage of the overall synthesis.

SCHEME 1. Possible Approaches to DPTL, 1



In this paper, we outline the synthesis of small useful deuterated oligoisomeric building blocks from ethylene glycol-d_4 (**EG**-d_4) and also incorporate an **EG4**-d_{16} derivative for the preparation of DPTL-d_{16} and its analogue with an octameric tether. An experimental description (only) of our preparation of nondeuterated DPTL, developed with the option of facile extension to the labeled manifold, has already been published.[26] Despite the large number of available protocols for OEG linker methods,[26,33−37] applications toward deuterated substrates have not been demonstrated.

(10) (a) Yu, Q.; Chen, S.; Taylor, A. D.; Homola, J.; Hock, B.; Jiang, S. *Sens. Actuators, B* **2005**, *B107*, 193−201. (b) Yam, C. M.; Gu, J.; Li, S.; Cai, C. *J. Colloid Interface Sci.* **2005**, *285*, 711−718. (c) Andruzzi, L.; Senaratne, W.; Hexemer, A.; Sheets, E. D.; Ilic, B.; Kramer, E. J.; Baird, B.; Ober, C. K. *Langmuir* **2005**, *21*, 2495−2504. (d) Riepl, M.; Oestblom, M.; Lundstroem, I.; Svensson, S. C. T.; Denier van der Gon, A. W.; Schaeferling, O. M.; Liedberg, B. *Langmuir* **2005**, *21*, 1042−1050. (e) Choi, N.-S.; Lee, Y. M.; Cho, K. Y.; Ko, D.-H.; Park, J.-K. *Electrochem. Commun.* **2004**, *6*, 1238−1242. (f) Neo, W. K.; Chan-Park, M. B.; Gao, J. X.; Dong, L. *Langmuir* **2004**, *20*, 11073−11083. (g) Yam, C. M.; Lopez-Romero, J. M.; Gu, J.; Cai, C. *Chem. Commun.* **2004**, 2510−2511. (h) Kleinert, M.; Roeckendorf, N.; Lindhorst, T. K. *Eur. J. Org. Chem.* **2004**, 3931−3940. (i) Ladd, J.; Boozer, C.; Yu, Q.; Chen, S.; Homola, J.; Jiang, S. *Langmuir* **2004**, *20*, 8090−8095. (j) Zhou, Y.; Valiokas, R.; Liedberg, B. *Langmuir* **2004**, *20*, 6206−6215. (k) Martins, M. C. L.; Naeemi, E.; Ratner, B. D.; Barbosa, M. A. *J. Mater. Sci.-Mater. Med.* **2003**, *14*, 945−954. (l) Yan, J.; Tender, L. M.; Hampton, P. D.; Lopez, G. P. *J. Phys. Chem. B* **2001**, *105*, 8905−8910.

(11) (a) Clare, B. H.; Guzman, O.; de Pablo, J.; Abbott, N. L. *Langmuir* **2006**, *22*, 7776−7782. (b) Clare, B. H.; Guzman, O.; De Pablo, J. J.; Abbott, N. L. *Langmuir* **2006**, *22*, 4654−4659. (c) Sun, X.-L.; Stabler, C. L.; Cazalis, C. S.; Chaikof, E. L. *Bioconjugate Chem.* **2006**, *17*, 52−57. (d) Mack, J.; Kienle, S.; Leipert, D.; Redemann, T.; Kraas, W.; Jung, G. *Lett. Pept. Sci.* **1999**, *6*, 135−142. (e) Clare, B. H.; Guzman, O.; de Pablo, Juan, J.; Abbott, Nicholas, L. *Langmuir* **2006**, *22*, 4654−4659.

(12) Naumann, R.; Walz, D.; Schiller, S. M.; Knoll, W. *J. Electroanal. Chem.* **2003**, *550−551*, 241−252.

(13) Sinner, E.-K.; Knoll, W. *Curr. Chem. Biol.* **2001**, *5*, 705−711.

(14) Guidelli, R.; Aloisi, G.; Becucci, L.; Dolfi, A.; Moncelli, M. R.; Tadini Buoninsegni, F. *J. Electroanal. Chem.* **2001**, *504*, 1−28.

(15) Zalipsky, S. *Bioconjugate Chem.* **1995**, *6*, 150−65.

(16) Yin, P. *Adv. Planar Lipid Bilayers Liposomes* **2005**, *2*, 49−76.

(17) Jeuken, L. J. C.; Bushby, R. J.; Evans, S. D. *Electrochem. Commun.* **2007**, *9*, 610−614.

(18) Kohli, N.; Hassler, B. L.; Parthasarathy, L.; Richardson, R. J.; Ofoli, R. Y.; Worden, R. M.; Lee, I. *Biomacromolecules* **2006**, *7*, 3327−3335.

(19) Jeuken, L. J. C.; Connell, S. D.; Henderson, P. J. F.; Gennis, R. B.; Evans, S. D.; Bushby, R. J. *J. Am. Chem. Soc.* **2006**, *128*, 1711−1716.

(20) He, L.; Robertson, J. W. F.; Li, J.; Kaercher, I.; Schiller, S. M.; Knoll, W.; Naumann, R. *Langmuir* **2005**, *21*, 11666−11672.

(21) Giess, F.; Friedrich, M. G.; Heberle, J.; Naumann, R. L.; Knoll, W. *Biophys. J.* **2004**, *87*, 3213−3220.

(22) Becucci, L.; Moncelli, M. R.; Guidelli, R. *Langmuir* **2006**, *22*, 1341−1346.

(23) Moncelli, M. R.; Becucci, L.; Schiller, S. M. *Bioelectrochemistry* **2004**, *63*, 161−167.

(24) Naumann, R.; Schiller, S. M.; Giess, F.; Grohe, B.; Hartman, K. B.; Kaercher, I.; Koeper, I.; Luebben, J.; Vasilev, K.; Knoll, W. *Langmuir* **2003**, *19*, 5435−5443.

(25) Schiller, S. M.; Naumann, R.; Lovejoy, K.; Kunz, H.; Knoll, W. *Angew. Chem., Int. Ed.* **2003**, *42*, 208−211.

(26) Kunze, J.; Leitch, J.; Schwan, A. L.; Faragher, R. J.; Naumann, R.; Schiller, S.; Knoll, W.; Dutcher, J. R.; Lipkowski, J. *Langmuir* **2006**, *22*, 5509−5519.

(27) Plech, A.; Salditt, T. In *Handbook of Polyelectrolytes and Their Applications*; Tripathy, S. K., Kumar, J., Nalwa, H. S., Eds.; American Scientific Publishers: Stevenson Ranch, CA, 2002; Vol. 1, pp 265−291.

(28) (a) Burgess, I.; Zamlynny, V.; Szymanski, G.; Schwan, A. L.; Faragher, R. J.; Lipkowski, J.; Majewski, J.; Satija, S. *J. Electroanal. Chem.* **2003**, *550−551*, 187−199. (b) Yaseen, M.; Lu, J. R.; Webster, J. R. P.; Penfold, J. *Langmuir* **2006**, *22*, 5825−5832. (c) Ott, F.; Cousin, F.; Menelle, A. *J. Alloys Compd.* **2004**, *382*, 29−38. (d) Tang, Y.; Su, T. J.; Armstrong, J.; Lu, J. R.; Lewis, A. L.; Vick, T. A.; Stratford, P. W.; Heenan, R. K.; Penfold, J. *Macromolecules* **2003**, *36*, 8440−8448. (e) Cooper, J. M.; Cubitt, R.; Dalgliesh, R. M.; Gadegaard, N.; Glidle, A.; Hillman, A. R.; Mortimer, R. J.; Ryder, K. S.; Smith, E. L. *J. Am. Chem. Soc.* **2004**, *126*, 15362−15363.

(29) Bin, X.; Horswell, S. L.; Lipkowski, J. *Biophys. J.* **2005**, *89*, 592−604.

(30) Anastasiadis, A.; Morton, C. J.; Talbo, G. H.; Koeppe, R. E., II; Separovic, F. *Int. J. Pept. Res. Ther.* **2006**, *12*, 243−252.

(31) Patel, A.; Perrin, D. M. *Bioconjugate Chem.* **2004**, *15*, 224−230.

