versed-phase preparative HPLC as described,[19] using a 2.5 × 100 cm column (Altex) packed with 25–40 μm Lichroprep RP-18 (E. Merck). The eluent contained various proportions of CH_3CN and H_2O, depending on the hydrophobicity of the compound (k'), but in each case the eluent was 0.03 M in NH_4OAc (pH 7).

The fractions containing the product (analytical HPLC) were pooled and concentrated to dryness. The bulk of the NH_4OAc was sublimed under vacuum from the flask into a Kjeldahl head by use of a 40 °C water bath. The residual traces were removed by lyophilization three times from H_2O to yield the pure product as a fluffy white powder.

Rat Antiovulatory Assay.[49] Adult female Simonsen Albino rats (Sprague-Dawley strain; >180 g) were acclimatized to laboratory conditions (14:10, light:dark with lights on at 5 a.m.) for at least 10 days. Daily vaginal lavages were taken from each rat between 7:30 and 9:00 a.m. for at least 12 days. Cytology of vaginal lavages was examined microscopically to determine the stage of the estrous cycle. Rats with at least two normal 4-day cycles preceding the test cycle were selected. The rats were injected subcutaneously with a solution of the analogue in vehicle (50% propylene glycol/0.9% saline or Mazola corn oil).

On the morning of expected estrus, the rats were sacrificed, and the oviducts were removed and examined under a dissecting microscope for the presence of freshly ovulated eggs. The eggs were teased out of the oviducts and counted. The percent of females ovulating was plotted against the log-dose to calculate the ED_{50} for antiovulatory activity. Usually three or more dose groups (data from a total of 10 animals per dose group) were used to determine the ED_{50}, but occasionally two dose groups were found to be sufficient.

(49) Corbin, A.; Beattie, C. W. *Endocr. Res. Commun.* **1975**, *2*, 1.

Acknowledgment. We gratefully acknowledge the technical assistance of A. Worden, B. Roberts, and D. Donahue in conducting the bioassays. We are indebted to J. Fazzari and S. Williams for HPLC and amino acid analyses. The encouragement and advice of Dr. John Moffatt is gratefully acknowledged.

Registry No. 1, 90531-06-9; 2, 90552-82-2; 3, 90531-07-0; 4, 75851-13-7; 5, 90531-08-1; 6, 90531-09-2; 7, 82778-58-3; 8, 85466-65-5; 9, 90531-10-5; 10, 82778-57-2; 11, 90531-11-6; 12, 90531-12-7; 13, 90531-13-8; 14, 90531-14-9; 15, 86855-16-5; 16, 81608-49-3; 17, 90531-15-0; 18, 90531-16-1; 19, 90531-17-2.

Analogues of Platelet Activating Factor (PAF). 1. Some Modifications of the Alkoxy Chain

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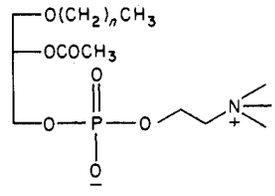
Analogues of platelet activating factor (PAF) in which the ether oxygen has been removed (**6**) and in which the alkoxy chain at C_1 has been replaced with a *o*-, *p*-, or *m*-alkylphenoxy group (**30**, **31**, and **35**, respectively) have been prepared. Compound **6** shows reduced platelet aggregation and hypotensive activity in comparison with C_{16} and C_{18} PAF. The results obtained for compounds **30**, **31**, and **35** indicate that the hypotensive and platelet aggregating responses are sensitive to structural modification of the ether chain. The ortho analogue **30** shows no platelet aggregating activity and only a weak hypotensive response. The para analogue **31** exhibits a moderate decrease in activity in both assays. The meta analogue **35** is the most active of the three.

1-*O*-Alkyl-2-*O*-acetyl-*sn*-glyceryl-3-phosphorylcholine in which the alkyl component is comprised largely of the C_{18} and C_{16} homologues (**1a** and **1b**) has been identified with platelet activating factor (PAF) by two independent research groups.[1] Coincidently, the same substance was



1a, $n = 17$
b, $n = 15$

prepared by Muirhead and colleagues and was found to have similar biological properties to those of a phospholipid isolated from the renal medulla and designated as the antihypertensive polar renomedullary lipid (APRL).[2] The identification of this structure has stimulated a vast amount of research on the biological properties of this substance in recent years. It is increasingly becoming apparent that PAF is not only one of the most potent aggregators of platelets known but is also one of the important mediators of anaphylaxis and inflammation promoting the activation of neutrophils, basophils, and other inflammatory cells.[4] In addition, Muirhead and coworkers have shown that this substance is exceedingly potent in its ability to lower blood pressure,[2] while our laboratory has presented evidence that this hypotensive effect is the result of a nonspecific vasodilation.[3] It is the latter property of PAF that has stimulated our interest.

We are proposing the hypothesis that the multiple biological activities of PAF need not be mediated by a common receptor and that the hypotensive effect is not a consequence of mediator cell (e.g., platelet) activation and release. If this indeed be the case, it should be possible to identify a structure in which these two properties have been separated sufficiently to allow the development of a therapeutically useful hypotensive agent.

(1) (a) Benveniste, J.; Tence, M.; Varenne, P.; Bidault, J.; Boullet, C.; Polonsky, J. *C. R. Hebd. Seances Acad. Sci., Ser. D* **1979**, *289*, 1037. (b) Polonsky, J.; Tence, M.; Varenne, P.; Das, B. C.; Lunel, J.; Benveniste, J. *Proc. Natl. Acad. Sci. U.S.A.* *77*, 7019. (c) Demopoulos, C. A.; Pinckard, R. N.; Hanahan, D. J. *J. Biol. Chem.* **1979**, *254*, 9355. (d) Hanahan, D. J.; Demopoulos, C. A.; Liehr, J.; Pinckard, R. N. *J. Biol. Chem.* **1980**, *255*, 5514.

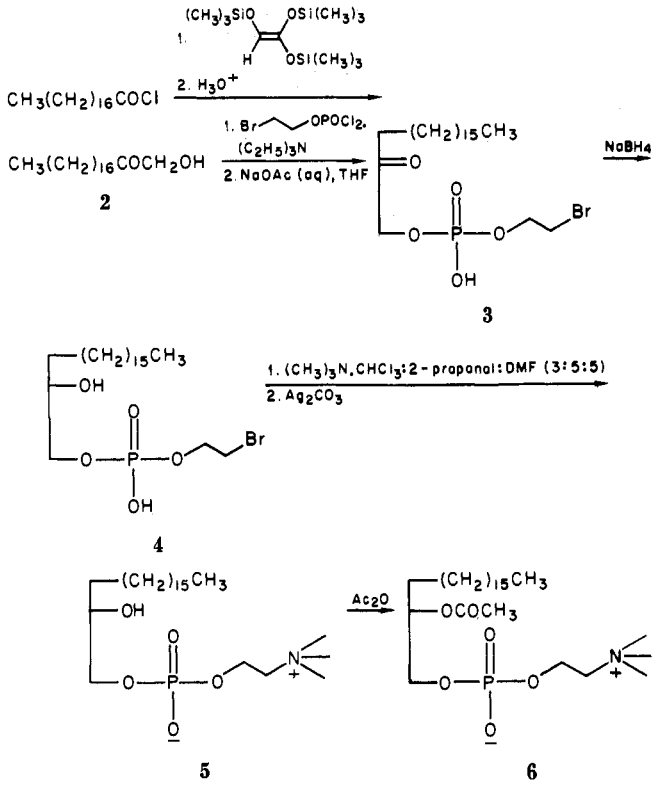
(2) Blank, M. L.; Snyder, F.; Byers, L. W.; Brooks, B.; Muirhead, E. E. *Biochem. Biophys. Res. Commun.* **1979**, *90*, 1194.

(3) Lai, F. M.; Shepherd, C. A.; Cervoni, P.; Wissner, A. *Life Sci.* **1983**, 1159.

(4) For recent reviews of the biological properties of PAF, see: (a) Snyder, F. *Ann. Rep. Med. Chem.* **1982**, *17*, 243. (b) Pinckard, R. N.; McManus, L. M.; Hanahan, D. J. *Adv. Inflammation Res.* **1982**, *4*, 147. (c) Vargaftig, B. B.; Chignard, M.; Benveniste, J.; Lefort, J.; Wal, F. *Ann. N.Y. Acad. Sci.* **1981**, *370*, 119.

0022-2623/84/1827-1174$01.50/0

Scheme I



We now describe the synthesis and biological properties of some of the PAF analogues in which we have incorporated modifications of the alkoxy group at C_1. Previous work in this area has been limited to variations of chain length.[5] Specifically, we have addressed two questions in this study: (1) To what extent is the ether oxygen necessary for hypotensive and platelet aggregating activities? (2) If these biological activities are receptor mediated, to what extent are they sensitive to the spacial arrangement of atoms in the alkoxy chain?

Chemistry. The preparation of a deoxy-PAF analogue **6** in which the ether oxygen of the alkoxy chain has been removed is depicted in Scheme I. Heating *n*-octadecanoyl chloride with 2 equiv of tris[(trimethylsilyl)oxy]ethylene[6] furnished after hydrolysis the hydroxymethyl ketone **2**. The phosphate group was introduced by the method of Hirt[7] to give the ketone **3**, which was reduced with sodium borohydride to the alcohol **4**. The quaternary nitrogen group was introduced by heating **4** with an excess of 40% trimethylamine in a mixture of chloroform:2-propanol: dimethylformamide (3:5:5). Treatment of the reaction product with silver carbonate (to remove the trimethylamine hydrobromide) and chromatography on Florisil furnished compound **5**. The desired deoxy analogue **6** was then prepared by acylation with acetic anhydride. The structure of **6** was confirmed by analytical and spectral data.

