# Darmstoff Analogues. 2. Ring and Side-Chain Effects on Smooth-Muscle Contraction

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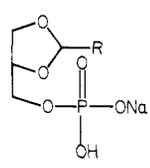
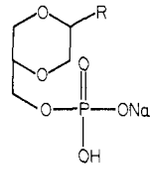
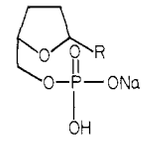
2-*cis*-$\Delta^8$-Heptadecenyl-4-(hydroxymethyl)-1,3-dioxolane monosodium phosphate (**1b**) has been shown to be present as a major component of Darmstoff in mammalian intestine and to be a potent inducer for contraction of intestinal smooth muscle. The analogous 2-pentadecyl material **1a**, also found abundantly in the intestine, is inactive. In this study, synthesis of phosphorylated hydroxymethyl-1,4-dioxanes, -tetrahydrofurans, -cyclopentanes, and -oxathiolanes bearing both oleyl and palmityl side chains is reported. Of these, 2-(hydroxymethyl)-5-*cis*-$\Delta^8$-heptadecenyl-1,4-dioxane monosodium phosphate (**2b**) exhibits about 12% of the activity of **1b**. Its pentadecyl analogue **2a**, like **1a**, is totally inactive, as are all other compounds prepared. The results indicate that Darmstoff-like compounds exhibit specific chemical requirements for activity and that where activity is encountered, the side-chain specificity noted in **1a** and **1b** can be preserved.

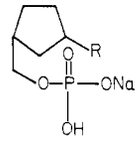
The mixture of acetal phosphatidic acids known as Darmstoff has been shown to contain primarily **1a** and

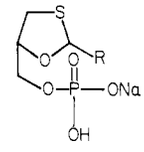






**1a**, R = $C_{15}H_{31}$ **2a**, R = $C_{15}H_{31}$ **3a**, R = $C_{15}H_{31}$
**b**, R = $C_{17}H_{33}$ **b**, R = $C_{17}H_{33}$ **b**, R = $C_{17}H_{33}$



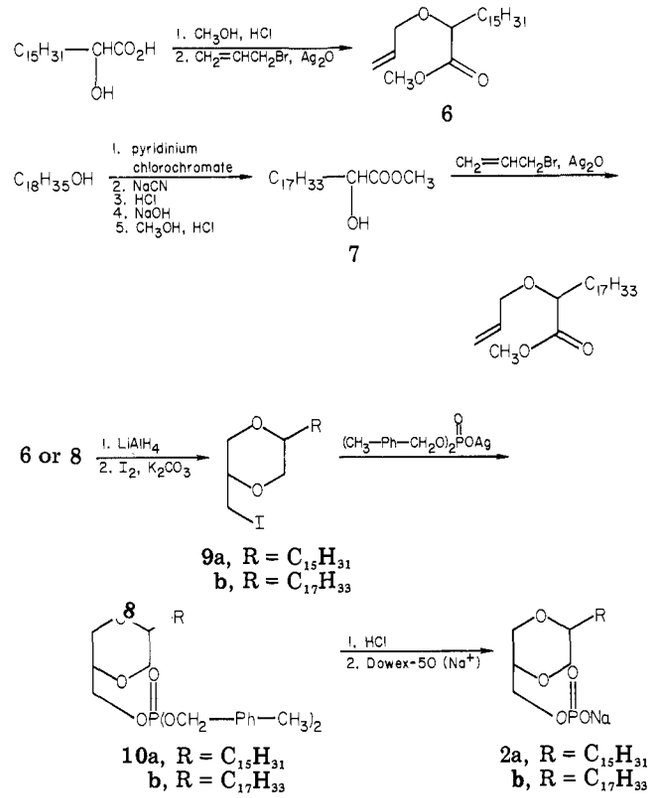


**4a**, R = $C_{15}H_{31}$ **5a**, R = $C_{15}H_{31}$
**b**, R = $C_{17}H_{33}$ **b**, R = $C_{17}H_{33}$

**1b**.[1,2] Remarkably, all the intestinal smooth muscle contracting activity possessed by Darmstoff resides in the oleyl acetal **1b**.[1,2] These compounds also display hypotensive and cardiodepressant properties;[3] the generally similar ether phospholipids recently described by Hanahan[4] and Muirhead[5] show hypotensive properties and cause platelet aggregation as well. Since it thus appears that phospholipids may exhibit specific pharmacological properties influenced by rather subtle structural changes, it was of interest to determine whether the significant side-chain specificity observed in **1** for smooth-muscle contraction would carry over into other compounds. Also, it was a goal of this work to determine to what extent deviation from the natural dioxolane ring would be consistent with good biological effect. The synthesis of compounds **3a** and **4a**, as well as their cardiodepressant properties, has been described earlier.[3] The synthesis of the other compounds and the ability of all compounds to induce guinea pig ileum contraction are reported here.