(32) An efficient and useful synthesis of a functionalized dimeric **EG2**-d_8 unit that does not begin with **EG**-d_4 has been described: see ref 31. A deuterated poly(ethylene glycol) material has been prepared. See: Poschalko, A.; Lancelot, N.; Marin, J.; Larras, V.; Limal, D.; Elbayed, K.; Raya, J.; Piotto, M.; Briand, J.-P.; Guichard, G.; Bianco, A. *Chemistry* (*Weinheim*) **2004**, *10*, 4532−4537.

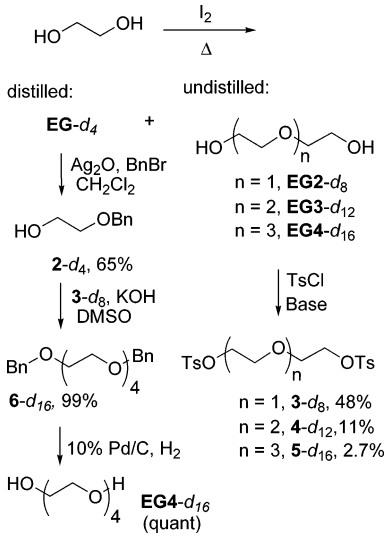
(33) (a) Svedhem, S.; Hollander, C. A.; Shi, J.; Konradsson, P.; Liedberg, B.; Svensson, S. C. *J. Org. Chem.* **2001**, *66*, 4494−4503. (b) Iyer, S. S.; Anderson, A. S.; Reed, S.; Swanson, B.; Schmidt, J. G. *Tetrahedron Lett.* **2004**, *45*, 4285−4288. (c) Loiseau, F. A.; Hii, K. K.; Hill, A. M. *J. Org. Chem.* **2004**, *69*, 639−647. (d) Lu, G.; Lam, S.; Burgess, K. *Chem. Commun.* **2006**, 1652−1654. (e) Houseman, B. T.; Mrksich, M. *J. Org. Chem.* **1998**, *63*, 7552−7555.

(34) Beadle, J. R.; Korzeniowski, S. H.; Rosenberg, D. E.; Garcia-Slanga, B. J.; Gokel, G. W. *J. Org. Chem.* **1984**, *49*, 1594−1603.

(35) Schwabacher, A. W.; Lane, J. W.; Schiesher, M. W.; Leigh, K. M.; Johnson, C. W. *J. Org. Chem.* **1998**, *63*, 1727−1729.

(36) Dicus, C. W.; Nantz, M. H. *Synlett* **2006**, 2821−2823.

SCHEME 2. Assembly of EG4-d_{16} Derivatives



Results and Discussion

The lone preparation of deuterated **EG**-d_4 oligomers was outlined by Schnabel.[38] As outlined in that paper, **EG**-d_4 was converted to numerous oligomers upon heating to 190 °C in the presence of 1.2 mol % of I_2. The mixture was subjected to fractional distillation and numerous perdeuterated EG oligomers were obtained including **EG4**-d_{16}, which was isolated in 8% yield; there is, however, no mention of the absolute scale of the reaction.[38] To ensure a complete separation of **EG3**-d_{12} and **EG5**-d_{20} from the target molecule, it was felt that significantly large amounts of **EG**-d_4 would be required.[39] Moreover the distillation had the potential to be exasperatingly slow, perhaps requiring numerous repetitions, to achieve but an 8% yield or be unsuccessful in the worst case scenario! Finally, we were concerned about our ability to be certain that **EG4**-d_{16} was completely free of other oligomers.

For these reasons, an alterative procedure was developed. The **EG**-d_4 monomer would be heated for a shorter period of time to access larger amounts of smaller oligomers and distillation in combination with functional group manipulations would complete the separation. The initial target under these conditions was a derivatized dimer that could then be extended by substitution reactions. To this end, the following treatment was performed. In a typical protocol, 6.4 g of **EG**-d_4 was heated at 160−170 °C for 7.5 h in the presence of 1.2% (mol) of I_2. The heat was reduced and the pot was subjected to vacuum distillation to recover 3.84 g of unreacted monomer. The remaining pot constituents were subjected to tosylation conditions. The result was three ditosylated compounds (**3**-d_8, **4**-d_{12}, **5**-d_{16}) which could be separated by flash chromatography (Scheme 2).

The subsequent plan was to take dimeric ditosylate **3**-d_8 and react it at each end with a monoprotected **EG**-d_4 such as **2**-d_4. Benzyl-protected **2**-d_4 has previously been prepared in good yield by reacting BnOH and $BrCH_2COOH$ followed by deuterium incorporation by ester enolate exchange and $LiAlD_4$ reduction events.[40,41] Adopting that protocol would mean we would have no use for the significant amount of **EG**-d_4 recovered during the aforementioned distillation. Hence we sought to develop a method to monobenzylate **EG**-d_4, thereby exploiting the excess available deuterated starting material.

Whereas many monofunctionalization protocols typically rely on an excess of the bifunctional reactant, this was unacceptable due to the high cost. After experimental evaluation of a number of methods, the use of Ag_2O, BnBr, and CH_2Cl_2 was adopted for its convenience.[42] In particular, the absence of a water workup would permit recovery of unreacted **EG**-d_4. Under the optimized conditions, the targeted **2**-d_4 was isolated (50%) in addition to starting material (8%) and doubly benzylated **EG**-d_4 (16%). Given that the latter could be converted to **EG**-d_4 through hydrogenation (H_2, 10% Pd/C, EtOAc), the yield of **3**-d_8 was 65% once corrected for recovered starting material from the two sources.

At this point, methods were explored for the reaction of **2**-d_4 with ditosylate **3**-d_8. The Williamson-like protocol of powdered KOH in DMSO forwarded by Johnstone and Rose,[43] which is used for alkylation procedures targeting ether containing phospholipids,[44] also proved amenable to the chemistry at hand. The tosic acid elimination that occurred during trials of other conditions was not observed and doubly benzylated **6**-d_{16} was cleanly provided in 99% yield (Scheme 2).

A quantitative hydrogenative deprotection of **6**-d_{16} to **EG4**-d_{16} was carried out under the conditions previously employed, and provided **EG4**-d_{16} of suitable purity for subsequent chemistry. The synthetic pathway from **EG**-d_4 to **EG4**-d_{16} occurred with only a 44% loss of consumed **EG**-d_4 over 5 steps[45] and demonstrates the first preparation of gram quantities of the useful precursor **EG4**-d_{16}.

The successful double alkylation procedure and the isolation of additional ditosylated compounds prompted us to evaluate the chemistry for other small oligomers of **EG**-d_4. The ditosylates (Scheme 2) were reacted with either BnOH or **2**-d_4 and in all cases but one, the yields of elongated product approached or exceeded 90%. The low yielding reaction involved the ditosylate of **EG**-d_4 where elimination chemistry dominated the process. Notwithstanding those results, successful reactions of ditosylates **3**-d_8, **4**-d_{12}, and **5**-d_{16} offer deuterated dibenzyl protected oligomers of **EG**-d_4 possessing 2−5 repeating units. The efficient deprotective hydrogenation of dibenzylated compounds **EG**-d_4 and **EG4**-d_{16} suggests the general applicability of the reaction for other oligomers. Higher doubly tosylated oligomers may be prepared more efficiently by longer heating in the original oligomerization reaction, or a more efficient method may involve debenzylation and further functionalization of the products of Table 1. Compounds with linker chains longer

(37) Waterhouse, J. E.; Harbottle, R. P.; Keller, M.; Kostarelos, K.; Coutelle, C.; Jorgensen, M. R.; Miller, A. D. *ChemBioChem* **2005**, *6*, 1212−1223.

(38) Schnabel, R. *J. Labelled Compd. Radiopharm.* **1992**, *31*, 91−94.

(39) At the time of these experiments, no oligomeric forms of **EG**-d_4 were commercially available.

(40) Bird, I.; Farmer, P. B. *J. Labelled Compd. Radiopharm.* **1989**, *27*, 199−216.

(41) Campbell, J. B. *J. Chem. Soc., Perkin Trans. 1* **1983**, 1213−1217.

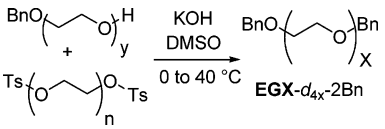
(42) Bouzide, A.; Sauve, G. *Tetrahedron Lett.* **1997**, *38*, 5945−5948.