Our approach to the problem concerning the sensitivity of the hypotensive and platelet aggregating responses to the spacial orientation of the alkoxy chain was to prepare a series of racemic *o*-, *p*-, and *m*-alkyl-substituted phenoxy analogues **30**, **31**, and **35**, respectively (Scheme II).[8] The

Table I. Hypotension and Platelet Aggregation Activities for PAF Analogues

compd	MABP[a,b]	platelet EC_{50}[c,h]	max platelet response[d]	ratio[e]
1a	3.17 (1.03, 10.22)	1.6×10^{-8} ($n = 12$)	79 (1×10^{-6})	1.98
1b	1.25 (0.30, 4.98)	2.22×10^{-8} ($n = 5$)	71 (1×10^{-6})	0.56
6	357.3 (89.2, 1113.4)	3.1×10^{-6} ($n = 1$)	31 (1×10^{-5})[i]	1.15
30	*f*	$>1 \times 10^{-5}$ ($n = 1$)	*g*	
31	12.56 (1.85, 78.93)	5.3×10^{-8} ($n = 1$)	80 (1×10^{-6})	2.37
35	0.43 (0.10, 1.83)	4.4×10^{-8} ($n = 1$)	70 (1×10^{-6})	0.10

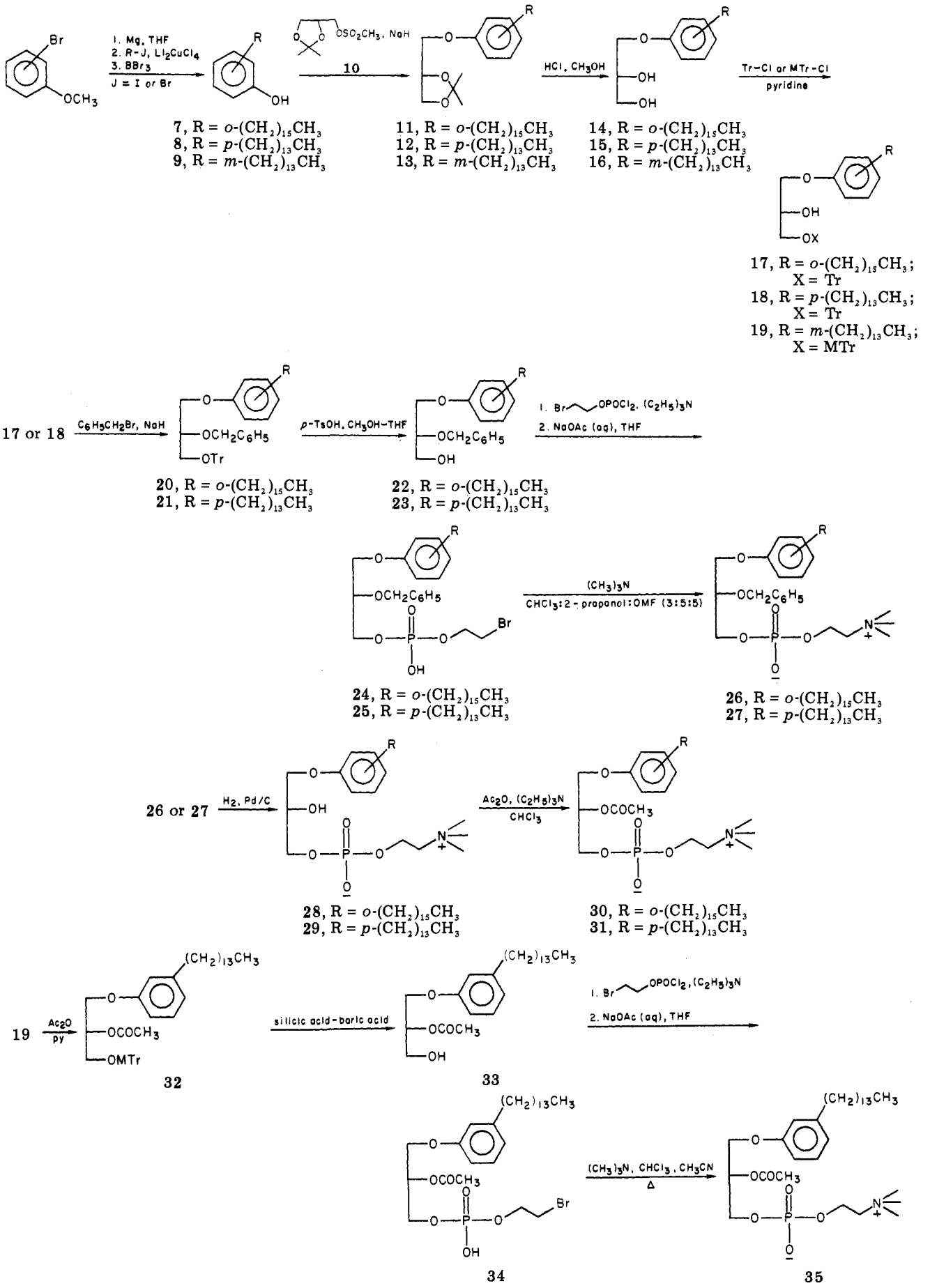
[a] Dose (μg/kg, iv) needed to decrease mean arterial blood pressure (MABP) 50 mmHg. [b] Values in parentheses are 95% confidence limits. [c] Molar concentration required to cause 50% of maximum aggregation. [d] Maximum aggregation units at specified molar concentration. [e] (MABP/platelet EC_{50}) $\times 10^{-8}$. [f] MABP decreased only 10 mmHg at 1000 μg/kg. [g] No response. [h] Values in parentheses are the number of experiments in which a dose–response curve was determined from two to six replicates per dose level. [i] This maximum response may indicate a partial agonist.

copper(I)-catalyzed[9] coupling of the appropriate Grignard reagent derived from the three possible monobromoanisoles with an alkyl bromide or iodide in refluxing tetrahydrofuran followed by demethylation with boron tribromide in methylene chloride at 0 °C provided the required alkyl phenols **7–9**. Alkylation of the mesylate **10** with phenols **7–9** using sodium hydride in dimethylformamide gave **11–13**. Acid hydrolysis of ketals **11–13** then furnished the diols **14–16**. While diols **14** and **15** were converted to the respective primary trityl derivatives **17** and **18** using trityl chloride (Tr-Cl) in pyridine, diol **16** was converted to the *p*-methoxytrityl derivative **19** by using *p*-methoxytrityl chloride (Mtr-Cl) in pyridine. The trityl-protected compounds **17** and **18** were benzylated, giving **20** and **21**, respectively. The trityl groups were then removed by using *p*-toluenesulfonic acid in a mixture of methanol and tetrahydrofuran. The resulting alcohols **22** and **23** were then converted to the phosphates **24** and **25**, respectively, as described above. The quaternary ammonium group was then introduced, furnishing **26** and **27** both of which were debenzylated by hydrogenolysis with a palladium catalyst to give compounds **28** and **29**, respectively. Finally, acetylation with acetic anhydride–triethylamine in refluxing chloroform gave the ortho and para analogues **30** and **31**.

A somewhat shorter route was subsequently developed to convert **10** to the meta-substituted analogue **35**. Acylation of **19** with acetic anhydride in pyridine gave the acetate **32**. Removal of the *p*-methoxytrityl group without concurrent acyl migration was accomplished by using the silicic acid–boric acid chromatographic procedure of Buchnea[10] to give **33**. The phosphate moiety was introduced as before, giving **34**. Finally, the quaternary nitrogen

(5) (a) Hanahan, D. J.; Munder, P. G.; Satouchi, K.; McManus, L.; Pinckard, R. N. *Biochem. Biophys. Res. Commun.* **1981**, *99*, 183. (b) Tence, M.; Coeffies, E.; Heymans, F.; Polonsky, J.; Godfroid, J.; Benveniste, J. *Biochimie* **1981**, *63*, 723.
(6) Wissner, A. *J. Org. Chem.* **1979**, *44*, 4617.
(7) Hirt, R.; Berchtold, R. *Pharm. Acta Helv.* **1958**, *33*, 349.
(8) (a) Since it has been established that the unnatural (*S*) enantiomer of PAF is devoid of both hypotensive and platelet activating properties (Blank, M. L.; Cress, E. A.; Lee, T.-C.; Malone, B.; Surles, J. R.; Piatadosi, C.; Hajdu, J.; Snyder, F. *Res. Commun. Chem. Pathol. Pharmacol.* **1982**, *38*, 3), we have decided in cases that would result in a shorter synthesis to prepare the racemic analogues. (b) We have elected to prepare the ortho analogue **30** with a longer alkyl chain in order to compensate for the decreased contribution to the overall length of the ether chain by the aromatic ring in this compound.
(9) Tomura, M.; Kochi, J. *Synthesis* **1971**, 303.
(10) Buchnea, D. *Lipids* **1974**, *9*, 55.