**Chemistry.** The key step in the successful synthesis of the dioxane analogue **2** is the iodine-induced cyclization of the alcohols obtained from **6** and **8** (Scheme I) to afford

Scheme I



the iodomelthyldioxanes **9**. This reaction is known to proceed much better when the product is a five-membered cyclic ether than when a six-membered ring is involved. After some difficulty, **9a** was obtained using Whittaker's heterogeneous reaction procedure[7] and a large excess of $I_2$. In the case of **9b**, excess $I_2$ reacted with the side-chain olefin. Since it was known[8] that dimethyl sulfoxide greatly accelerated the transformation of olefins into bromohydrins, it was felt that use of this solvent might be advantageous here also, since a halonium intermediate is probably involved in both cases. This was successful, and **9b** was obtained in reasonable yield using only 1.1 equiv of $I_2$. It is apparent that **9a** and **9b**, as well as many other compounds described here, can exist in cis and trans forms. In our Darmstoff synthesis,[1] we determined in a model

(1) W. Vogt, *Biochem. Pharmacol.*, **12**, 415 (1963).
(2) R. A. Wiley, D. D. Sumner, and E. J. Walaszek, *Lipids*, **5**, 803 (1970).
(3) A. N. Milbert and R. A. Wiley, *J. Med. Chem.*, **21**, 245 (1978).
(4) C. A. Demopoulos, R. N. Pinckard, and D. J. Hanahan, *J. Biol. Chem.*, **234**, 9355 (1979).
(5) M. L. Blank, F. Synder, L. W. Byers, B. Brooks, and E. E. Muirhead, *Biochem. Biophys. Res. Commun.*, **90**, 1194 (1978).
(6) M. L. Mihailovic, *Lect. Heterocycl. Chem.*, **3**, 111 (1976).
(7) N. Whittaker, *Tetrahedron Lett.*, 2805 (1977).
(8) D. R. Dalton, R. C. Smith, and D. C. Jones, *Tetrahedron*, **26**, 575 (1970).

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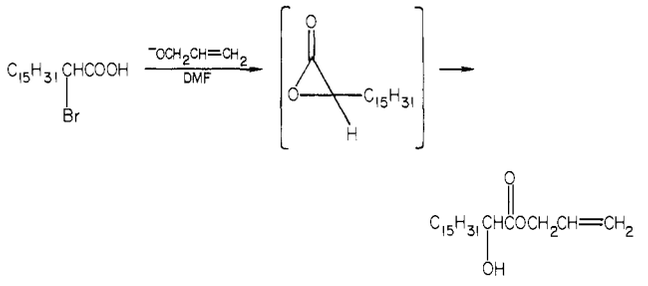
system (R = $C_3H_7$) that such dioxolane isomers are separable only by careful preparative gas chromatography and not by ohter chromatographic methods. The NMR spectra of the 5-spin system for protons $\alpha$ to oxygen are very complex, even in the pure isomers. In the present work, it must be presumed that cis and trans isomers are present in equal amounts, but no separation was achieved by any chromatographic method used. NMR signals for protons $\alpha$ to heteroatoms were, as expected, multiplets, which did not allow assignment of signals to specific protons.

The required allyloxy esters **6** and **8** were most conveniently obtained from the corresponding hydroxy esters and allyl bromide, using $Ag_2O$ as catalyst.[9] Attempted reaction of the hydroxy acids themselves with allyl bromide was unsuccessful, and it was observed that reaction of sodium allyl oxide and the bromo acid shown below resulted in formation of the allyl ester, apparently via the $\alpha$-lactone. This was surprising in view of the fact that



hydroxide ion is known to attack $\alpha$-lactones at the alkyl carbon and may have resulted from the steric requirements of the alkyl chain and the reagent.

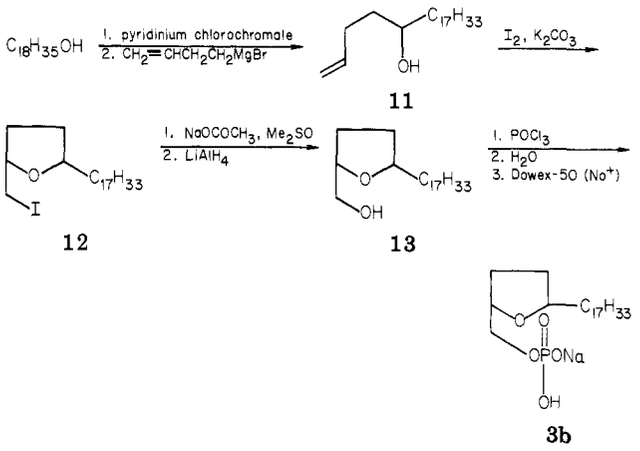
Phosphorylation to obtain **2** was most conveniently carried out directly on the iodides, using silver bis(*p*-methylbenzyl) phosphate.[10] Treatment of the intermediate esters **10** with dry HCl in chloroform gave the phosphates **2**, which were converted to sodium salts to facilitate purification and testing.

The synthesis of the oleyltetrahydrofuran phosphate **3b** was carried out as shown in Scheme II. As expected, cyclization to afford **12** was very facile, but attempted phosphorylation using the method above led to a complex mixture of products. Thus, the iodide was converted to the hydroxymethyl compound **13** via the acetate ester. This was then phosphorylated using $POCl_3$; it was found helpful to use *p*-(dimethylamino)pyridine as a basic catalyst.