(43) Johnstone, R. A. W.; Rose, M. E. *Tetrahedron* **1979**, *35*, 2169−2173.

(44) (a) Wang, Z.; Schwan, A. L.; Lairson, L. L.; O'Donnell, J. S.; Byrne, G. F.; Foye, A.; Holm, B. A.; Notter, R. H. *Am. J. Physiol.* **2003**, *285*, L550−L559. (b) Chang, Y.; Wang, Z.; Notter, Robert, H.; Wang, Z.; Qu, L.; Schwan, A. L. *Bioorg. Med. Chem. Lett.* **2004**, *14*, 5983−5986.

(45) This value arises in the following manner: 52% losses occur in the two steps providing ditosylate **3**-d_8 and 35% losses occur making **3**-d_4. Since these components each deliver one-half of the required four EG units to **6**-d_{16}, those losses become 26% and 17.5%, respectively. The total losses are therefore 43.5% before the final two near-quantitative steps completing the preparation of **EG4**-d_{16}.

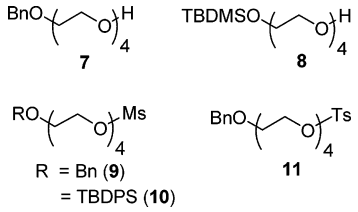
TABLE 1. General Preparation for Doubly Benzylated Perdeuterated OEG's



no.	*n*	*y*	*X*[a]	product	yield (%)
1	2	0	2	**EG2**-d_8-2Bn	89
2	1	1	3	**EG3**-d_{12}-2Bn	32[b]
3	3	0	3	**EG3**-d_{12}-2Bn	91
4	2	1	4	**6**-d_{16}	99
5	4	0	4	**6**-d_{16}	91
6	3	1	5	**EG5**-d_{20}-2Bn	98

[a] $X = (n + 2y)$. [b] Only **EG3**-d_{12}-2Bn was recovered from a mixture of three products. On scrutiny of the ^{1}H NMR of the crude reaction mixture, the other two products appeared to possess unsaturation.

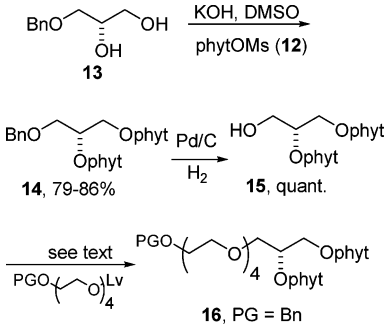
CHART 1. Some Functionalized EG4 Derivatives



than five units can be accessed through coupling reactions presented herein (vide infra) or the oligomeric diols can be extended by using already demonstrated protocols.[35,46,47]

For eventual construction of DPTL, the **EG4**-d_{16} would require further adaptation by way of a monofunctionalization.[48] As such, known **EG4** derivatives **7**,[37,49] **8**,[36] **9**,[50] **10**,[34] and **11**[49] (Chart 1) were prepared as potential building blocks for DPTL construction by adapting and in some cases improving the yields or atom efficiencies of past procedures. Given these options for bifunctional **EG4** linkers, attention was turned toward the elaboration of the glycerol portion and introduction of the hydrophobic phytanyl groups. Phytanyl mesylate (**12**) was prepared from phytanol by adapting a literature protocol (95%).[51] The mesylate was then employed for the double alkylation of benzyl glycerol (**13**), using powdered KOH/DMSO, to afford the desired benzyl diphytanyl glycerol (**14**) in 86−95% yield after optimization. Concluding with a reductive debenzylation (H_2, 10% Pd/C, quant.), this preparation of diphytanyl glycerol (**15**) represents a significant improvement over other methods.[51,52] This particular disubstituted glycerol, which is naturally occurring in the lipid membranes of *H. cutirubrum*,[53,54] has been sought for a variety of reasons, as phytanyl chains improve both the chemical stability and fluidity of a molecule compared to conventional linear fatty alkyl options.[55]

SCHEME 3. Functionalization of Glycerol



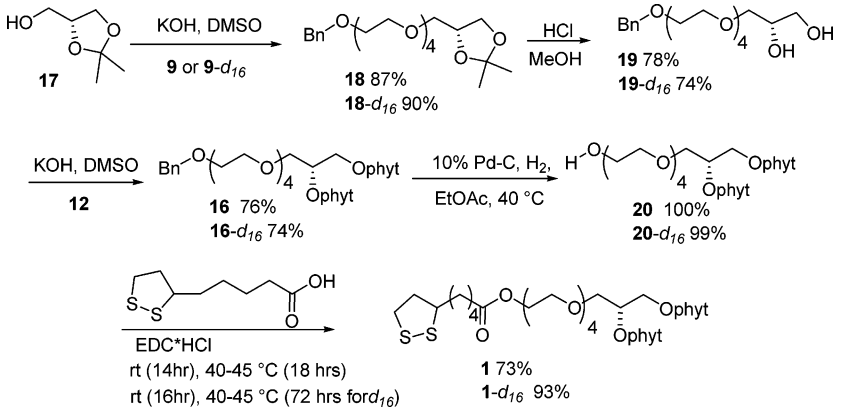
The direct linking of diphytanyl glycerol (**15**) and compounds **9**−**11** was attempted under a number of conditions. It was quickly established that TBDMS-protected **10** readily degraded under a variety of basic conditions and was ineffective.[56] Mesylate **9** proved unreactive to the reaction conditions and the coupled material was eventually achieved by using NaH and tosylate **11** in THF. However, the reaction was slow and incomplete even after 7 days, but afforded **16** in 72% based on consumed starting material (Scheme 3).

The synthetic approach to this juncture, based on retrosynthetic path B of Scheme 1, assembled the tether and glycerol components of DPTL in an efficient and partially convergent manner, one amenable to label incorporation. However, since the last step was sluggish, we turned our focus fully toward path A (Scheme 1), on which we had been simultaneously working. It should be noted that notwithstanding the attachment of the tether to the glycerol component, pathway B is still viable for DPTL preparation. Since a number of steps had been optimized and improved, particularly the double diphytanylation, incorporation of labeled material early in the reaction sequence, as retrosynthetic path A requires, was viewed as a satisfactory approach. Scheme 4 shows the steps eventually chosen for this protocol, both in the unlabeled[26] and in the labeled form. The full explanation of the methods follows.

Enantiopure **17** ((*S*)-solketal, (*S*)-(+)-1,2-isopropylideneglycerol) or its mesylate could serve as a protected glycerol for tether attachment. The nucleophilic attack by monobenzyl derivative **7** on the mesylate proved unsuitable but solketal performed efficiently as a nucleophile with the optimized KOH/DMSO conditions affording **18** in 87% yield. Repetition of the protocol with **9**-d_{16}[57] was equally effective, offering **18**-d_{16} in 90% yield. Cleavage of the isopropylidene in **18** was not trivial and many reagents brought about breakdown of the polyether tether. The diol was eventually freed by using 1.0 M HCl in MeOH (74−78%), an outcome comparable in our hands to the Dowex reagent previously employed.[58] The phytanyl groups

(46) Zalipsky, S. *Bioconjugate Chem.* **1995**, *6*, 150−165.
(47) Burkett, B. A.; Chan, T. H. *Synthesis* **2004**, 1007−1010.
(48) It was felt that employing one end of **EG4** as a nucleophile under harsh basic conditions may bring about its breakdown. Hence monoprotection schemes of commercial **EG4** (protiated form) were investigated for eventual creation of electrophilic **EG4** derivatives.
(49) Richard, A.; Marchi-Artzner, V.; Lalloz, M.-N.; Brienne, M.-J.; Artzner, F.; Gulik-Krzywicki, T.; Guedeau-Boudeville, M.-A.; Lehn, J.-M. *Proc. Natl. Acad. Sci.* (*U.S.A.*) **2004**, *101*, 15279−15284.
(50) Azzam, T.; Peppas, N. A.; Slomkowski, S.; Domb, A. J. *Polym. Adv. Technol.* **2002**, *13*, 788−797.
(51) Eguchi, T.; Arakawa, K.; Terachi, T.; Kakinuma, K. *J. Org. Chem.* **1997**, *62*, 1924−1933.
(52) (a) Aoki, T.; Poulter, C. D. *J. Org. Chem.* **1985**, *50*, 5634−5636. (b) Kamikawa, T.; Nogawa, K.; Yamagiwa, Y. *Glycoconjugate J.* **1993**, *10*, 235−239.
(53) Joo, C. N.; Kates, M. *Biochim. Biophys. Acta*, **1969**, *176*, 278−297.
(54) Kates, M.; Palameta, B.; Yengoyan, L. S. *Biochemistry* **1965**, *4*, 1595−1599.
(55) Raguse, B.; Braach-Maksvytis, V.; Cornell, B. A.; King, L. G.; Osman, P. D. J.; Pace, R. J.; Wieczorek, L. *Langmuir* **1998**, *14*, 648−659.
(56) A few trials with the mesylate of diphytanyl glycerol and a monoproptected **EG4** derivative gave no reaction and this option was discarded.
(57) The monobenzylation of **EG4**-d_{16} using the conditions of Scheme 2 occurs in 47% yield, but after recovery of starting material and reduction of a small amount of doubly benzylated **EG4**-d_{16}, the effective yield was 93%.