Scheme II


1. Mg, THF
2. R-J, Li2CuCl4
3. BBr3
J = I or Br
OSO2CH3, NaH
10
HCl, CH3OH
Tr-Cl or MTr-Cl
pyridine
7, R = o-(CH2)15CH3
8, R = p-(CH2)13CH3
9, R = m-(CH2)13CH3
11, R = o-(CH2)15CH3
12, R = p-(CH2)13CH3
13, R = m-(CH2)13CH3
14, R = o-(CH2)15CH3
15, R = p-(CH2)13CH3
16, R = m-(CH2)13CH3
17, R = o-(CH2)15CH3; X = Tr
18, R = p-(CH2)13CH3; X = Tr
19, R = m-(CH2)13CH3; X = MTr
17 or 18
C6H5CH2Br, NaH
p-TsOH, CH3OH-THF
1. Br-CH2CH2-OPOCl2, (C2H5)3N
2. NaOAc (aq), THF
20, R = o-(CH2)15CH3
21, R = p-(CH2)13CH3
22, R = o-(CH2)15CH3
23, R = p-(CH2)13CH3
(CH3)3N
CHCl3:2-propanol:DMF (3:5:5)
24, R = o-(CH2)15CH3
25, R = p-(CH2)13CH3
26, R = o-(CH2)15CH3
27, R = p-(CH2)13CH3
26 or 27
H2, Pd/C
Ac2O, (C2H5)3N
CHCl3
28, R = o-(CH2)15CH3
29, R = p-(CH2)13CH3
30, R = o-(CH2)15CH3
31, R = p-(CH2)13CH3
19
Ac2O
py
silicic acid-boric acid
1. Br-CH2CH2-OPOCl2, (C2H5)3N
2. NaOAc (aq), THF
32
33
(CH3)3N, CHCl3, CH3CN
Δ
34
35

functionality was added to furnish **35**, in this case under nonaqueous conditions in order to avoid hydrolysis of the acetate group, by refluxing **34** in a mixture of chloroform–acetonitrile containing a large excess of trimethylamine.

Biology. Table I presents the hypotension and platelet aggregation data we obtained for compounds **6**, **30**, **31**, and **35** as well as that for racemic C_{18} and C_{16}-PAF (**1a** and **1b**), our standard compounds.

Rats were chosen for blood pressure studies since it has been determined that rat platelets do not respond well to PAF[4c,11] and that the hypotensive effect of PAF is not mediated by platelets in this species.[4c] In contrast, rabbits were chosen to obtain platelets for in vitro platelet aggregation studies since the platelets of this species are known to be very sensitive to PAF.[4c,12] The hypotensive data were obtained in the spontaneously hypertensive rat and are expressed as the intravenous dose needed to reduce the mean arterial blood pressure (MABP) 50 mmHg as extrapolated from the least-squares regression line.

Platelet aggregation responses were obtained for rabbit platelet rich plasma. The data are expressed as the molar concentration that is needed of a given compound to obtain 50% of its maximum aggregation response (EC_{50}). Also presented in the table is the maximum aggregation response of a particular compound obtained at the indicated molar concentration; this value can be used to distinguish partial and full agonists.

The last column in the table gives the ratio of the hypotension and platelet aggregation values, which can be used as a measure of the degree of separation of biological activities for a particular compound relative to the standard compounds. An analogue that has a ratio smaller than that observed for the standard compounds would indicate increased selectivity in favor of the hypotensive effect, while an analogue with a larger ratio would indicate selectivity in favor of platelet aggregation.

Results and Discussion

The hypotensive and platelet aggregation data obtained for racemic C_{18}- and C_{16}-PAF (**1a** and **1b**) and the analogues are presented in Table I. Both standard compounds **1a** and **1b** show comparable activities in either assay.

The results obtained for the deoxy analogue **6** indicate that removal of the ether oxygen linkage at C_1 produces a significant decrease in activity both with respect to its ability to reduce blood pressure and its ability to stimulate platelet aggregation. Moreover, the hypotension to platelet aggregation ratio of 1.15 for this compound falls inside the range (0.56–1.98) obtained for our standard compounds **1a** and **1b** and suggests that this modification of the PAF structure has resulted in no separation of the two biological properties.

The results obtained for the *o*-, *p*-, *m*-alkyl-substituted phenoxy analogues **30**, **31**, and **35**, respectively, indicate that the hypotensive and platelet aggregating activities are sensitive to structural modification of the alkoxy chain. The ortho-substituted analogue **30** shows no platelet aggregation response at concentrations up to 1×10^{-5} M. In addition, the hypotensive potency of this compound is extremely weak when compared to that of the standard compounds. While a dose of 1000 μg/kg of **30** results in a 10-mm decrease of the mean arterial blood pressure (MABP), a dose of 0.41 μg/kg of C_{18}-PAF (**1a**) results in the same decrease in MABP. In contrast, the para-substituted analogue **31** shows only a moderate decrease in activity; when compared to **1a**, there appears to be a 4-fold decrease in potency as a hypotensive agent and only about a 3-fold decrease in potency as a platelet activating agent. The meta-substituted compound **35** is the most active of the three in both assays; while it appears to be somewhat more potent than the standard compounds with respect to the hypotensive effect, it appears to be slightly (2–3-fold) less potent with respect to platelet aggregation. The ratios of the hypotension and platelet aggregation values for **31** and **35** are 2.37 and 0.10, respectively. Since these values fall outside opposite ends of the range (0.56–1.98) exhibited by the standards **1a** and **1b**, there is the suggestion of a slight change in selectivity with variation of structure although this may not be statistically significant.

This marked difference in biological activities associated with variation of the nature of the alkoxy chain is consistent with the suggestion that both the hypotensive and platelet aggregating responses of PAF are receptor-mediated processes.[13] Since the possibility exists that the receptor associated with hypotension and that associated with cell activation may have different steric requirements, we are currently involved in the preparation of additional analogues containing modifications of the alkoxy chain and other portions of the molecule in an attempt to prepare a compound showing increased selectivity in favor of the hypotensive response.

Experimental Section

General Methods. Unless otherwise stated, the following are implied. Melting points were determined on a Mel-Temp capillary melting point apparatus and are uncorrected. The nuclear magnetic resonance (NMR) spectra were recorded either on a Varian EM 390 spectrometer or Varian FT-80 spectrometer, and chemical shifts in parts per million (ppm) are reported with tetramethylsilane (Me_4Si) or chloroform as internal references. Infrared spectra (IR) were recorded on a Nicolet FT 7000 spectrophotometer. Mass spectra were determined on a Finnegan-MAT Model CH 7 mass spectrometer. The field desorption (FD), (FD-*p*-TSA), and fast atom bombardment (FAB) mass spectra were obtained by using a Kratos MS 50 mass spectrometer. Where analyses are indicated only by symbols of the elements, analytical results obtained for these elements were within ±0.4% of the theoretical value.

Blood Pressure Studies. Male spontaneously hypertensive rats (Taconic Farms) between the ages of 18 and 24 weeks were utilized in these experiments. Under ether anesthesia, the left femoral artery and vein were cannulated with saline-filled catheters composed of a $1^3/_4$-in. length of polyethylene (PE) 10 tubing heat-sealed to a 6-in. length of PE 20. All catheters were tunneled subcutaneously and exteriorized at the back of the neck. All incisions were closed with wound clips, and the rats were returned to single cages.

At least 16 h following surgery, the rats were placed in Broome style restraining cages for the duration of the experiment. The femoral artery catheter was connected to a Statham P23 ID pressure transducer with PE 100 tubing and a stainless steel stepdown connector. The arterial pulse pressure was monitored on a Grass Model 7 physiological recorder. The output from each channel of the recorder was processed through a MINC-23 computer (Digital Equipment Corp.), which analyzed each waveform at the rate of 100 samples/s and averaged the collected data over 15-s periods. Values derived from the arterial pulse pressure included mean, systolic, and diastolic blood pressure and heart rate. All values were updated on a video display every 15 s and stored to disk at the same frequency. An additional length of

(11) Cargill, D. I.; Cohen, D. S.; Von Valen, R. G.; Klimek, J. J.; Leven, R. P. *Thromb. Haemostasis* **1983**, *49*, 204.

(12) (a) Benveniste, J.; Henson, P. M.; Cochrane, C. G. *J. Exp. Med.* **1972**, *136*, 1356. (b) Comussi, G.; Tetta, C.; Deregibus, M. C.; Bussolino, F.; Segoloni, G.; Vercellone, A. *J. Immunol.* **1982**, *128*, 86.

(13) (a) Valone, F. H.; Coles, E.; Reinhold, V. R.; Goetzl, E. J. *J. Immunol.* **1982**, *129*, 1637. (b) Valone, F. H.; Gotzl, E. J. *Immunology* **1983**, 141. (c) Hwang, S-B.; Lee, C-S. C.; Cheak, M. J.; Shen, T. Y. *Biochemistry*, **1983**, *22*, 4756.

PE 20 tubing terminated at one end with a 1-mL syringe and needle was attached to the femoral vein catheter with use of a stainless steel adaptor.

Stock solutions were prepared by dissolving the test compounds in saline or 10–25% ethanol in saline. Aliquots of the stock solutions were appropriately diluted such that the volume of all doses was 1 mL/kg. Each dose was administered by rapid, intravenous bolus injection and followed by a 0.2-mL flush.

The computer files were read at 1-min intervals for 5 min preceding dosing and for 15 min after administration of the compounds. Depending on the dose, some of the responses were followed from 15 to 90 min at 5–10-min intervals.

At individual doses, the maximum change in mean arterial blood pressure was obtained for each rat, and the responses for that dose were averaged. The equation for the least-squares regression line (response = slope × dose + intercept) was determined and the best fit line plotted using the averaged responses for each dose. The dose required to reduce mean arterial blood pressure by 50 mmHg was determined by solving the equation of the regression line for dose and entering −50 for the response value.