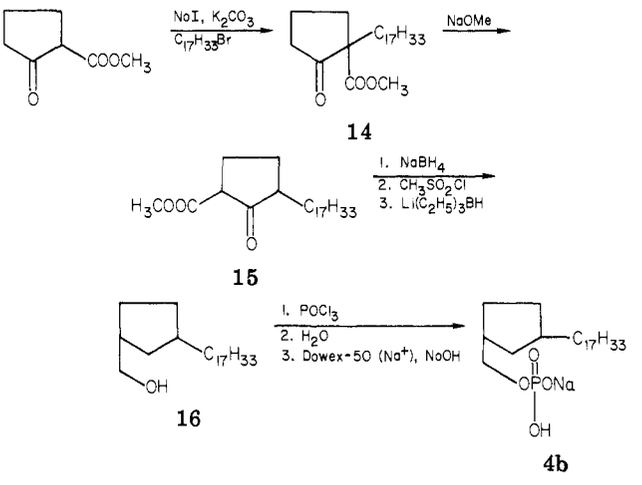
The cyclopentane analogue was prepared as shown in Scheme III. It was necessary to add 1 equiv of NaI to obtain **14** in good yield, and attempts to introduce the required hydroxymethyl substituent directly were unsuccessful. Advantage was therefore taken of the fact that treatment of compounds like **14** with base results in cleavage; it the alcohol is removed by distillation, recyclization to afford the 2,5-disubstituted compound will occur,[12] apparently because its enolate is highly resistant to nucleophilic attack. Reduction, removal of the carbonyl group, and phosphorylation were carried out in conventional fashion.

The oxathiolane compounds were obtained as shown in

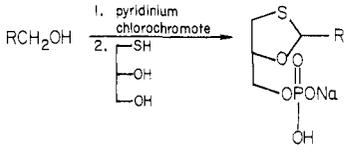
Scheme II



Scheme III

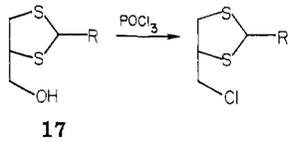


Scheme IV



**5a**, R = $C_{15}H_{31}$
**b**, R = $C_{17}H_{33}$

Scheme IV. Literature reports indicate that condensation of the mercaptodiol with aldehydes affords only oxathiolanes, but in our hands a substantial amount of the corresponding oxathiane was formed as well. These were separated with some difficulty by chromatographic means, and the oxathiolane was phosphorylated as shown. Synthesis of the analogous dithiolane materials was also tried, but attempted phosphorylation of the thioacetals **17** resulted only in formation of the chloro thioacetal. This apparently arose from the intramolecular participation by the sulfur attached to C-2 at the dichlorophosphate stage. Although the chloro compound could be used to produce phosphate esters analogous to **10**, these were unstable to the hydrolysis conditions.



**17**

(9) P. A. Lavene and P. S. Yang, *J. Biol. Chem.*, **102**, 557 (1933).
(10) J. G. Malotovsky, L. F. Nikulina, and L. D. Bergelson, *Chem. Phys. Lipids*, **17**, 108 (1976).
(11) G. Hofle, W. Steglich, and H. Vorbruggen, *Angew. Chem., Int. Ed. Engl.*, **17**, 569 (1978).
(12) K. Sisido, K. Utinoto, and T. Isida, *J. Org. Chem.*, **29**, 2781 (1964).

## Biological Results

Compounds **2a,b**, **3a,b**, **4a,b**, and **5a,b** were tested for their ability to cause contraction of guinea pig ileum. Only the oleyldioxane **2b** was active. The concentration required for half-maximal contraction was 16.0 mM; the potency relative to acetylcholine is $5.02 \times 10^{-4}$ (0.05%). The comparable ratio for the dioxolane **1b** is $4.06 \times 10^{-3}$ (0.4%), so that **2b** is about one-eighth as active at **1b**. Like **1a**, the palmityl analogue **2a** is totally inactive. These results indicate that the side-chain specificity previously observed with Darmstoff (**1**) carries over into other closely related ring systems and that in order to be active as intestinal smooth muscle contracting agents, cyclic phospholipids of this type must very closely resemble natural Darmstoff. It is surprising that no activity was observed in the oxathiolane **5b**, since the differences between sulfur and oxygen would seem subtle and related mostly to small changes in atomic size. No definitive explanation is available, but the difference in chemical behavior between **17** and its oxathiolane analogue emphasizes that substitution of sulfur for oxygen might cause important differences in the electronic structure and chemical behavior of the molecules.

## Experimental Section

Melting points (uncorrected) were determined on a Thomas-Hoover apparatus. Elemental microanalyses were performed on a Hewlett-Packard Model 185B analyzer at the University of Kansas. Where analyses are indicated by symbols of the elements, experimental results were within 0.4% of the calculated values. All compounds exhibited IR spectra consistent with the structures assigned, and all compounds except salts were also characterized by NMR.