SCHEME 4. Preparation of DPTL and DPTL-d_{16}



were introduced to diol **19** through the agency of KOH/DMSO, affording fully functionalized glycerol derivative **16** in about 75% yield, a 26% improvement over the previous report, despite employing less alkylating agent.[58]

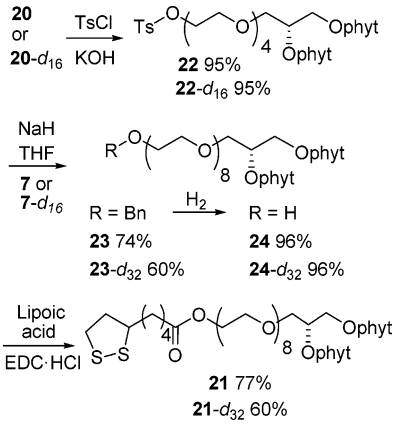
Quantitative hydrogenation readied the tethered material (**20**) for esterification with lipoic acid. With use of unlabeled **20**, the coupling was achieved in 73% after exposure to 1.25 equiv of lipoic acid and 1.3 equiv of carbodiimide coupling agent EDC·HCl. Interestingly, this reaction was stirred for 14 h at room temperature followed by reflux conditions for an additional 18 h. When **20**-d_{16} was converted into **1**-d_{16} and the reaction was stirred 16 h at rt followed by reflux conditions for 72 h, DPTL with the deuterated linker (**1**-d_{16}) was isolated in 93% yield!

The NMR spectra of **1**-d_{16}, though not fully diagnostic of the assigned structure, were nevertheless consistent. Further characterization was found in the IR with strong bands at C−D stretching frequencies of 2182 and 2082 cm^{-1}. The electrospray mass spectrum was also consistent, depicting peaks at *m/z* 1034 ($(M + H)^+$) and 1051 ($(M + NH_4)^+$). A small peak possessing ca. 20% of the intensity of the $(M + H)^+$ peak was observed at *m/z* 1033 and is predicted based on the enrichment of the original **EG**-d_4 (98.3%), but more importantly, indicates no substantial loss of deuterium during the preparation.

Given this successful synthesis and the valuable coupling chemistry of some small EG mesylates and tosylates, we sought to prepare an octameric analogue of DPTL, both unlabeled and possessing a fully deuterated octameric EG linker. Specifically, diphytanyl glycerol octaethylene glycol lipoates (DPOL) **21** and **21**-d_{32} were targeted for IR spectroscopy and neutron reflectivity analysis of water transportation/encapsulation across biomimetic membranes. As with the **EG4** linker of DPTL, the **EG8** tether is expected to demonstrate an affinity for water, but EG oligomers of five or more units are known to coil in a helical fashion with a 7/2 coil parameter and not extend linearly.[59]

The synthetic plan was based closely on the preparation of **1**. A successful octameric linker was prepared through tosylation of **20** and a Williamson connection to monofunctionalized **7** (Scheme 5). The tosylation was efficient (95%), but the coupling was lower yielding. The KOH/DMSO conditions could not be effectively applied and **23** was instead obtained in 74% yield

SCHEME 5. Preparation of 21 and 21-d_{32}



with NaH/THF. The deuterated version affording **23**-d_{32} proceeded at 60%. After debenzylation (96%), lipoic acid condensation was again performed with EDC·HCl. The final reactions were very sluggish and although DPOL (**21**) could be obtained in 77% yield, the **21**-d_{32} reaction required additional reactants and heating for 7 days for a 60% yield.

Consistent with the bonds within each compound, DPOL (**21**) did not contain bands from 2200 to 2050 cm^{-1} but DPOL-d_{32} did possess characteristic C−D stretching frequencies at 2182 and 2082 cm^{-1}. As well, the mass spectral data confirmed that **21** and **21**-d_{32} were prepared and **21**-d_{32} contained the required 32 deuterons. An examination of the spectrum from **21** with EI provided a $(M + H)^+$ peak at *m/z* 1194 and a $(M + NH_4)^+$ signal at *m/z* 1211. The mass spectrum of **21**-d_{32} did not possess intense M^+ or $(M + H)^+$ peaks compared to a dominant $(M + NH_4)^+$ signal of *m/z* 1243. However, expansion of the $(M + H)^+$ region showed that the $(M + H - 1)^+$ peak was 41% of the $(M + H)^+$intensity, proving that no losses of deuterium occurred during the synthesis.

Conclusions

Whereas synthetic applications of OEG-containing materials to linker technology are well established, a protocol for the preparation of a selection of *deuterated* oligo(ethylene glycol) synthetic precursors is introduced herein. The materials are suitable components for elaboration using the methods described and are expected to mesh well with already published protocols for nonlabeled analogues.[35,40,41,46,47] The methods demonstrated are high yielding and are broadly applicable to a variety of fields

(58) Schiller, S.; Naumann, R.; Knoll, W. EP 1364948, 2003.

(59) (a) Harder, P.; Grunze, M.; Dahint, R.; Whitesides, G. M.; Laibinis, P. E. *J. Phys. Chem. B* **1998**, *102*, 426−436. (b) Valiokas, R.; Oestblom, M.; Svedhem, S.; Svensson, S. C. T.; Liedberg, B. *J. Phys. Chem. B* **2000**, *104*, 7565−7569. (c) Vanderah, D. J.; Meuse, C. W.; Silin, V.; Plant, A. L. *Langmuir* **1998**, *14*, 6916−6923.

related to linker technology and may provide new opportunities for mass spectrometry-based pharmacokinetics of tethered drugs and for proteomics.[31]

To exhibit the usefulness of the labeled building blocks, monobenzylated tetra(ethylene glycol) (**7**) was functionalized to prepare labeled and unlabeled DPTL, a popular surface active agent for membrane biomimetic studies. The chemistry achieved herein shows that two synthetic approaches represented in Scheme 1 are viable for the preparation of DPTL. A derivative of DPTL containing an octameric OEG linker has also been synthesized, both in unlabeled and d_{32} form. As a part of this study we have improved the published preparation of DPTL (**1**). Our unlabeled material possesses purity and surface activity adequate for use in a number of surface studies[26,60,61] and the labeled analogue was equally useful.[62]

Experimental Section

The general procedures have been published elsewhere.[63] The experimental procedures for the preparation of nonlabeled DPTL has been published previously.[26] Procedures for most nondeuterated compounds are found in the Supporting Information. **EG**-d_4 (98.3% D) was purchased from C-D-N Isotopes.

Conversion of Ethylene Glycol-d_4 to Higher Oligomers. To a round-bottomed flask was added **EG**-d_4 (6.4 g) and iodine (1.2 mol %). A Dean Stark apparatus was attached and the mixture was placed into a pre-equilibrated oil bath (160−170 °C) for 7.5 h. The mixture was allowed to cool and then starting material was distilled off (38−40 °C, 0.15 mmHg) to give 3.84 g of **EG**-d_4.