Platelet Aggregation Studies. Platelet aggregation was measured in the usual manner. Briefly, 90–120 mL of blood was collected by cardiac puncture from unanesthetized male New Zealand white rabbits (Whaley's Summit View Farms, Belvedere, NJ) with use of 3.2% sodium citrate anticoagulant (1 pt of citrate/10 pt of blood). The blood was centrifuged at low speed (800 rpm × 15 min) at room temperature to recover the platelet-rich plasma (PRP). Dilutions (1:3000) of PRP in Isoton diluent were made and platelet counts were determined on a Coulter Thrombocounter, which was standardized with platelet reference standards (Interscience Platelet Control, Portland, OR). Sample platelet counts ranged 350 000–600 000/μL of PRP and could be adjusted to a specific value by the addition of platelet-poor plasma (PPP), which was prepared by centrifuging PRP at 3000 rpm × 30 min.

Incubation mixtures consisted of 450 μL of PRP and 50-μL aliquots of stock solutions of **1a**, **1b**, or PAF analogues. Samples of PRP in a cuvette were stabilized for 1–2 min at 37 °C in the aggregometer to achieve a stable base line. A stock solution of $1–3 \times 10^{-3}$ M PAF compound was made in 10% EtOH/90% H_2O. Dilutions were made with saline to give working stock solutions. Fifty microliters of the working stocks were added to the cuvette to give final concentrations ranging from 1×10^{-9} to 1×10^{-4} M, and the aggregation response was monitored for a 3–5-min period. The final EtOH concentration was kept below 0.5% to avoid inhibitory effects of the diluent. Each compound was tested in two to six trials per dose level.

The recording equipment consisted of two single channel Chronolog aggregometers connected to a dual-channel 10-mv full scale deflection Omniscribe chart recorder (Houston Instruments). The chart recorder was calibrated daily with use of a suspension of Bio-Rad latex beads (S-X 12 400 mesh) which had a density slightly greater than the PRP sample for 0% light transmission and clear water for 100% light transmission. These limits defined a full-scale deflection. Aggregation tracings were analyzed by a digitizing method[14] and x, y coordinate data was automatically stored on files in a computer. A basic program then computed parameters of interest such as the base line and the magnitude of aggregation for each curve.

The mean dose–response curve for each compound was determined and EC_{50} values (the dose required to give a response that is 50% of the maximum attainable with that compound) was determined.

1-Hydroxy-2-nonadecanone (2). A mixture of *n*-octadecanoic acid chloride (96.5 g, 0.32 mol) and tris[(trimethylsilyl)oxy]-ethylene[6] (205.06 g, 0.7 mol) was heated at 110–115 °C for 2 h. Another 65 g of tris[(trimethylsilyl)oxy]ethylene was added and heating was continued for one more hour. The solution was poured slowly into a mixture of 300 mL of tetrahydrofuran, 300 mL of water, and 50 mL of concentrated hydrochloric acid with vigorous stirring (exothermic). The mixture was stirred at reflux for 15 min and then diluted with ethyl acetate. The water layer was separated from the hot mixture. The ethyl acetate layer was kept hot, saturated aqueous sodium bicarbonate was added, and the mixture was stirred for 10 min. The organic layer was separated, dried, and evaporated and crude product was recrystallized from methanol, giving 81.3 g (85%) of the desired title compound **2** as a white solid: mp 78–80 °C; IR (KBr) 1720, 3250 cm^{-1}; NMR ($CDCl_3$) δ 0.9 (t, 3 H), 1.1–1.8 (m, 30 H), 2.4 (t, 2 H), 3.02 (brs, 1 H), 4.2 (s, 2 H); MS, *m/e* 298 (M^+). Anal. ($C_{19}H_{38}O_2$) C, H.

2-Bromoethyl 2-Oxononadecyl Phosphate (3). To a solution of 1-hydroxy-2-nonadecanone (**2**) (65 g, 0.22 mol) in 1 L of warm carbon tetrachloride was added 2-bromoethyl phosphorodichlorodate (68.46 g, 0.28 mol), followed by triethylamine (28.64 g, 0.28 mol). After 2 h, the mixture was filtered and the solvent removed. The residue was stirred overnight in a mixture of 1 L of 0.5 M sodium acetate and 1 L of tetrahydrofuran. Most of the tetrahydrofuran was removed, and ether and concentrated hydrochloric acid were added. The mixture was extracted with ether. The ether extract was dried and evaporated, and the residue was recrystallized twice with methanol, giving 65 g of crude product. A 50-g portion of this crude product was chromatographed on a Florisil column, eluting first with chloroform and then with chloroform–methanol (1:1). The isolated product was then recrystallized twice from methanol–2-propanol, giving 41.0 g (50%) of the desired title compound **3** as a white solid: mp 94–97 °C; IR (KBr) 1725 cm^{-1}; NMR ($CDCl_3/CD_3OD$) δ 0.9 (t, 3 H), 1.1–1.75 (m, 30 H), 2.43 (t, 2 H), 3.3 (brs, 1 H), 3.5 (t, 2 H), 4.1 (s, 2 H), 4.4–4.8 (m, 2 H); MS *m/e* 405 (M^+ − Br). Anal. ($C_{21}H_{42}O_3P$-$Br \cdot H_2O$) C, P; H: calcd, 8.81; found, 8.08; Br: calcd, 15.87; found, 15.20.

2-Bromoethyl 2-Hydroxynonadecyl Phosphate (4). 2-Bromoethyl 2-oxononadecyl phosphate (**3**) (34 g, 0.07 mol) was heated in 650 mL of 2-propanol until all solid was dissolved. This solution was partially cooled in a water bath, and while the solution was still warm, sodium borohydride (3.31 g, 0.087 mol) was added in portions, with stirring, over a period of 10 min. The reaction mixture was stirred for 1.5 h, most of the 2-propanol was removed, and the residue was diluted with dilute hydrochloric acid. The mixture was extracted with ether. The ether extract was dried and evaporated, and the residue was recrystallized from hexanes with cooling, giving 32.8 g (96%) of the desired title compound **4**: mp 68–70 °C; IR (KBr) 3460 cm^{-1}; NMR ($CDCl_3$), CD_3OD) δ 0.9 (t, 3 HO), 1.1–1.7 (m, 32 H), 3.5 (t, 2 H), 3.75–4.75 (m, 7 H); MS (CI), *m/e* 485.9, 487.2. Compound **3** was used directly in the next step without further purification.

2-[[Hydroxy[(2-hydroxynonadecyl)oxy]phosphinyl]-oxy]-*N,N,N*-trimethylethanaminium, Hydroxide, Inner Salt (5). A mixture of 2-bromoethyl 2-hydroxynonadecyl phosphate (**4**) (20 g, 0.041 mol), 600 mL of a mixture of chloroform, 2-propanol, and dimethylformamide (3:5:5), and 375 mL of 40% aqueous trimethylamine was stirred at 55 °C for 5 h. The solvents were removed (with the addition of toluene to prevent foaming). The residue was dissolved in a mixture of 200 mL of ethanol and 100 mL of water, and silver carbonate (5.66 g, 0.02 mol) was added and the mixture stirred overnight. The mixture was then filtered through Celite, the solvents were removed, and the residue was dissolved in a small amount of ethanol and precipitated with ether, giving 17.2 g (90%) of desired title compound **5** as a white solid: IR (KBr) 3240 cm^{-1}; NMR ($CDCl_3/CD_3OD$) δ 0.9 (t, 3 H, 1.1–1.7 (m, 32 H), 3.2 (s, 9 H), 3.5–4.0 (m, 4 H), 4.25 (m, 4 H); MS, *m/e* 465 (M^+). Anal. ($C_{24}H_{52}O_5PN \cdot H_2O$) C, P; H: calcd, 11.25; found, 10.65; N: calcd, 2.90; found 2.21.

7-Heptadecyl-4-hydroxy-*N,N,N*-trimethyl-9-oxo-3,5,8-trioxa-4-phosphadecan-1-aminium 4-Oxide, Hydroxide, Inner Salt (6). A mixture of 2-[[hydroxy[(2-hydroxynonadecyl)oxy]-phosphinyl]oxy]-*N,N,N*-trimethylethanaminium, hydroxide, inner salt (**5**) (7 g, 0.015 mol), sodium acetate (0.62 g, 0.0075 mol), and 35 mL of acetic anhydride was refluxed for 15 min. The excess acetic anhydride was removed by distillation at reduced pressure. The residue was dissolved in chloroform and filtered through Celite, and the solvent was removed. The residue was chromatographed on silica gel, eluting first with chloroform–methanol (7:3) and then with chloroform–methanol (1:1), and finally the product was eluted with chloroform–methanol (3:7). The solvents were removed at room temperature, and the residue was stirred with moist ether overnight, giving 4.5 g (59%) of the desired product **6** as a hygroscopic powder: mp 204–225 °C dec; IR (KBr)

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1735 cm^{-1}; NMR ($CDCl_3$, CD_3OD) δ 0.9 (t, 3 H, 1.1–1.8 (m, 32 H), 2.05 (s, 3 H), 3.2 (s, 9 H), 3.5–3.75 (m, 2 H), 3.88 (t, 2 H), 4.0–4.4 (m, 2 H), 4.98 (m, 1 H); MS (FD), *m/e* 508 (M + H). Anal. ($C_{26}H_{54}NO_6P \cdot {}^1/_2H_2O$) C, H, N, P.