**Methyl 2-(Allyloxy)heptadecanoate (6).** In a flask equipped with a magnetic stirrer and a drying tube was placed 3.8 g (0.013 mol) of 2-hydroxyheptadecanoic acid[13] and 125 mL of 3% methanolic HCl (prepared from anhydrous methanol and acetyl chloride). The solution was stirred for 18 h at room temperature, and the solvent was removed. $CH_2Cl_2$ (150 mL) was added and evaporated to remove water as an azeotropic mixture; this gave the methyl ester as a white solid. This was dissolved in 50 mL of benzene, 10 mL (0.116 mol) of allyl bromide (freshly distilled) and 3.07 g (0.013 mol) of $Ag_2O$[14] were added, and the mixture was heated under reflux for 18 h with stirring. The mixture was then cooled, and the inorganic salts were removed by filtration through a short pad of Florisil. The solvent was evaporated to give a yellow oil, which was purified by column chromatography on silica gel (50:50 benzene/hexane), followed by vacuum distillation, to give 2.23 g (51.2%) of **6** as a colorless oil, bp 146–151 °C (0.2 mm). Anal. ($C_{21}H_{40}O_3$) C, H.

**Methyl 2-Hydroxy-*cis*-$\Delta^{10}$-nonadecenoate (7).** In a flask equipped with a magnetic stirrer and a drying tube was placed 7.42 g (34.5 mmol) of pyridinium chlorochromate in 50 mL of methylene chloride. To this stirred suspension was added 6.2 g (23 mmol) of oleyl alcohol in one portion. The mixture was stirred at room temperature for 1.5 h. The tarry reaction mixture was diluted with 300 mL of dry ether, the solution was filtered through a pad of Florisil, and the filtrate was evaporated to give the crude aldehyde. This was stirred with 2.91 g (28 mmol) of sodium bisulfite and 1.47 g (30 mmol) of sodium cyanide in 300 mL of water for 20 h at room temperature. The organic product was extracted with 2–150 mL portions of ether. The combined ether extracts were dried ($Na_2SO_4$) and filtered, and the filtrate was evaporated to give the crude cyanohydrin. This was treated with 150 mL of concentrated HCl, and the mixture was stirred at room temperature for 18 h in a well-ventilated hood. The mixture was diluted with 300 mL of water and extracted with 2–300 mL portions of $CHCl_3$. The combined organic layers were dried ($Na_2SO_4$) and filtered, and the filtrate was evaporated to give a yellow semisolid. The residue was then heated under reflux for 20 h with 12.2 g (0.217 mol) of potassium hydroxide and 150 mL of 65% aqueous ethanol. Most of the ethanol was removed by distillation, and the mixture was cooled and diluted with 200 mL of $H_2O$. The cloudy aqueous solution was acidified to pH 1 with concentrated $H_2SO_4$. The organic product was extracted with 2–100 mL portions of ether. The combined ether extracts were dried ($Na_2SO_4$) and filtered, and the filtrate was evaporated to give the crude hydroxy acid. This was dissolved in 125 mL of 3% methanolic HCl and stirred at room temperature for 18 h. The methanol was removed, and the residue treated with 150 mL of $CH_2Cl_2$, followed by evaporation to remove the water produced. The crude product was purified by medium-pressure liquid chromatography on silica gel (hexane/ethyl acetate, 9:1) to give 2.3 g (30.4% overall yield) of **7** as a light yellow oil. Anal. ($C_{20}H_{38}O_3$) C, H.

**Methyl 2-(Allyloxy)-*cis*-$\Delta^{10}$-nonadecenoate (8).** To a solution of 1.83 g (5.6 mmol) of **7** and 30 mL of benzene was added 4.2 mL (48.6 mmol) of allyl bromide and 1.3 g (5.6 mmol) of $Ag_2O$. The reaction mixture was heated under reflux with stirring for 14 h. The mixture was then cooled to room temperature, and the inorganic salts were removed by filtration through a sintered glass funnel. The filter cake was washed with 100 mL of ether, and the organic solution was evaporated to dryness to give a yellow oil, which was purified by column chromatography on silica gel (benzene/hexane, 50:50), followed by distillation to give 1.08 g (52.5%) of **8** as a colorless oil, bp 165–171 °C (0.4 mm). Anal. ($C_{23}H_{42}O_3$) C, H.

**2-(Iodomethyl)-5-pentadecyl-1,4-dioxane (9a).** To a mixture of 50 mL of anhyrous ether and 0.23 g (6.17 mmol) of $LiAlH_4$ under a nitrogen atmosphere was added dropwise a solution of 2.1 g (6.17 mmol) of **6** in 10 mL of ether over 10 min. The reaction mixture was stirred at room temperature for 30 min and worked up by the method of Micovic.[15] To the reaction mixture was added 0.23 mL of $H_2O$, 0.23 mL of 15% aqueous NaOH, and 0.69 mL of $H_2O$ consecutively with constant stirring. The precipitate was removed, and the ether solution was evaporated to dryness to give the corresponding alcohol. To this alcohol (2.47 g, 7.92 mmol) was added 10.1 g (39.6 mmol) of iodine, 10.9 g (79.2 mmol) of anhydrous $K_2CO_3$, and 200 mL of 50% aqueous ether. This mixture was stirred at room temperature for 48 h and then heated for 6 h in an oil bath at 60 °C. TLC revealed that the starting material was almost completely consumed. Heating was discontinued, and the mixture was allowed to stir overnight. Excess iodine was removed by the addition of solid $Na_2S_2O_3$. The ether layer was separated, and the aqueous layer was extracted with 100 mL of $CH_2Cl_2$. The combined organic layers were dried ($MgSO_4$) and filtered, and the filtrate was evaporated to give a yellow solid, which was purified by column chromatography on silica gel (benzene/hexane, 50:50) to give 2.01 g (57.93%) of **9a** as a white solid. An analytical sample was obtained by recrystallization from methanol, mp 76–83 °C. Anal. ($C_{20}H_{39}IO_2$) C, H.