Distosylates. To the distillation pot was added dry CH_2Cl_2 (35 mL) and TsCl (7.84 g, 41.1 mmol). The mixture was cooled to −30 °C and powdered KOH (4 equiv, 2.31 g, 41.1 mmol) was added portionwise. The mixture was slowly warmed to rt and stirred overnight. H_2O (100 mL) and enough CH_2Cl_2 (200 mL) were added to dissolve solid material, followed by $Na_2S_2O_3$ (aq) (50 mL). The mixture was extracted with CH_2Cl_2 (2 × 40 mL) and the combined extracts were washed with $Na_2S_2O_3$ (50 mL), H_2O (50 mL), and brine (50 mL) and dried over $MgSO_4$. The solvent was evaporated in vacuo yielding a solid (∼6.5 g) that was chromatographed (flash conditions (20−60% EtOAc/hexanes). Yields are based on recovered **EG**-d_4: **3**-d_8 (3.89 g, 48%), **4**-d_{12} (680 mg, 11%), and **5**-d_{16} (139 mg, 2.7%). The ditosylate of **EG**-d_4 was sometimes obtained when distillation times were minimized. See the Supporting Information for characterization data.

Monobenzylated EG-d_4 (2-d_4). To a flame-dried round-bottomed flask was added **EG**-d_4 (1.14 g, 17.3 mmol), dry CH_2Cl_2 (30 mL), and Ag_2O (1.5 equiv, 5.99 g, 25.8 mmol) and the mixture was heated to 40 °C. Then benzyl bromide (1 equiv, 2.95 g, 17.3 mmol) was added neat and the reaction was stirred overnight. The mixture was filtered through a small pad of silica gel, which was washed liberally with EtOAc, and the combined organics were concentrated in vacuo to yield an oil. Flash chromatography (10−50% EtOAc/hexanes then EtOH) yielded doubly benzylated **EG**-d_4 (676 mg, 16%), **2**-d_4 (1.34 g, 50%), and **EG**-d_4 (91 mg, 8%). See the Supporting Information for characterization data.

Doubly Benzylated Tetra(ethylene glycol)-d_{16} (6-d_{16}). To a flame-dried round-bottomed flask was added KOH (5 equiv, 2.42 g, 43.2 mmol) and dry DMSO (30 mL) and the suspension was stirred for 1 h. The mixture was cooled with ice water and a solution of **2**-d_4 (2 equiv, 2.70 g, 17.3 mmol) and **3**-d_8 (1 equiv, 3.65 g, 8.64 mmol) in dry DMSO (20 mL) was added. The cooling bath was removed and the mixture was stirred for 48 h at rt and then 24 h at 40 °C. After cooling, H_2O (100 mL) was added and the mixture was extracted with EtOAc (4 × 100 mL). The combined organics were washed with H_2O (7 × 100 mL) and brine and dried over $MgSO_4$. Concentration in vacuo yielded **6**-d_{16} as an oil, 3.35 g (99%), which was used directly in the next step. Spectral data: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.34−7.27 (m, 10H), 4.56 (s, 4H); ^{2}H NMR (61 MHz, $CDCl_3$) δ 3.68 (s, 16H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 137.72, 127.57, 126.87, 126.75, 72.27, 68.95 (pent, J_{CD} = 21.3 Hz), 67.91 (pent, J_{CD} = 21.5 Hz); IR (neat) (cm^{-1}) 3088, 3063, 3030, 2932, 2854, 2183, 2082.

Preparation of Tetra(ethylene glycol)-d_{16} (EG4-d_{16}). To a round-bottomed flask was added **6**-d_{16} (4.54 g, 11.6 mmol) and 10% Pd/C (1.0 g) and EtOAc (40 mL). The suspension was degassed (3×) then hydrogen was introduced under balloon pressure (15 to 16 psi) and the reaction was placed into a pre-equilibrated oil bath at 40 °C for 3 days. Then the mixture was filtered through Celite, which was washed liberally with EtOH (400 mL), and the combined organics were concentrated in vacuo yielding 2.40 g (99%) of **EG4**-d_{16} as an oil.[38] Spectral data for **EG4**-d_{16}: ^{1}H NMR (400 MHz, $CDCl_3$) δ 4.15−3.37 (br, 2H); ^{2}H NMR (61 MHz, $CDCl_3$) δ 3.69 (s, 4H), 3.66 (s, 8H), 3.60 (s, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 70.93 (pent, J_{CD} = 21.0 Hz), 68.71 (pent, J_{CD} = 21.5 Hz), 68.35 (pent, J_{CD} = 21.4 Hz), 59.66 (pent, J_{CD} = 21.5 Hz); IR (neat) (cm^{-1}) 3405 (br), 2930, 2205, 2084.

General Procedure for the Preparation of Dibenzyl Perdeuterated Oligomeric EG's (Table 1). To a flame-dried round-bottomed flask was added KOH (5 equiv) and dry DMSO, and the suspension was stirred for 30 to 60 min. The mixture was then cooled in an ice bath for 5 min followed by the addition of a solution of **2**-d_4 or benzyl alcohol (2.0 equiv), and ditosylated OEG's (1.0 equiv) in DMSO was added over 5 min. The mixture was allowed to warm to rt for 1−3 days and then warmed at 40 °C for 1−3 days. See the Supporting Information for isolation and purification procedures and for characterization data.

Preparation of 13-Phenyl-3,6,9,12-tetraoxatridecan-1-ol-d_{16} (7-d_{16}, monobenzylated EG4-d_{16}). To a flame-dried round-bottomed flask was added **EG4**-d_{16} (1.01 g, 4.81 mmol), Ag_2O (1.5 equiv, 1.65 g, mmol), and dry CH_2Cl_2 (50 mL), and the mixture was placed into a pre-equilibrated oil bath at 40 °C. The mixture was allowed to reflux while freshly distilled BnBr (828 mg, 4.84 mmol) was added and the resulting mixture was stirred overnight. The mixture was filtered through a small plug of silica gel, which was washed liberally with EtOH (250 mL), and the combined organics were concentrated in vacuo yielding an oil. Flash chromatography (30% EtOAc/hexanes to 30% EtOH/EtOAc, R_f 0.1 (EtOAc)) yielded **7**-d_{16} in 93% after correction for recovered **EG4**-d_{16} and reductive debenzylation of **6**-d_{16}. Spectral data for **7**-d_{16}: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.35−7.28 (m, 5H), 4.56 (s, 2H), 2.24 (br s, 1H); ^{2}H NMR (61 MHz, $CDCl_3$) δ 3.74−3.56 (m, 16H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 137.6, 127.6, 126.9, 126.8, 72.3, 71.0 (pent, J_{CD} = 21.6 Hz), 68.9 (pent, J_{CD} = 21.1 Hz), 67.9 (pent, J_{CD} = 21.5 Hz), 59.9 (pent, J_{CD} = 21.6 Hz); IR (neat) (cm^{-1}) 3455 (br), 3031, 2857, 2186, 2082. Anal. Calcd for $C_{15}H_8D_{16}O_5$: C 59.98, H 8.05. Found :C 59.99, H 7.96.

1-Phenyl-2,5,8,11-tetraoxatridecan-13-yl Methanesulfonate-d_{16} (9-d_{16}). To a flame-dried round-bottomed flask was added **7**-d_{16} (1.48 g, 4.92 mmol), Et_3N (1.8 equiv, 0.895 g, 8.85 mmol, 1.23 mL), and dry CH_2Cl_2 (10 mL). The mixture was cooled to −30 °C and mesyl chloride (1.5 equiv, 0.845 g, 7.38 mmol) was added over 5 min. The mixture was allowed to slowly warm to rt. After 2.75 h, sat. $NH_4Cl_{(aq)}$ (30 mL) and H_2O (20 mL) were added and the mixture was extracted with EtOAc (3 × 25 mL). The combined organics were washed with sat. $NH_4Cl_{(aq)}$, sat. $NaHCO_{3(aq)}$, sat. $NH_4Cl_{(aq)}$, and brine, dried over $MgSO_4$, and then concentrated in vacuo to yield **9**-d_{16} (1.73 g, 97%) as an oil that was used without further purification. Spectral data for **9**-d_{16}: ^{1}H NMR (400 MHz, $CDCl_3$)

(60) Becucci, L.; Guidelli, R.; Karim, C. B.; Thomas, D. D.; Veglia, G. *Biophys. J.* **2007**, 93, 2678−2687.
(61) Becucci, L.; Santucci, A.; Guidelli, R. *J. Phys. Chem. B*,**2007**, *111*, 9814−9820.
(62) Kunze, J.; Leitch, J.; Schwan, A. L.; Faragher, R. J.; Naumann, R.; Knoll, W.; Dutcher, J. R.; Lipkowski, J. Manuscript in preparation.
(63) Faragher, R. J.; Alberico, D.; Schwan, A. L. *Tetrahedron* **2005**, *61*, 1115−1125.