2-*n*-Hexadecylphenol (7). Magnesium (4.86 g, 0.2 mol) was placed in a 250-mL, two-necked, round-bottom flask. The flask was flamed several times and *o*-bromoanisole (37.4 g, 0.2 mol) in 200 mL of tetrahydrofuran was added in several portions. As soon as the vigorous reaction stopped, it was refluxed for 2 h and added dropwise to a boiling solution of hexadecyl iodide (35.3 g, 0.1 mol) in tetrahyrofuran. Dilithium tetrachlorocopper(II) solution (10 mL, 0.1 M in THF) was added. The mixture was refluxed for 2 h and stirred at room temperature for 16 h. The solution was then quenched with dilute hydrochloric acid and extracted with ether. The organic layer was washed with ammonium chloride solution and saturated sodium bicarbonate solution and dried over anhydrous magnesium sulfate. Solvents were evaporated and the side products were removed by kugelrohr distillation at ca. 110 °C (1 mm). The yield of crude product was 34 g. Recrystallization from ether–methanol gave 31 g (93%) of 1-hexadecyl-2-methoxybenzene: mp 38 °C; IR (KBr) 1600 cm^{-1}; NMR ($CDCl_3$) δ 0.9 (t, 3 H), 1.1–1.9 (m, 28 H), 2.65 (t, 2 H), 3.85 (s, 3 H), 6.75–7.4 (m, 4 H); MS, *m/e* 332 (M^+). Anal. ($C_{23}H_{38}O_1$) C, H.

Boron tribromide (55.12 g, 0.22 mol) was added dropwise, under argon, to a solution of the 1-hexadecyl-2-methoxybenzene (72.96 g, 0.22 mol) in 500 mL of methylene chloride at 0 °C. The reaction was stirred at room temperature for 3 h and was quenched slowly with water. Ether was added and the organic layer was separated. The organic layer was stirred with a minimum amount of 10% aqueous sodium hydroxide for 1 h, acidified with dilute hydrochloric acid, and extracted with ether. The combined organic layers were washed with aqueous sodium bicarbonate solution and dried, and solvents were evaporated, giving 67 g (96%) of 2-hexadecylphenol (7): mp 54–55 °C; IR (KBr) 3350 cm^{-1}; NMR $CDCl_3$) δ 0.95 (t, 3 H), 1.2–1.8 (m, 28 H), 2.65 (t, 2 H), 4.7 (brs, 1 H), 6.65–7.5 (m, 4 H); MS, *m/e* 318 (M^+). Anal. ($C_{22}H_{38}O_1$) C, H.

4-Tetradecylphenol (8). The procedure used to prepare 4-methoxy-1-tetradecylbenzene was the same as the employed in the preparation of 1-hexadecyl-2-methoxybenzene. The reagents used were *p*-bromoanisole (28.5 g, 0.15 mol), magnesium (3.7 g, 0.15 mol), and dilithium tetrachlorocopper(II) (9 mL, 0.1 M in THF). Recrystallization of the crude product from ether–methanol gave 20 g (88%) of 4-methoxy-1-tetradecylbenzene: mp 38 °C; IR (KBr) 1605 cm^{-1}; NMR ($CDCl_3$) δ 0.87 (t, 3 H), 1.1–1.7 (m, 24 H), 2.57 (t, 2 H), 3.76 (s, 3 H), 6.75–7.3 (m, 4 H); MS, *m/e* 304 (M^+). Anal. ($C_{21}H_{36}O_1$) C, H.

4-Tetradecylphenol (8) was prepared in the similar fashion as described above for 2-hexadecylphenol (7). The reagents used were boron tribromide (52.62 g, 0.21 mol) and 4-methoxy-1-tetradecylbenzene (63.84 g, 0.21 mol). The yield of compound 8 was 57 g (94%): mp 70–71 °C; IR (KBr) 3400 cm^{-1}; NMR ($CDCl_3$) δ 0.9 (t, 3 H), 1.1–1.8 (m, 24 H), 2.55 (t, 2 H), 4.65 (s, 1 H), 6.6–7.15 (m, 4 H); MS, *m/e* 290 (M^+). Anal. ($C_{20}H_{34}O_1$) C, H.

3-Tetradecylphenol (9). The method used to prepare 3-methoxy-1-tetradecylbenzene was similar to that described above in the preparation of 1-hexadecyl-2-methoxybenzene. The reagents used were *m*-bromoanisole (37.4 g, 0.2 mol), magnesium (4.87 g, 0.2 mol), tetradecyl bromide (52.68 g, 0.19 mol), and dilithium tetrachlorocopper(II) (10 mL, 0.1 M in THF). The isolated yield of 3-methoxy-1-tetradecylbenzene was 52.4 g (91%): IR (neat) 1600 cm^{-1}; NMR ($CDCl_3$) δ 0.95 (t, 3 H), 1.2–1.9 (m, 24 H), 2.6 (t, 2 H), 3.85 (s, 3 H), 6.75–7.5 (m, 4 H); MS, *m/e* 304 (M^+). Anal. ($C_{21}H_{36}O_1$) C, H.

The method used to prepare 3-tetradecylphenol (9) was the same as that described above for 2-hexadecylphenol (7). The reagents used were boron tribromide (112.75 g, 0.4 mol) and 3-methoxy-1-tetradecylbenzene (136.8 g, 0.45 mol). The yield of 3-tetradecylphenol (9) was 107 g (82%): mp 38–40 °C; IR (KBr) 3310 cm^{-1}; NMR ($CDCl_3$) δ 0.95 (t, 3 H), 1.2–1.8 (m, 24 H), 2.6 (t, 2 H), 4.95 (brs, 1 H), 6.5–7.3 (m, 4 H); MS, *m/e* 290 (M^+); Anal. ($C_{20}H_{34}O$) C, H.

2,2-Dimethyl-4-[[(methylsulfonyl)oxy]methyl]-1-3-dioxolane (10). To a solution of soketal (132 g, 1.0 mol) in 500 mL of methylene chloride was added triethylamine (152.78 g, 1.5 mol) at 0 °C. Methanesulfonyl chloride (137.46 g, 1.2 mol) in 500 mL of methylene chloride was added dropwise to the above solution over 1 h. The reaction was stirred at 0 °C for 4 h, diluted with ether, washed with water, sodium bicarbonate solution, and brine, and dried $MgSO_4$). Solvents were removed under reduced pressure to give 201.8 g (96%) of compound 9, which was used directly in the next reaction without further purification: NMR ($CDCl_3$) δ 1.39 (s, 3 H), 1.46 (s, 3 H), 2.76 (s, 3 H), 3.7–4.7 (m, 5 H).

2,2-Dimethyl-4-[(2-hexadecylphenoxy)methyl]-1,3-dioxolane (11). To a suspension of prewashed (hexanes) sodium hydride (16.8 g, 50% in oil) in 200 mL of dimethylformamide was added at room temperature dropwise a solution of 2-hexadecylphenol (7) (75.05 g, 0.236 mol) in 200 mL of dimethylformamide. The mixture was refluxed for 2 h and cooled to room temperature, and 2,2-dimethyl-4-[[(methylsulfonyl)oxy]-methyl]-1,3-dioxolane (10) (59.47 g, 0.283 mol) was added. The mixture was then refluxed for 24 h, cooled, diluted with ether, washed with water, and dried. Solvents were removed under reduced pressure. Methanol was added to the residue. The white solid was collected by filtration, giving 85 g (83%) of the desired title compound 11: mp 37–38 °C; IR (KBr) 1580, 1600 cm^{-1}; NMR ($CDCl_3$) δ 0.9 (t, 3 H), 1.1–1.75 (m, 28 H), 2.6 (t, 2 H), 3.75–4.75 (m, 5 H), 6.65–7.4 (m, 4 H); MS, *m/e* 432 (M^+). Anal. ($C_{28}H_{48}O_3$) C, H.

2,2-Dimethyl-4-[(4-tetradecylphenoxy)methyl]-1,3-dioxolane (12). This compound was prepared by using the same methods as described above for compound 11. The reagents used were sodium hydride (10.8 g, 0.225 mol), 4-tetradecylphenol (8) (43.5 g, 0.15 mol), and 2,2-dimethyl-4-[[(methylsulfonyl)oxy]-methyl]-1,3-dioxolane (10) (37.8 g, 0.18 mol). The yield of the title compound was 58.7 g (97%): mp 52–53 °C; IR (KBr) 1610 cm^{-1}; NMR ($CDCl_3$) δ 0.87 (t, 3 H), 1.1–1.5 (m, 24 H), 1.4 (s, 3 H), 1.45 (s, 3 H), 2.52 (t, 2 H), 3.75–4.25 (m, 4 H), 4.25–4.65 (m, 1 H), 6.65–7.25 (m, 4 H); MS, *m/e* 404 (M^+). Anal. ($C_{26}H_{44}O_3$) C, H.

2,2-Dimethyl-4-[(3-tetradecylphenoxy)methyl]-1,3-dioxolane (13). The compound was prepared by the same procedure as that employed in the preparation of compound 11. The reagents used were sodium hydride (14.4 g, 0.3 mol), 3-tetradecylphenol (9) (58 g, 0.2 mol), and 2,2-dimethyl-4-[[(methylsulfonyl)oxy]methyl]-1,3-dioxolane (10) (50.4 g, 0.24 mol). The yield of 13 was 65.2 g (80.6%): mp 31–32 °C; IR (KBr) 1600 cm^{-1}; NMR ($CDCl_3$) δ 0.9 (5, 3 H), 1.1–1.75 (m, 24 H), 1.4 (s, 3 H), 1.45 (s, 3 H), 2.55 (t, 2 H), 3.75–4.3 (m, 4 H), 4.35–4.7 (m, 1 H), 6.55–7.35 (m, 4 H); MS, *m/e* 404 (M^+). Anal. ($C_{26}H_{44}O_3$) C, H.