**2-(Iodomethyl)-5-*cis*-$\Delta^8$-heptadecenyl-1,4-dioxane (9b).** Reduction of 2.73 mmol of **8** was carried out by the same procedure as that described for compound **6** to give the corresponding alcohol as an oil. A mixture of 1.26 g (3.72 mmol) of this alcohol, 1.33 g (5.24 mmol) of $I_2$, and 30 mL of $Me_2SO$ was heated in an oil bath at 60 °C for 48 h. The dark solution was cooled and treated with 20 mL of 6% aqueous $Na_2S_2O_3$ and 20 mL of saturated NaCl solution. The mixture was then extracted with 5–15 mL portions of ether. The combined ether extracts were dried ($MgSO_4$) and filtered, and the filtrate was evaporated to give a dark brown oil, which was purified by column chromatography on silica gel (benzene/hexane, 50:50) to give 0.54 g (31.2%) of **9b** as a low-melting solid. An analytical sample was obtained by preparative TLC on silica gel (hexane/ether, 9:1). Anal. ($C_{22}H_{41}IO_2$) C, H.

**2-(Hydroxymethyl)-5-pentadecyl-1,4-dioxane Bis(*p*-methylbenzyl) Phosphate (10a).** A heterogeneous mixture of

(13) H. R. LeSeur, *J. Chem. Soc.*, **85**, 827 (1904).
(14) R. Wilstatter and A. Pfannensteil, *Chem. Ber.*, **37**, 4744 (1904).
(15) V. K. Micovic and M. L. J. Mihalovic, *J. Org. Chem.*, **18**, 1190 (1953).

0.25 g (0.583 mmol) of **9a**, 0.35 g (0.875 mmol) of the silver salt of bis(*p*-methylbenzyl) phosphate,[10] and 20 mL of benzene was heated under reflux for 24 h. The benzene was removed, and the residue was triturated with 30 mL of ether. The inorganic material was removed by filtration through a pad of Florisil. The ether solution was evaporated to give a solid material, which was a nonpolar impurity. The filter cake was then washed with 75 mL of ethyl acetate to give the crude triester **10a**, which was purified by preparative TLC on silica gel (hexane/ethyl acetate, 70:30) to give 0.193 g (54%) of **10a** as a low-melting solid. Anal. ($C_{36}H_{57}O_6P$) C, H.

**2-(Hydroxymethyl)-5-*cis*-$\Delta^8$-heptadecenyl-1,4-dioxane Bis(*p*-methylbenzyl) Phosphate (10b).** A mixture of 0.32 g (0.689 mmol) of **9b**, 0.43 g (1.03 mmol) of the silver salt of bis-(*p*-methylbenzyl) phosphate,[10] and 20 mL of toluene was heated under reflux for 20 h. After the mixture was cooled and the toluene removed, the residue was triturated with 30 mL of ether, and the inorganic material was removed by filtration through a pad of Florisil. The filter cake was then washed with 100 mL of ethyl acetate. The ethyl acetate solution was evaporated to give a yellow oil, which was purified by preparative TLC on silica gel (hexane/ethyl acetate, 60:40) to yield 0.32 g (72.7%) of **10b** as a light yellow oil. Anal. ($C_{38}H_{59}O_6P$) C, H.

**2-(Hydroxymethyl)-5-pentadecyl-1,4-dioxane Monosodium Phosphate (2a).** A solution of 0.1844 g (0.299 mmol) of **10a** in 25 mL of $CHCl_3$ was cooled in an ice bath, and anhydrous HCl was bubbled through the solution with magnetic stirring for 1.5 h. TLC revealed that the starting material had disappeared. The solvent was evaporated to give 0.13 g of the dihydrogen phosphate as a low-melting solid. This was converted to the monosodium salt **2a** by batch treatment with Dowex-50 ($Na^+$) resin, followed by lyophilization to give 0.10 g (78%) of **2a** as a white solid. Anal. ($C_{20}H_{40}O_6PNa$) C, H.

**2-(Hydroxymethyl)-5-*cis*-$\Delta^8$-heptadecenyl-1,4-dioxane Monosodium Phosphate (2b).** The phosphotriester **10b**, 0.23 g (0.358 mmol) was hydrolyzed by the same procedure described for compound **10a**. The yield of the monosodium salt **2b** was 0.14 g (80%). Anal. ($C_{22}H_{42}O_6PNa$) H; C: calcd, 57.89; found, 57.00.