δ 7.29−7.18 (m, 5H), 4.50 (s, 2H), 3.06 (s, 3H); ^{2}H NMR (61 MHz, $CDCl_3$) δ 4.36 (s, 2H), 3.75 (s, 2H), 3.68−3.61 (m, 12H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 137.8 127.7, 127.0, 126.9, 72.3, 68.9 (br pent, J_{CD} = 21.5 Hz), 68.3 (pent, J_{CD} = 22.3 Hz), 68.0 (pent, J_{CD} = 21.1 Hz), 67.3 (pent, J_{CD} = 21.5 Hz), 36.8.

(4*R*)-2,2-Dimethyl-4-(15-phenyl-2,5,8,11,14-pentaoxapentadec-1-yl)-1,3-dioxolane-*d*$_{16}$ (18-*d*$_{16}$). To a flame-dried round-bottomed flask was added powdered KOH (2.5 equiv, 0.642 g, 11.5 mmol) and dry DMSO (30 mL) and the suspension was allowed to stir for 30 min. The mixture was cooled in an ice bath and a solution of **9**-d_{16} (1 equiv, 1.73 g, 4.58 mmol) and **17** (1 equiv, 0.605 g, 4.58 mmol) in dry DMSO (25 mL) was added. The mixture was then warmed to rt and stirred for 18 h and then warmed at 40 °C for 18 h. H_2O (100 mL) was added and the solution was extracted with EtOAc (4 × 50 mL) and the combined organics were washed with H_2O (6 × 50 mL) and brine, dried over $MgSO_4$, and concentrated in vacuo to yield an oil, 1.71 g (90%), suitable for use in the next step. A small sample was purified from flash chromatography (20−50% EtOAc/hexanes), R_f 0.50 (1:1 EtOAc:hexanes). Spectral data for **18**-d_{16}: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.37−7.28 (m, 5H), 4.59 (s, 2H), 4.30 (app quint, J = 6.0 Hz, 1H), 4.07 (dd, J = 8.3, 6.4 Hz, 1 H), 3.74 (dd, J = 8.3, 6.4 Hz, 1 H), 3.59 (dd, J = 10.0, 5.8 Hz, 1 H), 3.50 (dd, J = 10.0, 5.5 Hz, 1H), 1.44 (s, 3H), 1.38 (s, 3H); ^{2}H NMR (61 MHz, $CDCl_3$) δ 3.63 (br s, 16H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 137.6, 127.4, 126.7, 126.6, 108.2, 73.8, 72.1, 71.3, 69.2 (pent, J_{CD} = 21.6 Hz), 68.8 (pent, J_{CD} = 21.3 Hz), 67.8 (pent, J_{CD} = 21.3 Hz), 65.9, 25.9, 24.6; IR (neat) (cm^{-1}) 3063, 3030, 2986, 2934, 2864, 2184, 2082; $[\alpha]^{24}_D$ +5.96 (CH_2Cl_2, *c* 14.2); MS(EI) *m*/*z* (%) 414 (M+, 4), 399 (27), 272 (80), 257 (32), 229 (100), 185 (10), 173 (13), 161 (10), 145 (20), 132 (27), 101 (31), 91 (64), 84 (77); HRMS calcd for $C_{21}H_{18}D_{16}O_7$ 414.3293, found 414.3299.

(2*S*)-17-Phenyl-4,7,10,13,16-pentaoxaheptadecane-1,2-diol-*d*$_{16}$ (19-*d*$_{16}$). To a round-bottomed flask was added **18**-d_{16} (1.45 g, 3.50 mmol) and MeOH (30 mL) and the mixture was cooled to −40 °C. Then a solution of freshly prepared 1 M HCl in MeOH (31.1 mL, precooled to −40 °C) was added over 2 min and the mixture was slowly warmed over 4 h after which time the mixture was concentrated in vacuo to yield an oil. Flash chromatography (EtOAc to 7:1 EtOAc:EtOH, R_f 0.55 in 9:1 EtOAc/EtOH) yielded **19**-d_{16} as an oil, 0.970 g (74%). Spectral data: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.37−7.28 (m, 5H), 4.58 (s, 2H), 3.87−3.83 (m, 1H), 3.72−3.54 (m, 4 H); ^{2}H NMR (61 MHz, $CDCl_3$) δ 3.67 (br s, 16H); ^{13}C NMR (75 MHz, $CDCl_3$) δ:137.2, 127.4, 126.7, 126.6, 72.1, 71.0 (br), 69.4 (br), 68.7 (br pent, J_{CD} = 21 Hz), 67.6 (br pent, J_{CD} = 21 Hz), 62.0; IR (neat) (cm^{-1}) 3311 (br), 3036, 2929, 2864, 2186, 2083.

(16*S*,25*R*,29*R*)-1-Phenyl-21,25,29,33-tetramethyl-16-[[(7*R*,11*R*)-3,7,11,15-tetramethylhexadecyl]oxy]-2,5,8,11,14,18-hexaoxatetratriacontane-*d*$_{16}$ (16-*d*$_{16}$). To a flame-dried round-bottomed flask was added powdered KOH (5 equiv, 0.710 g, 12.5 mmol) and dry DMSO (25 mL) and the suspension was stirred for 30 min. The mixture was then cooled in an ice bath and a solution of **19**-d_{16} (1 equiv, 0.937 g, 2.50 mmol) and phytanyl mesylate (**12**) (3 equiv, 2.82 g, 7.50 mmol) in dry DMSO (25 mL) was added and the mixture was allowed to warm to rt and stirred at rt for 18 h and at 40 °C for 3 days. H_2O (100 mL) was added and the aqueous mixture was extracted with EtOAc (5 × 30 mL) and the combined organics were washed with H_2O (6 × 50 mL) and brine, dried over $MgSO_4$, and concentrated in vacuo. Flash chromatography (20% EtOAc:hexanes, R_f 0.45 (25% EtOAc/hexanes)) yielded **16**-d_{16} as an oil, 1.68 g (74%). Spectral data: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.34−7.27 (m, 5H), 4.56 (s, 2H), 3.62−3.43 (m, 9H), 1.63−1.47 (m, 6H), 1.38−1.01 (m, 44H), 0.86 (br d, J = 6.6 Hz, 18 H), 0.84 (v br d, J = 6.6 Hz. 12H); ^{2}H NMR (61 MHz, $CDCl_3$) δ 3.64 (br s, 16H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 138.0, 127.9, 127.3, 127.1, 77.7, 72.8, 71.1, 70.7, 70.6, 69.5, 69.4 (pent, J_{CD} = 21.0 Hz), 68.4, 68.3 (pent, J_{CD} = 21.4 Hz), 39.1, 37.2, 37.1, 37.0, 36.9, 36.8, 36.5, 36.4, 32.4, 29.5, 29.4, 27.6, 24.5, 24.2, 24.1, 22.4, 22.3, 19.46, 19.40, 19.36; IR (neat) (cm^{-1}) 3064, 3031, 2953, 2925, 2867, 2181, 2081; $[\alpha]^{24}_D$ −0.308 (CH_2Cl_2, *c* 14.6). Anal. Calcd for $C_{58}H_{94}D_{16}O_7$: C 74.47, H 11.85. Found: C 74.28, H 11.61.