3-(2-Hexadecylphenoxy)-1,2-propanediol (14). A mixture of 2,2-dimethyl-4-[(2-hexadecylphenoxy)methyl]-1,3-dioxolane (11) (83.38 g, 0.193 mol), 500 mL of methanol, and 100 mL of 1 N hydrochloric acid was heated on a steam bath for 3 h. The reaction mixture was allowed to cool. Solvents were partially evaporated, and the solid was collected, washed with ice-cold methanol, and dried under vacuum. Recrystallization from methanol–ether gave 46 g (61%) of the title compound 14: mp 70–71 °C; IR (KBr) 1600, and 3250 cm^{-1}; NMR ($CDCl_3$) δ 0.95 (t, 3 H), 1.1–1.75 (m, 28 H), 2.05 (brs, 1 H), 2.55 (brs, 1 H), 2.65 (t, 2 H), 3.75–4.5 (m, 5 H), 6.75–7.5 (m, 4 H); MS, *m/e* 392 (M^+). Anal. ($C_{25}H_{44}O_3$) C, H.

3-(4-Tetradecylphenoxy)-1,2-propanediol (15). The method used to prepare compound 15 was the same as that described above for 14. The yield of 15 was 38 g (95%): mp 81.5–83.5 °C; IR (KBr) 1608, 3400 cm^{-1}; NMR ($CDCl_3$) δ 0.9 (t, 3 H), 1.1–1.5 (m, 24 H), 2.15 (brs, 1 H), 2.55 (t, 2 H), 3.5–4.4 (m, 6 H), 6.5–7.5 (m, 4 H); MS, *m/e* 364 (M^+). Anal. ($C_{23}H_{40}O_3$) C, H.

3-(3-Tetradecylphenoxy)-1,2-propanediol (16). This compound was prepared by using the same method as described above for 14. The reagents used were 2,2-dimethyl-4-[(3-tetradecylphenoxy)methyl]-1,3-dioxolane (13) (62.2 g, 0.154 mol), 100 mL of 1 N hydrochloric acid, 500 mL of methanol, and 200 mL of tetrahydrofuran. The yield of 16 was 54 g (96%): mp 72–73 °C; IR (KBr) 1608, 3350 cm^{-1}; NMR ($CDCl_3$) δ 0.9 (t, 3 H), 1.1–1.75 (m, 24 H), 2.4 (brs, 2 H), 2.55 (t, 2 H), 3.7–4.3 (m, 5 H), 6.5–7.4 (m, 4 H); MS, *m/e* 364 (M^+). Anal. ($C_{23}H_{40}O_3$) C, H.

1-(2-Hexadecylphenoxy)-3-(triphenylmethoxy)-2-propanol (17). To a solution of 3-(2-hexadecylphenoxy)-1,2-propanediol (14) (46.26 g, 0.118 mol) in 150 mL of dry pyridine was added trityl chloride (49.34 g, 0.177 mol). The mixture was stirred under

argon at room temperature for 48 h. The reaction mixture was diluted with ether, washed with sodium bicarbonate solution, and brine, and then dried (Na_2SO_4). Solvent was evaporated and the crude product was treated with hexanes and the crystallized triphenylcarbinol and removed by filtration. Evaporation of hexanes gave 72.9 g (97%) of the desired title compound **17**. A small sample of **17** was purified by column chromatography, using hexanes–Et_2O (9:1) as eluent: IR (neat), 1600, 3450 cm^{-1}; NMR ($CDCl_3$) δ 0.95 (t, 3 H), 1.1–1.7 (m, 28 H), 2.55 (t, 2 H), 3.45 (d, 2 H), 4.0–4.4 (m, 3 H), 6.6–7.25 (m, 19 H); MS, m/e 634 (M^+). Anal. ($C_{44}H_{58}O_3$) C, H.

1-(4-Tetradecylphenoxy)-3-(triphenylmethoxy)-2-propanol (18). The procedure used to prepare **18** was the same as that described for **17**. The reagents used were 3-(4-tetradecylphenoxy)-1,2-propanediol (**15**) (17.29 g, 0.047 mol), 70 mL of dry pyridine, and trityl chloride (19.86 g, 0.071 mol). The crude product **18** was purified by column chromatography, using silica gel and hexanes–ether (9:1) as eluting solvents, giving 12 g (42%) of desired title compound **18**: IR (neat) 1608, 3450 cm^{-1}; NMR ($CDCl_3$) δ 0.9 (t, 3 H), 1.1–1.75 (m, 24 H), 2.45 (brs, 1 H), 2.55 (t, 2 H), 3.35 (d, 2 H), 4.0–4.4 (m, 3 H), 6.7–7.75 (m, 19 H); MS, m/e 606 (M^+). Anal. ($C_{42}H_{54}O_3$) C, H.

1-[(4-Methoxyphenyl)diphenylmethoxy]-3-(3-tetradecylphenoxy)-2-propanol (19). To a solution of 3-(3-tetradecylphenoxy)-1,2-propanediol (**16**) (21.84 g, 0.06 mol) in 40 mL of pyridine and 25 mL of tetrahydrofuran was added 4-methoxytrityl chloride (25.94, 0.084 mol). The mixture was stirred at room temperature, under argon, for 24 h. Solvents were removed under reduced pressure. The residue was dissolved in chloroform (500 mL), washed with sodium bicarbonate solution, and water, and dried, and the solvent was evaporated. The crude product **19** was purified by chromatography on silica gel, using hexanes–ether (5:1), giving 34.34 g (90%) of the desired compound **19**: IR (neat) 1600, 3450 cm^{-1}; NMR ($CDCl_3$) δ 0.9 (t, 3 H), 1.1–1.5 (m, 24 H), 2.45 (t, 2 H), 2.65 (brs, 1 H), 3.35 (d, 2 H), 3.76 (s, 3 H), 3.9–4.25 (m, 3 H), 6.65–7.6 (m, 18 H). Anal. ($C_{43}H_{36}O_4$) C, H.

1-[2-(Phenylmethoxy)-3-(triphenylmethoxy)propoxyl]-2-hexadecylbenzene (20). To a suspension of prewashed (hexanes) sodium hydride (8.28 g, 0.17 mol) in dimethylformamide was added dropwise a solution of 1-(2-hexadecylphenoxy)-3-(triphenylmethoxy)-2-propanol (**17**) (72.91 g, 0.115 mol) in dimethylformamide. A solution of benzyl bromide (25.57 g, 0.15 mol) was then added and the reaction mixture was stirred at room temperature for 3 h. The reaction was quenched slowly with water and extracted twice with equal mixture of ether and hexanes. Combined organic layers were dried (Na_2SO_4), and solvents were removed under reduced pressure to give 60 g (73%) of the desired title compound **20**: IR (neat) 1600 cm^{-1}; NMR ($CDCl_3$) δ 0.95 (t, 3 H), 1.1–1.7 (m, 28 H), 2.55 (t, 2 H), 3.43 (d, 2 H), 4.0–4.4 (m, 3 H), 4.85 (brs, 2 H), 6.45–7.75 (m, 24 H); MS, m/e 724 (M^+). Anal. ($C_{57}H_{64}O_3$) C, H.

1-[2-(Phenylmethoxy)-3-(triphenylmethoxy)propoxy]-4-tetradecylbenzene (21). Compound **21** was prepared by using the same method as described above in the preparation of **20**. The reagents used were sodium hydride (1.4 g, 0.03 mol), 1-(4-tetradecylphenoxy)-3-(triphenylmethoxy)-2-propanol (**18**) (13.78 g, 0.023 mol), and benzyl bromide (4.06 g, 0.024 mol). The yield of **21** was 15.7 g (99%): IR (KBr) 1608 cm^{-1}; NMR ($COCl_3$) δ 0.88 (t, 3 H), 1.1–1.7 (m, 24 H), 2.55 (t, 2 H), 3.35 (d, 2 H), 3.8–4.2 (m, 3 H), 4.65 (s, 2 H), 6.6–7.6 (m, 24 H); MS, m/e 696 (M^+). Anal. ($C_{49}H_{60}O_3$) C, H.

2-(Phenylmethoxy)-3-(2-hexadecylphenoxy)-1-propanol (22). A mixture of 1-[2-(phenylmethoxy)-3-(triphenylmethoxy)propoxy]-2-hexadecylbenzene (**20**) (49.96 g, 0.069 mol), 200 mL of methanol, 200 mL of tetrahydrofuran, and 0.5 g of *p*-toluenesulfonic acid was stirred at room temperature under argon for 24 h. Solvents were removed under reduced pressure, and the residue was dissolved in ether, washed with saturated sodium bicarbonate solution, and dried ($MgSO_4$). The solvent was evaporated. The crude product was purified by silica gel chromatography, using hexanes–ether (6:1), giving 11 g (33%) of the desired title compound **22**: mp 39–40 °C; IR (KBr) 1600, 3450 cm^{-1}; NMR ($CDCl_3$) δ 0.86 (t, 3 H), 1.1–1.75 (m, 28 H), 2.0 (t, 1 H), 2.65 (t, 2 H), 3.6–4.25 (m, 5 H), 4.8 (m, 2 H), 6.6–7.6 (m, 9 H); MS, m/e 482 (M^+). Anal. ($C_{32}H_{50}O_3$) H; C: calcd, 79.62; found 80.29.