***cis*-1,13-Docasadien-5-ol (11).** To a mixture of 5.99 g (27.9 mmol) of pyridinium chlorochromate and 30 mL of methylene chloride was added 5.0 g (18.6 mmol) of oleyl alcohol in 20 mL of $CH_2Cl_2$. The mixture was stirred at room temperature for 1.5 h and then diluted with 300 mL of anhydrous ether; the solution was filtered through a pad of Florisil. The tarry residue was washed with 3–50 mL portions of ether. The filtrate was evaporated to dryness to give 4.77 g (96%) of crude oleyl aldehyde. In a dry flask was placed 0.61 g (0.025 mol) of magnesium turnings, 0.1 g of $I_2$, and 50 mL of anhydrous ether. This mixture was stirred, four drops of 1-bromo-4-butene were added, and stirring was continued until the iodine color disappeared. Then the remainder of 2.4 mL (0.024 mol) of 1-bromo-4-butene diluted with 15 mL of ether was added dropwise with stirring over 20 min. The mixture was stirred at room temperature until reaction had ceased (approximately 30 min). Then 4.0 g (0.015 mol) of the crude oleyl aldehyde was added dropwise in 20 mL of ether over 20 min with constant stirring. After the addition, the mixture was heated under reflux for 1 h. TLC (silica gel; hexane/ether, 80:20) revealed the disappearance of starting material; then 50 mL of 10% aqueous ammonium chloride was added slowly. The ether layer was separated, and the aqueous phase was extracted with 3–50 mL portions of ether. The combined ether layers were dried ($MgSO_4$) and filtered, and the filtrate was evaporated to give a yellow oil, which was purified by medium-pressure liquid chromatography and distillation to give 2.1 g (43.4%) of **11** as a colorless oil, bp 145–159 °C (0.3 mm). Anal. ($C_{22}H_{42}O$) C, H.

**2-*cis*-$\Delta^8$-Heptadecenyl-5-(iodomethyl)tetrahydrofuran (12).** To a mixture of 0.74 g (2.3 mmol) of **11**, 3.18 g (23 mmol) of $K_2CO_3$, and 30 mL of $CH_2Cl_2$ was added 0.7 g (2.76 mmol) of $I_2$. The reaction mixture was stirred at room temperature for 15 h. TLC (silica gel; hexane/ether, 80:20) revealed that starting material had disappeared. The reaction mixture was treated with 20 mL of $H_2O$, and the excess $I_2$ was decomposed with $Na_2S_2O_3$. The $CH_2Cl_2$ layer was separated, and the aqueous phase was extracted with 3–10 mL portions of ether. The combined organic layers were dried ($MgSO_4$) and filtered, and the filtrate was evaporated to give a dark oil, which was purified by column chromatography on silica gel (hexane/benzene, 50:50) to give 0.94 g (85%) of **12** as a yellow oil. This compound appeared to be unstable; further purification was accomplished by preparative TLC on silica gel (6% ether in hexane). Anal. ($C_{22}H_{31}IO$) C: calcd, 58.92; found, 59.40. H: calcd, 9.21; found, 9.78.

**2-*cis*-$\Delta^8$-Heptadecenyl-5-(hydroxymethyl)tetrahydrofuran (13).** A mixture of 1.5 g (3.34 mmol) of **13**, 2.78 g (33.89 mmol) of anhydrous sodium acetate, and 30 mL of $Me_2SO$ was heated at 90 °C under a nitrogen atmosphere with stirring for 24 h. The mixture was then cooled and poured into 100 mL of ice–water. The organic product was extracted with 5–25 mL portions of ether. The combined ether layers were washed with water, dried ($MgSO_4$), and filtered, and the filtrate was evaporated to give the crude acetate. This was not purified but was dissolved in 10 mL of anhydrous ether and added dropwise to a stirred slurry of 0.3 g (7.89 mmol) of $LiAlH_4$ in 50 mL of ether under a nitrogen atmosphere. The mixture was stirred at room temperature and worked up by adding consecutively 0.3 mL of water, 0.3 mL of 15% aqueous NaOH, and 0.9 mL of water with constant stirring. After the solids were removed, the ether was dried ($MgSO_4$) and evaporated to give a yellow oil, which was purified by medium-pressure liquid chromatography on silica gel to afford 0.22 g (20% overall yield) of **13** as a yellow oil. Anal. ($C_{22}H_{42}O_2$) C, H.

**2-*cis*-$\Delta^8$-Heptadecenyl-5-(hydroxymethyl)tetrahydrofuran Monosodium Phosphate (3).** In a dry flask was placed a solution of 0.12 g (0.796 mmol) of $POCl_3$ in 25 mL of $CH_2Cl_2$ under a $N_2$ atmosphere. The flask was cooled in an ice bath, and a solution of 0.27 g (0.796 mmol) of **13**, 0.19 g (2.4 mmol) of pyridine, and 0.01 g (0.08 mmol) of 4-(dimethylamino)pyridine (DMAP) in 5 mL of $CH_2Cl_2$ was added dropwise over 10 min. The mixture was stirred at room temperature for 6 h, the solvent was removed, and the residue was dissolved in 30 mL of ether. The mixture was hydrolyzed by adding 1 mL of water and stirring for 30 min. The solvent was dried ($Na_2SO_4$) and filtered, and the filtrate was evaporated. The residue was placed on a 20-g silica gel column packed with $CHCl_3$. The column was eluted with 200 mL of $CHCl_3$ to remove nonplar impurities. The dihydrogen phosphate was then eluted with 400 mL of $CH_3OH$. The methanol was evaporated, the residue was dissolved in 20 mL of ether, the solution was filtered, and the filtrate was evaporated to give the dihydrogen phosphate as a waxy semisolid. This product was treated batchwise with Dowex-50 ($Na^+$) in 20 mL of water, filtered, lyophilized, and washed with acetone to give 0.068 g (19%) of the monosodium salt **3** as a yellow semisolid. Anal. ($C_{22}H_{42}O_5PNa$) C, H.