(14*S*,23*R*,27*R*)-19,23,27,31-Tetramethyl-14-[[(7*R*,11*R*)-3,7,11,15-tetramethylhexadecyl]oxy]-3,6,9,12,16-pentaoxadotriacontan-1-ol-*d*$_{16}$ (20-*d*$_{16}$). To a round-bottomed flask was added **16**-d_{16} (1.65 g, 1.76 mmol), EtOAc (60 mL), and 10% Pd/C (0.400 g), and the solution was degassed (3×). Then hydrogen gas was introduced under balloon pressure (15−16 psi) and the mixture was warmed at 40 °C for 13 h. The mixture was filtered through Celite, which was washed liberally with EtOAc, and the combined organics were concentrated in vacuo. Flash chromatography (30% EtOAc/hexanes to EtOAc, R_f 0.1 (50% EtOAc/hexanes)) yielded **20**-d_{16} as an oil, 1.43 g (96%). Spectral data: ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.59−3.45 (m, 9H), 2.51 (br s, 1H), 1.64−1.47 (m, 6H), 1.40−1.00 (m, 42H), 0.86 (br d, J = 6.6 Hz, 18H), 0.84 (v br d, J = 6.6 Hz, 12 H); ^{2}H NMR (61 MHz, $CDCl_3$) δ 3.70−3.56 (br s, 16H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 77.6, 77.7, 71.3 (pent, J_{CD} = 21.3 Hz), 71.0, 70.54, 70.49, 69.4, 69.3 (pent, J_{CD} = 20.8 Hz), 68.3, 60.3 (pent, J_{CD} = 21.3 Hz), 37.0, 36.9, 36.8, 36.7, 36.4, 36.3, 29.4, 29.3, 27.5, 24.4, 24.1, 24.0, 22.34, 22.25, 19.4, 19.3; IR (neat) (cm^{-1}) 3476 (br), 2954, 2930, 2868, 2183, 2082; $[\alpha]^{24}_D$ −0.511 (CH_2Cl_2, *c* 9.22). Anal. Calcd for $C_{51}H_{88}D_{16}O_7$: C 72.46, H 12.40. Found: C 72.38, H 12.31.

(3*R*)-(14*S*,23*R*,27*R*)-19,23,27,31-Tetramethyl-14-[[(7*R*,11*R*)-3,7,11,15-tetramethylhexadecyl]oxy]-3,6,9,12,16-pentaoxadotriacont-1-yl-ester-1,2-dithiolane-3-pentanoate-*d*$_{16}$ (1-*d*$_{16}$, DPTL-*d*$_{16}$). To a flame-dried round-bottomed flask was added lipoic acid (2 equiv, 0.409 g, 1.98 mmol), EDC·HCl (2.5 equiv, 0.475 g, 2.48 mmol), DMAP (cat, 26.7 mg), and dry CH_2Cl_2 (10 mL). Then a solution of **20**-d_{16} (1 equiv, 0.837 g, 0.990 mmol) and Et_3N (2.5 equiv, 0.251 g, 2.48 mmol) in dry CH_2Cl_2 (15 mL) was added and the mixture was stirred at rt for 24 h and warmed at 40 °C for 3 days. Saturated $NH_4Cl_{(aq)}$ (20 mL) and H_2O (20 mL) were added and the solution was extracted with CH_2Cl_2 (4 × 50 mL). The combined organics were washed with sat. $NH_4Cl_{(aq)}$, H_2O, sat. $NaHCO_{3(aq)}$, H_2O, and brine, dried over $MgSO_4$, and concentrated in vacuo. Flask chromatography (20−50% EtOAc/hexanes, R_f 0.13 in 50% EtOAc/hexanes) yielded **1**-d_{16} as an oil, 0.952 g (93%). Spectral data: ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.60−3.45 (m, 9H), 3.23−3.16 (m, 2H), 2.51−2.40 (m, 1H), 2.35 (t, J = 7.5 Hz, 2H), 1.93−1.82 (m, 1H), 1.71−1.03 (m, 55H), 0.86 (br d, J = 6.6 Hz, 18H), 0.84 (v br d, J = 6.6 Hz, 12H); ^{2}H NMR (61 MHz, $CDCl_3$) δ 4.08 (s, 2H), 3.50 (s, 14H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 172.7, 77.6, 71.0, 70.6, 70.5, 69.43, 69.38 (br pent, J_{CD} = 21.1 Hz), 68.3, 67.9 (pent, J_{CD} = 21.6 Hz), 62.3 (pent, J_{CD} = 21.5 Hz), 55.8, 39.8, 39.0, 38.0, 37.1, 37.0, 36.9, 36.8, 36.74, 36.4, 36.3, 34.2, 33.5, 32.4, 29.5, 29.4, 28.3, 27.6, 24.4, 24.2, 24.1, 24.0, 22.4, 22.3, 19.41, 19.35; IR (neat) (cm^{-1}) 2953, 2926, 2867, 2182, 2082, 1737; $[\alpha]^{24}_D$ −0.359 (CH_2Cl_2, *c* 9.22); MS(TOF ES) *m*/*z* (%) 1051 ((M + NH_4)$^+$), 1033 ((M)$^+$); HRMS $C_{59}H_{104}D_{16}NO_8S_2$ calcd 1033.9194, found 1033.9158. Anal. Calcd for $C_{59}H_{100}D_{16}O_8S_2$: C 68.55, H 11.31. Found: C 68.61, H 11.11.

(16*S*,25*R*,29*R*)-19,23,27,31-Tetramethyl-1-*p*-toluenesulfonate-14-[[(7*R*,11*R*)-3,7,11,15-tetramethylhexadecyl]oxy]-3,6,9,12,16-pentaoxadotriacontane-*d*$_{16}$ (22-*d*$_{16}$). To a flame-dried round-bottomed flask was added **20**-d_{16} (1 equiv, 0.564 g, 0.668 mmol), TsCl (1.95 equiv, 0.248 g, 1.30 mmol), and dry CH_2Cl_2 (45 mL). The mixture was cooled to −30 °C and powdered KOH (3.2 equiv, 0.120 g, 2.14 mmol) was added portionwise and the mixture was allowed to warm to rt overnight and was stirred vigorously for 3 days. H_2O (100 mL) was added and the mixture was extracted with CH_2Cl_2 (4 × 50 mL). The combined organics were washed with H_2O (40 mL) and brine, dried over $MgSO_4$, and concentrated in vacuo. Flash chromatography (25−75% EtOAc/hexanes, R_f 0.13 in 50% EtOAc/hexanes) yielded **22**-d_{16} as a slightly yellow oil, 0.634 g (95%). Spectral data: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.80 (d, J = 8.4 Hz, 2H), 7.34 (d, J = 8.4 Hz, 2H), 3.56−3.44 (m, 9H), 2.45 (s, 3H), 1.56−1.49 (m, 6H), 1.37−1.03 (m, 42H), 0.86 (br d,

J = 6.7 Hz, 18H), 0.84 (v br d, J = 6.7 Hz, 12H); ^{2}H NMR (61 MHz, $CDCl_3$) δ 4.14 (s, 2H), 3.67 (s, 2H), 3.62 (s, 8H), 3.56 (s, 4H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 144.2, 132.9, 129.4, 127.6, 77.6, 71.0, 70.6, 69.5, 69.3 (pent, J_{CD} = 21.6 Hz), 68.4, 68.2 (pent, J_{CD} = 21.5 Hz), 67.4 (pent, J_{CD} = 21.6 Hz), 39.0, 37.2, 37.1, 36.9, 36.8, 36.44, 36.35, 32.4, 29.5, 29.4, 27.6, 24.4, 24.1, 24.0, 22.4, 22.3, 21.2, 19.44, 19.37, 19.31; IR (neat) (cm^{-1}) 3030, 2954, 2926, 2867, 2186, 2083, 1598; $[\alpha]^{24}_D$ −0.441 (CH_2Cl_2, *c* 3.54). Anal. Calcd for $C_{58}H_{94}D_{16}O_9S$: C 69.69, H 11.09. Found: C 69.61, H 11.12.