2-(Phenylmethoxy)-3-(4-tetradecylphenoxy)-1-propanol (23). This compound was prepared by the same procedure as that employed in the preparation of compound **22**. The reagents used were 1-[2-(phenylmethoxy)-3-(triphenylmethoxy)propoxy]-4-tetradecylbenzene (**21**) (14.62 g, 0.021 mol), 50 mL of methanol, 50 mL of tetrahydrofuran, and 0.1 g of *p*-toluenesulfonic acid. The isolated yield of **23** was 7.6 g (80%): mp 50–51 °C; IR (KBr) 1608, 3400 cm^{-1}; NMR ($CDCl_3$) δ 0.95 (t, 3 H), 1.1–1.5 (m, 24 H), 2.57 (t, 2 H), 3.6–4.2 (m, 6 H), 4.8 (m, 2 H), 6.75–7.25 (m, 4 H), 7.25–7.6 (m, 5 H); MS, m/e 454 (M^+). Anal. ($C_{30}H_{46}O_3$) C, H.

2-Bromoethyl 2-(Phenylmethoxy)-3-(2-hexadecylphenoxy)propyl Phosphate (24). To a solution of 2-(phenylmethoxy)-3-(2-hexadecylphenoxy)-1-propanol (**22**) (9.64 g, 0.02 mol) in 80 mL of carbon tetrachloride was added 2-bromoethyl phosphorodichloridate (5.76 g, 0.024 mol), followed by triethylamine (2.42 g, 0.024 mol). The mixture was stirred at room temperature for 2 h and filtered through Celite, and the solvent was removed under reduced pressure. The residue was dissolved in 80 mL of tetrahydrofuran, and 80 mL of 0.5 M aqueous sodium acetate was added. The mixture was stirred for 24 h. Dilute hydrochloric acid and brine were added, and the mixture was extracted several times with ethyl acetate. The combined organic layer were washed with brine and dried, and the solvents were evaporated. The crude compound was purified by Florisil column chromatography, eluting with chloroform, chloroform–methanol (9:1), and then with chloroform–methanol (1:1), giving 12.9 g (97%) of the desired title compound **24**: IR (KBr) 1600, 3400 cm^{-1}; NMR ($CDCl_3/CD_3OD$) δ 0.9 (t, 3 H), 1.1–1.7 (m, 28 H), 2.55 (t, 2 H), 3.0–3.75 (m, 3 H), 3.8–4.7 (m, 7 H), 4.65 (brs, 2 H), 6.5–7.5 (m, 9 H); MS (FD-*p*-TSA), m/e 669, 671 (M + H). Anal. ($C_{34}H_{54}O_6BrP\cdot H_2O$) C, H, P, Br.

2-Bromoethyl 2-(Phenylmethoxy)-3-(4-tetradecylphenoxy)propyl Phosphate (25). This compound was prepared in the same manner as described above in the preparation of compound **24**. The regents used were 2-(phenylmethoxy)-3-(4-tetradecylphenoxy)-1-propanol (**23**) (4.54 g, 0.01 mol), 2-bromoethyl phosphorodichloridate (2.88 g, 0.012 mol), and triethylamine (1.21 g, 0.012 mol). The isolated yield of **25** after Florisil column chromatography was 5.3 g (83%): IR (KBr) 1608, 3500 cm^{-1}; NMR ($CDCl_3/CD_3OD$) δ 0.9 (t, 3 H), 1.15–1.8 (m, 24 H), 2.53 (t, 2 H), 3.1 (brs, 1 H), 3.4 (t, 2 H), 3.85–4.25 (m, 5 H), 4.67 (brs, 2 H), 6.67–7.2 (m, 4 H), 7.2–7.45 (m, 5 H); MS (FD), m/e 640, 642 (M + H). Anal. ($C_{32}H_{50}O_6BrP\cdot H_2O$) C, H, P, Br.

4-Hydroxy-*N*,*N*,*N*-trimethyl-9-phenyl-7-[(2-hexadecylphenoxy)methyl]-3,5,8-trioxa-4-phosphanonan-1-aminium 4-Oxide, Hydroxide, Inner Salt (26). A mixture of 2-bromoethyl 2-(phenylmethoxy)-3-(2-hexadecylphenoxy)propyl phosphate (**24**) (11.36 g, 0.017 mol), 120 mL of 40% aqueous trimethylamine, and 260 mL of chloroform–2-propanol–dimethylformamide (3:5:5) was heated at 65 °C for 5 h. The volume of solvents was reduced to half and then 150 mL of ethanol followed by silver carbonate (1.4 g) was added. The mixture was stirred at room temperature for 1 h and filtered through Celite. The solvents were removed under reduced pressure. The crude material was purified by silica gel chromatography, eluting first with chloroform–methanol–H_2O (125:25:1) and then with the same constituents (30:30:1), giving 7.2 g (66%) of compound **26**: IR (KBr) 1650 cm^{-1}; NMR ($CDCl_3/CD_3OD$) δ 0.9 (t, 3 H), 1.1–1.75 (m, 28 H), 2.5 (t, 3 H), 3.06 (brs, 9 H), 3.5 (m, 2 H), 3.8–4.4 (m, 7 H), 4.65 (m, 2 H), 6.7–7.25 (m, 4 H), 7.25–7.5 (m, 5 H); MS (FD), m/e 648 (M + H). Anal. ($C_{37}H_{62}O_6NP$) C, H, N, P.

4-Hydroxy-*N*,*N*,*N*-trimethyl-9-phenyl-7-[(4-tetradecylphenoxy)methyl]-3,5,8-trioxa-4-phosphanonan-1-aminium 4-Oxide, Hydroxide, Inner Salt (27). The method used was the same as that employed for the preparation of **26**. The reagents used were 2-bromoethyl 2-(phenylmethoxy)-3-(4-tetradecylphenoxy)propyl phosphate (**25**) (4.86 g, 0.0076 mol), 60 mL of 40% aqueous trimethylamine, and silver carbonate (0.64 g). The isolated yield of compound **27** after purification was 3.5 g (74%): IR (mull) 1608, 3400 cm^{-1}; NMR ($CDCl_3/CD_3OD$) δ 0.9 (t, 3 H), 1.1–1.5 (m, 24 HO, 2.54 (t, 2 H), 3.03 (s, 9 H), 3.35–3.55 (m, 2 H), 3.8–4.3 (m, 7 H), 4.7 (brs, 2 H), 6.7–7.2 (m, 4 H), 7.2-7.45 (m, 5 H); MS (FD-*p*-TSA), m/e 620 (M + H). Anal. ($C_{35}H_{58}NO_6P\cdot 1^1/_2 H_2O$) C, H, N, P.

2-[[Hydroxy[2-Hydroxy-3-(2-hexadecylphenoxy)propoxy]phosphinyl]oxy]-*N,N,N*-trimethylethanaminium, Hydroxide, Inner Salt (28). A mixture of 4-hydroxy-*N,N,N*-trimethyl-9-phenyl-7-[(2-hexadecylphenoxy)methyl]-3,5,8-trioxa-4-phosphanonan-1-aminium 4-oxide, hydroxide, inner salt (**26**) (5.18 g, 0.008 mol), 0.6 g, 5% palladium on carbon, 50 mL of methanol, and 50 mL of glacial acetic acid was hydrogenated in a Parr shaker for 18 h. The catalyst was filtered off through Celite, and the solvents were removed under reduced pressure at 40–50 °C, giving 4.21 g (94%) of compound **28**: IR (KBr) 1610, 3400 cm^{-1}; NMR ($CDCl_3/CD_3OD$) δ 0.9 (t, 3 H), 1.1–1.7 (m, 28 H), 2.0 (brs, 1 H), 2.55 (t, 2 H), 3.2 (brs, 9 H), 3.5–4.7 (m, 9 H), 6.75–7.5 (m, 4 H); MS (FD-*p*-TSA), *m/e* 558 (M + H). Anal. ($C_{30}H_{58}NO_6P$) H, N, P; C: calcd, 64.60; found, 64.13.

2-[[Hydroxy[2-hydroxy-3-(4-tetradecylphenoxy)propoxy]phosphinyl]oxy]-*N,N,N*-trimethylethanaminium, Hydroxide, Inner Salt (29). This compound was prepared by using the same method as described above for compound **28**. The reagents used were 4-hydroxy-*N,N,N*-trimethyl-9-phenyl-7-[(4-tetradecylphenoxy)methyl]-3,5,8-trioxa-4-phosphanonan-1-aminium 4-oxide, hydroxide, inner salt (**27**) (2.85 g, 0.0046), 0.3 g of 5% palladium on carbon, 30 mL of methanol, and 30 mL of glacial acetic acid. The yield of **29** was 2.4 g (99%): IR (KBr) 1610, 3200–3400 cm^{-1}; NMR ($CDCl_3/CD_3OD$) δ 0.9 (t, 3 H), 1.1–1.7 (m, 24 H), 2.55 (t, 3 H), 3.2 (brs, 9 H), 3.3–3.6 (m, 2 H), 3.8–4.4 (m, 4 H), 6.65–7.25 (m, 4 H); MS (FD-*p*-TSA), *m/e* 530 (M + H). Anal. ($C_{28}H_{52}N_1O_6P_1$) C, H, N, P.