**Methyl 1-*cis*-$\Delta^8$-Heptadecenyl-2-oxocyclopentanecarboxylate (14).** To a solution of 5.15 g (36.2 mmol) of 2-carbomethoxycyclopentanone and 11.5 g (36.2 mmol) of *cis*-$\Delta^8$-1-bromoheptadecene[16] in 200 mL of acetone was added 5.43 g (36.2 mmol) of NaI and 12.51 g (90.5 mmol) of anhydrous $K_2CO_3$. The mixture was heated at reflux for 48 h, at which time TLC revealed almost complete disappearance of starting material. The mixture was cooled and filtered to remove the inorganic material. The filtrate was evaporated to give a yellow oil, which was purified by column chromatography on silica gel (6% ether in hexane) to give 10.99 g (80%) of **14** as an almost colorless oil. Anal. ($C_{24}H_{42}O_3$) C, H.

**Methyl 3-*cis*-$\Delta^8$-Heptadecyl-2-oxocyclopentanecarboxylate (15).** In a 250-mL, three-necked, round-bottom flask equipped with a magnetic stirrer, Vigreux column, and variable reflux–partial distillation apparatus was placed 10 mL of anhydrous methanol, 100 mL of dry xylene (distilled from Na), and 0.17 g (7.554 mmol) of Na. The mixture was heated with stirring at 60 °C until the sodium had been consumed. Then 2.86 g (7.554 mmol) of **14** was added, and the mixture was heated at reflux for 12 h. The methanol was removed by distillation and approximately half of the xylene was distilled out. The mixture was cooled to room temperature and diluted with 100 mL of 10% aqueous acetic acid. The organic product was extracted with 2–50 mL portions of ether, and the combined ether extracts were washed three times with 50-mL portions of water. The organic solution was dried ($MgSO_4$) and filtered, and the filtrate was evaporated

(16) H. J. Goller and D. S. Sgoutas, *Biochemistry*, **9**, 4801 (1970).

to give a dark oil, which was purified by medium-pressure liquid chromatography on silica gel (4% ethyl acetate in hexane) to give 1.39 g (48%) of **15** as a yellow oil. Anal. ($C_{24}H_{42}O_3$) H; C: calcd, 76.14; found, 75.47.

**1-(Hydroxymethyl)-3-*cis*-$\Delta^8$-heptadecenylcyclopentane (16).** To 1.85 g (4.89 mmol) of **15** in 50 mL of anhydrous methanol under a nitrogen atmosphere was added 0.28 g (7.4 mmol) of $NaBH_4$. This solution was stirred for 2 h at room temperature, at which time TLC revealed the disappearance of starting material. The alcohol was isolated by acidification with $CH_3COOH$, dilution with 50 mL of water, and extraction with 4–20 mL portions of ether. The ether extracts were dried ($MgSO_4$) and filtered, and the filtrate was evaporated to give the crude alcohol. This was then dissolved in 25 mL of $CH_2Cl_2$, and 0.74 g (7.33 mmol) of triethylamine (distilled from $CaH_2$) was added. The solution was cooled in an ice bath, 0.67 g (5.89 mmol) of freshly distilled $CH_3SO_2Cl$ was added, and the mixture was allowed to stir at ice-bath temperature for 30 min and then at room temperature for 2 h. The solution was washed with 50 mL of ice–water, followed by 50 mL of 10% ice-cold aqueous HCl, 50 mL of saturated aqueous $NaHCO_3$, and 50 mL of saturated NaCl solution. The organic phase was dried ($Na_2SO_2$) and filtered, and the filtrate was evaporated to give the crude methanesulfonate. This was dissolved in 25 mL of dry tetrahydrofuran in a dry $N_2$-filled flask. This was cooled in an ice bath, 30 mL (30 mmol) of a 1 M solution of $LiEt_3BH$ was added, and the mixture was stirred for 15 min at ice-bath temperature and then at room temperature for 2 h. Then 0.15 g (3.95 mmol) of $LiAlH_4$ was added, and the mixture was stirred for 1 h. Then 20 mL of water was cautiously added, followed by 100 mL of 10% aqueous HCl. The product was extracted with 3–50 mL portions of ether. The combined ether extracts were washed with saturated aqueous $NaHCO_3$, dried ($MgSO_4$), and filtered, and the filtrate was evaporated to give a dark oil, which was purified by column chromatography on alumina (benzene/hexane, 75:25) to give 0.56 g (34% overall yield) of **6** as a light yellow oil. Anal. ($C_{23}H_{44}O$) C, H.