(28*S*,27*R*,41*R*)-33,37,41,45-Tetramethyl-1-phenyl-28-[[(7*R*,-11*R*)-3,7,11,15-tetramethylhexadecyl]oxy]-2,5,8,11,14,17,20,23,-26,30-decaoxahexatetracontane-*d*$_{32}$ (23-*d*$_{32}$). To a flame-dried round-bottomed flask was added NaH (4.0 equiv, 81.0 mg, 2.03 mmol), which was washed with hexanes (2 × 25 mL), and then dry THF (10 mL) was added. The suspension was cooled to −78 °C and a solution of **22**-d_{16} (0.506 g, 0.506 mmol) and **7**-d_{16} (0.152 g, 0.506 mmol) in dry THF (20 mL) was added dropwise. The mixture was slowly warmed to rt and stirred for 3 days at rt and then warmed at 40 °C for 3 h. H_2O (20 mL) was added and the mixture was extracted with EtOAc (4 × 25 mL) and the combined organics were washed with H_2O (20 mL) and brine, dried over $MgSO_4$, and concentrated in vacuo. Flash chromatography (20−75% EtOAc/hexanes then 50% EtOH/EtOAc, R_f 0.1 (EtOAc)) yielded **23**-d_{32} as an oil, 0.346 g (60%). Spectral data: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.34−7.27 (m, 5H), 4.56 (s, 2H), 3.61−3.43 (m, 9H), 1.37−0.98 (m, 42H), 0.86 (br d, J = 6.6 Hz, 18H), 0.84 (v br d, J = 6.6 Hz, 12H); ^{2}H NMR (61 MHz, $CDCl_3$) δ 3.58 (br s, 32 H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 138.1, 128.1, 127.5, 127.3, 77.7, 72.9, 71.2, 70.7, 69.7, 69.5 (br pent, J_{CD} = 21.4 Hz), 68.6 (br), 68.4 (pent, J_{CD} = 21.6 Hz), 39.2, 37.3, 37.2, 37.1, 37.0, 36.9, 36.6, 36.5, 32.6, 29.7, 29.6, 27.8, 24.6, 24.3, 24.2, 22.54, 22.45, 19.57, 19.51; IR (neat) (cm^{-1}) 3090, 3066, 3030, 2955, 2930, 2865, 2180, 2082; $[\alpha]^{24}_D$ −0.161 (CH_2Cl_2, *c* 4.77). Anal. Calcd for $C_{66}H_{94}D_{32}O_{11}$: C 70.29, H 11.26. Found: C 70.48, H 11.05.

(26*S*,35*R*,39*R*)-31,35,39,51-Tetramethyl-26-[[(7*R*,11*R*)-3,7,11,-15-tetramethylhexadecyl]oxy]-3,6,9,12,15,18,21,24,28-nonaoxatetratetracontan-1-ol-*d*$_{32}$ (24-*d*$_{32}$). To a round-bottomed flask was added **23**-d_{32} (0.295 g, 0.265 mmol), EtOAc (20 mL), and 10% Pd/C (75 mg), and the mixture was degassed (3×). Then hydrogen gas was introduced under balloon pressure (15−16 psi) and the mixture was warmed at 40 °C for 2.5 h. The mixture was filtered over Celite, which was washed liberally with EtOAc, and combined organics were concentrated in vacuo. Flash chromatography (EtOAc to 9:1 EtOAc:EtOH), R_f 0.5 (9:1 EtOAc:EtOH), yielded **24**-d_{32} as an oil, 0.265 g (95%). Spectral data: ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.60−3.40 (m, 9H), 3.11 (br s, 1H), 1.60−1.53 (m, 2H), 1.49 (app heptet, J = 6.6 Hz, 4H), 1.34−1.02 (m, 42H), 0.83 (br d, J = 6.6 Hz, 18H), 0.81 (v br d, J = 6.6 Hz, 12H); ^{2}H NMR (61 MHz, $CDCl_3$) δ 3.60 (br s, 32H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 77.2, 71.5 (pent, J_{CD} = 21.6 Hz), 71.3, 70.9, 70.8, 69.9, 69.5 (br pent, J_{CD} = 21.2 Hz), 68.8 (br), 60.8 (pent, J_{CD} = 21.6 Hz), 39.3, 37.4, 37.32, 37.30, 37.2, 37.1, 37.0, 36.7, 36.6, 32.7, 29.8, 29.74, 29.72, 29.6, 27.9, 24.7, 24.4, 24.3, 22.6, 22.5, 19.7, 19.60, 19.55, 19.5; IR (neat) (cm^{-1}) 3479 (br), 2954, 2926, 2868, 2182, 2082; $[\alpha]^{24}_D$ −0.160 (CH_2Cl_2, *c* 4.96); MS(ESI) *m*/*z* (%) 1055 ($(M + NH_4)^+$), 1038 ($(M + H)^+$); HRMS calcd for $C_{59}H_{92}D_{32}NO_{11}$ 1055.1183, found 1055.1232.

(3*R*)-(26*S*,35*R*,39*R*)-31,35,39,51-Tetramethyl-26-[[(7*R*,11*R*)-3,7,11,15-tetramethylhexadecyl]oxy]-3,6,9,12,15,18,21,24,28-nonaoxatetratetracontan-1-ylester-1,2-dithiolane-3-pentanoate-*d*$_{32}$ (21-*d*$_{32}$). To a flame-dried round-bottomed flask was added lipoic acid (2.0 equiv, 58.0 mg, 0.280 mmol), EDC·HCl (2.5 equiv, 67.0 mg, 0.350 mmol), DMAP (cat, 3 mg), and dry CH_2Cl_2 (5 mL). A solution of **24**-d_{32} (1.0 equiv, 0.145 g, 0.140 mmol) and Et_3N (2.5 equiv, 35 mg, 0.350 mmol) in dry CH_2Cl_2 (10 mL) was added and the mixture was stirred at rt for 24 h and refluxed for 3 days. Then an additional amount of lipoic acid (45 mg) and EDC·HCl (55 mg) was added and stirring was continued at reflux for 4 days. Saturated $NH_4Cl_{(aq)}$ (10 mL) and H_2O (10 mL) were added and the aqueous solution was extracted with CH_2Cl_2 (4 × 25 mL). The combined organics were washed with sat. $NH_4Cl_{(aq)}$, H_2O, sat. $NaHCO_{3(aq)}$, H_2O, and brine, dried over $MgSO_4$, and concentrated in vacuo. Flash chromatography (1:1 EtOAc:hexanes then 1:1 EtOAc:EtOH) yielded **21**-d_{32} as a yellow oil, 98 mg (60% after recovery of **24**-d_{32} (28 mg)). Spectral data for **21**-d_{32}: ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.64−3.42 (m, 9H), 3.21−3.08 (m, 2H), 2.47 (app pent, J = 6.3 Hz, 0.5H), 2.45 (app pent, J = 6.3 Hz, 0.5H), 2.35 (t, J = 7.4 Hz, 2H), 1.91 (app hextet, J = 6.9 Hz, 1H), 1.71−1.42 (m, 13H), 1.41−1.01 (m, 42H), 0.86 (br d, J = 6.7 Hz, 18H), 0.84 (v br d, J = 6.7 Hz, 12H); ^{2}H NMR (61 MHz, $CDCl_3$) δ 4.18 (s, 2H), 3.59 (s, 30H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 173.3, 77.8, 71.3, 70.81, 70.76, 69.8, 69.6 (pent, J_{CD} = 21.5 Hz), 68.9, 68.8, 68.2 (br pent, J_{CD} = 22.0 Hz), 62.6 (pent, J_{CD} = 22.4 Hz), 56.2, 40.1, 39.3, 38.4, 37.4, 37.29, 37.28, 37.24, 37.2, 37.1, 37.0, 36.64, 36.56, 34.5, 33.8, 32.7, 29.8, 29.72, 29.69, 28.6, 27.9, 24.7, 24.5, 24.4, 24.3, 22.6, 22.5, 19.7 19.6, 19.54, 19.52; IR (neat) (cm^{-1}) 2953, 2925, 2867, 2181, 2082, 1736; $[\alpha]^{24}_D$ −0.284 (CH_2Cl_2, *c* 2.75); ES MS (+) *m*/*z* (%) 1243 ($(M + NH_4)^+$); HR TOF ES MS(+) $C_{67}H_{100}D_{32}O_{12}S_2$ ($(M + H)^+$), calcd 1226.1247, found 1226.1199; $C_{67}H_{104}D_{32}NO_{12}S_2$ ($(M + NH_4)^+$), calcd 1243.1513, found 1243.1506. Anal. Calcd for $C_{67}H_{100}D_{32}O_{12}S_2$: C 65.64, H 10.85. Found: C 66.20, H 10.44.

Acknowledgment. The authors are indebted to the Natural Sciences and Engineering Research Council for funding this project. The authors also thank Dr. Julia Kunze, Prof. Jacek Lipkowski, and the Lipkowski group for their guidance and their confidence in us. R.J.F. thanks the Ontario Government for a graduate scholarship.

Supporting Information Available: Additional experimental procedures and copies of ^{1}H and ^{13}C NMR spectra for nondeuterated and selected deuterated compounds. This material is available free of charge via the Internet at http://pubs.acs.org.

JO701979Z