**1-(Hydroxymethyl)-3-*cis*-$\Delta^8$-heptadecenylcyclopentane Monosodium Phosphate (4b).** To a solution of 0.21 g (1.34 mmol) of $POCl_3$ in 40 mL of $CH_2Cl_2$ maintained at 0 °C under $N_2$ in a dry flask was added dropwise over 10 min a solution of 0.03 g (0.26 mmol) of 4-(dimethylamino)pyridine, 0.11 g (1.39 mol) of pyridine, and 0.45 g (1.34 mmol) of **16** in 10 mL of $CH_2Cl_2$. The mixture was allowed to stir at room temperature for 16 h. The solvent was evaporated, and the residue was dissolved in 30 mL of ether. Then 1 mL of $H_2O$ was added, and this mixture was stirred for 30 min. The solvent was then dried ($NaSO_4$) and filtered, and the filtrate was evaporated to give a dark oil. The residue was placed on a 20-g silica gel column prepared with $CHCl_3$. The column was eluted with 200 mL of chloroform to remove nonpolar impurities. The dihydrogen phosphate ester was then eluted with 400 mL of methanol. The methanol was evaporated, the residue was taken up in 20 mL of ether, the solution was filtered, and the filtrate was evaporated. After batchwise treatment with Dowex-50 ($Na^+$) and lyophilization, the monosodium salt **4b** was obtained as a very hygroscopic solid. This substance was treated with 1 equiv of NaOH in 20 mL of methanol. The methanol was evaporated, the residue was dissolved in 10 mL of 9:1 $CHCl_3$/methanol saturated with water, the solution was filtered, and the solvent was evaporated. The residue was washed thoroughly with acetone and then dissolved in 20 mL of $H_2O$. The aqueous solution was lyophilized to give 0.12 g (20%) of the disodium salt as the monohydrate. Anal. ($C_{23}H_{43}O_4PNa_2$) C, H.

**4-(Hydroxymethyl)-2-pentadecyl-1,3-oxathiolane (17a).** Hexadecanal was prepared from commercially available 1-hexadecanol by oxidation with pyridinium chlorochromate as described for **7**. To 4.75 g (0.0198 mol) of hexadecanal and 15 mL of glacial acetic acid was added 1.63 mL (0.020 mol) of 3-mercapto-1,2-propanediol and 0.5 mL of boron trifluoride etherate. The mixture was stirred at room temperature for 2 h and then diluted with 100 mL of $H_2O$. The organic product was extracted with 3–100 mL portions of ether. The combined ether solutions were washed with 100 mL of $H_2O$, 100 mL 10% aqueous $Na_2CO_3$, and 100 mL of $H_2O$. The ether was dried ($MgSO_4$) and filtered, and the filtrate was evaporated to give the crude product, which was purified by medium-pressure liquid chromatography to give 0.77 g (11.1%) of **17a** as a white solid. Anal. ($C_{19}H_{38}O_2S$) C, H.

**2-*cis*-$\Delta^8$-Heptadecenyl-4-(hydroxymethyl)-1,3-oxathiolane (17b).** This compound was prepared from *cis*-$\Delta^9$-octadecenal in 10.3% yield by the same procedure as that described for the synthesis of compound **17a**. Anal. ($C_{21}H_{40}O_2S$) C, H.

**4-(Hydroxymethyl)-2-pentadecyl-1,3-oxathiolane Monosodium Phosphate (5a).** To a mixture of 0.255 g (1.67 mmol) of $POCl_3$ and 15 mL of anhydrous ether at 0 °C under a nitrogen atmosphere was added dropwise over 10 min a solution of 0.55 g (1.67 mmol) of **17a** and 0.13 g (1.67 mmol) of pyridine in 5 mL of ether. The mixture was stirred for 1.5 h at 0 °C. The mixture was then allowed to warm to room temperature. The solvent was evaporated, and the residue stirred with 50 mL of ice–water. The aqueous phase was extracted with 2–50 mL portions of ether. The combined ether layers were dried ($MgSO_4$) and filtered, and the filtrate was evaporated. The residue was then converted to the monosodium salt **5a** by batchwise treatment with Dowex-50 ($Na^+$). After lyophilization and washing with acetone, 0.32 g (40%) of the very hygroscopic **5a** was obtained: IR (KBr) 2940, 2860, 1460, 1250, 1000 $cm^{-1}$. Anal. ($C_{19}H_{38}O_5PS$ Na) H; C: calcd, 52.76; found, 51.81.

**2-*cis*-$\Delta^8$-Heptadecenyl-4-(hydroxymethyl)-1,3-oxathiolane Monosodium Phosphate (5b).** Compound **5b** was prepared by the same procedure as that described for compound **5a**. Treatment of **17b** (2.5 g, 7.01 mmol) as described previously yielded 0.7 g (22%) of **5b** as a very hygroscopic solid: IR (KBr) 2940, 2860, 1460, 1220, 1160, 1010, 760 $cm^{-1}$. Anal. ($C_{21}H_{40}O_5PSNa$) C; H: calcd, 8.73; found, 9.19.

**Pharmacological Testing.** A portion of guinea pig terminal ileum was rapidly removed from a sacrificed animal and maintained in modified Tyrodes's solution at 37 °C. Tissues were oxygenated by bubbling 95% $O_2$/5% $CO_2$ through the buffer. For testing, the tissue was suspended in a 10-mL organ bath under 1-g tension, and contraction magnitude was measured with a Beckman dynograph recorder. A control response curve was measured for acetylcholine; following tissue recovery a response curve for the test compound was measured, and the ratio of doses required to elicit 50% maximal response was determined. All results are the means of three experiments.

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