4-Hydroxy-*N,N,N*-trimethyl-9-oxo-7-[(2-hexadecylphenoxy)methyl]-3,5,8-trioxa-4-phosphadecan-1-aminium 4-Oxide, Hydroxide, Inner Salt (30). A mixture of 2-[[hydroxy[2-hydroxy-3-(2-hexadecylphenoxy)propoxy]phosphinyl]oxy]-*N,N,N*-trimethylethanaminium, hydroxide, inner salt (**28**) (2.75 g, 0.005 mol), acetic anhydride (10.21 g, 0.1 mol), triethylamine (5.06 g, 0.05 mol), and 120 mL of chloroform was refluxed for 24 h. The solvents were removed under reduced pressure. The crude product was purified by silica gel column chromatography, eluting first with chloroform–methanol (9:1), then chloroform–methanol (8:1), and finally chloroform–methanol–H_2O (20:10:1), giving 1 g of the pure title compound **30**: IR (neat) 1600, 1735 cm^{-1}; NMR ($CDCl_3/CD_3OD$) δ 0.88 (t, 3 H), 1.1–1.75 (m, 28 H), 2.05 (s, 3 H), 2.55 (t, 2 H), 3.25 (brs, 9 H), 3.5–4.5 (m, 8 H), 5.15–5.6 (m, 1 H), 6.65–7.25 (m, 4 H); MS (FD-*p*-TSA), *m/e* 600 $(M + H)^+$. Anal. ($C_{32}H_{58}NO_7P\cdot 2^1/_2H_2O$) C, H, N, P.

4-Hydroxy-*N,N,N*-trimethyl-9-oxo-7-[(4-tetradecylphenoxy)methyl]-3,5,8-trioxa-4-phosphadecan-1-aminium 4-Oxide, Hydroxide, Inner Salt (31). Compound **31** was prepared by using the same procedure as described above for the preparation of **30**. The reagents used were 2-[[hydroxy[2-hydroxy-3-(4-tetradecylphenoxy)propoxy]phosphinyl]oxy]-*N,N,N*-trimethylethanaminium hydroxide, inner salt (**29**) (2.12 g, .004 mol), acetic anhydride (10.2 g, 0.1 mol), and triethylamine (4.05 g, 0.04 mol). The isolated yield was 1.8 g (79%): IR (KBr) 1608, 1735 cm^{-1}; NMR ($CDCl_3/CD_3OD$) δ 0.9 (t, 3 H), 1.1–1.7 (m, 24 H), 2.08 (s, 3 H), 2.5 (t, 2 H), 3.3 (brs, 9 H), 3.65–4.5 (m, 8 H), 5.25–5.5 (m, 1 H), 6.65–7.25 (m, 4 H); MS (FD-*p*-TSA), 572 (M + H). Anal. ($C_{30}H_{54}NO_7P\cdot{}^1/_2H_2O$) C, H, N, P.

1-[(4-Methoxyphenyl)diphenylmethoxy]-3-(3-tetradecylphenoxy)propan-2-yl Acetate (32). A mixture of 1-[(4-methoxyphenyl)diphenylmethoxy]-3-(3-tetradecylphenoxy)-2-propanol (**19**) (63.6 g, 0.1 mol), pyridine (200 mL), and acetic anhydride (60 mL) was stirred at room temperature for 72 h. The mixture was then poured onto ice and extracted several times with chloroform. The combined organic layers were washed with dilute aqueous sodium bicarbonate and water and dried ($MgSO_4$). The solvents were removed under reduced pressure. Toluene was added and removed to give 62.6 g (92%) of the desired title compound **32**: IR (neat) 1600, 1735 cm^{-1}; NMR ($CDCl_3$) δ 0.9 (t, 3 H), 1.1–1.7 (m, 24 H), 2.1 (s, 3 H), 2.55 (t, 2 H), 3.35 (d, 2 H), 3.8 (s, 3 H), 4.15 (d, 2 H), 5.25–5.55 (m, 1 H), 6.6–7.75 (m, 18 H); MS, *m/e* 678 (M^+). Anal. ($C_{45}H_{58}O_5$) C, H.

1-Hydroxy-3-(3-tetradecylphenoxy)propan-2-yl Acetate (33). Three-hundred and twenty grams of silicic acid–boric acid[10] was suspended in 800 mL of petroleum ether (bp 30–60 °C) and packed in a column (5 × 55 cm). 1-[(4-Methoxyphenyl)diphenylmethoxy]-3-(tetradecylphenoxy)propan-2-yl acetate (**31**) (30 g, 0.044 mol) was dissolved in 50 mL of petroleum ether and introduced to the column. The column was then eluted with 3 L of petroleum ether followed by 4.5 L of petroleum ether–ether (19:1) until all the 4-methoxytriphenylcarbinol had eluted. Petroleum ether–ether (3:1) was used to elute the desired product, giving 14.3 g (80%) of **33**: NMR ($CDCl_3$) δ 0.9 (t, 3 H), 1.1–1.7 (m, 24 H), 2.1 (s, 3 H), 2.58 (t, 2 H), 3.85–4.35 (m, 5 H), 5.15–5.5 (m, 1 H), 6.65–7.35 (m, 4 H); MS, *m/e* 406 (M^+). Anal. ($C_{25}H_{42}O_4$) C, H.

2-Bromoethyl 2-(Acetyloxy)-3-(3-tetradecylphenoxy)propyl Phosphate (34). This compound was prepared in the same manner as described above for the preparation of compound **24**. The reagents used were 1-hydroxy-3-(3-tetradecylphenoxy)propan-2-yl acetate (**33**) (4.06 g, 0.01 mol), 2-bromoethyl phosphorodichlorodate (2.88 g, 0.012 mol), and triethylamine (1.21 g, 0.012 mol). The isolated yield of **33** after Florisil column chromatography was 3.8 g (64%): IR (KBr) 1600, 1735 cm^{-1}; NMR ($CDCl_3/CD_3OD$) δ 0.90 (t, 3 H), 1.1–1.75 (m, 24 H), 2.05 (s, 3 H), 2.55 (t, 2 H), 2.75 (m, 2 H), 3.45 (m, 2 H), 4.0–4.5 (m, 5 H), 5.35 (m, 1 H), 6.5–7.4 (m, 4 H); MS (FD-*p*-TSA), *m/e* 593, 595 (M + H). Anal. ($C_{27}H_{47}O_7BrP\cdot H_2O$) H, P; C: calcd, 53.02; found, 53.50; Br: calcd, 13.06; found, 12.13.

4-Hydroxy-*N,N,N*-trimethyl-9-oxo-7-[(3-tetradecylphenoxy)methyl]-3,5,8-trioxa-4-phosphadecan-1-aminium 4-Oxide, Hydroxide, Inner Salt (35). A mixture of 2-bromoethyl 2-(acetyloxy)-3-(3-tetradecylphenoxy)propyl phosphate (**34**) (2.96 g, 0.005 mol), 50 mL of 33% trimethylamine in acetonitrile, and 30 mL of chloroform was heated to reflux for 4 h. An additional 20 mL of 33% trimethylamine in acetonitrile was added and the reaction refluxed for 16 h. The reaction was allowed to cool, and the solvents were evaporated. The residue was chromatographed on silica gel (100 g), using chloroform–methanol (9:1) followed by chloroform–methanol–water (120:25:4), to give 1.8 g (63%) of the desired product **34**: IR (KBr) 1725 cm^{-1}; NMR ($CDCl_3/CD_3OD$) δ 0.85 (t, 3 H), 1.1–1.75 (m, 24 H), 2.03 (s, 3 H), 2.5 (t, 2 H), 3.2 (s, 9 H), 3.25–4.5 (m, 8 H), 5.3 (m, 1 H), 6.55–7.25 (m, 4 H); MS (FAB), 572 (M + H); Anal. ($C_{30}H_{54}N_1O_7P\cdot H_2O$) C, H, N, P.

Acknowledgment. We thank Dr. L. Gehrlein and staff for microanalytical determinations, Dr. M. Siegel and G. O. Morton and staff for spectral data, R. A. Gabel for his assistance with the hypotension assay, and D. Nytko for her assistance with the platelet aggregation assay.

Registry No. **1a**, 85026-88-6; **1b**, 77286-68-1; **2**, 90857-71-9; **3**, 90857-72-0; **4**, 90857-73-1; **5**, 90857-74-2; **6**, 90857-75-3; **7**, 25401-86-9; **8**, 25401-89-2; **9**, 90866-51-6; **10**, 34331-40-3; **11**, 90857-76-4; **12**, 90857-77-5; **13**, 90857-78-6; **14**, 90857-79-7; **15**, 90857-80-0; **16**, 90857-81-1; **17**, 90857-82-2; **18**, 90857-83-3; **19**, 90857-84-4; **20**, 90857-85-5; **21**, 90857-86-6; **22**, 90857-87-7; **23**, 90857-88-8; **24**, 90857-89-9; **25**, 90857-90-2; **26**, 90857-91-3; **27**, 90857-92-4; **28**, 90857-93-5; **29**, 90857-94-6; **30**, 90857-95-7; **31**, 90857-96-8; **32**, 90857-97-9; **33**, 90857-98-0; **34**, 90857-99-1; **35**, 90858-00-7; *n*-octadecanoic acid chloride, 112-76-5; tris[(trimethylsilyl)oxy]ethylene, 69097-20-7; 2-bromoethyl phosphorodichloridate, 4167-02-6; *o*-bromoanisole, 578-57-4; hexadecyl iodide, 544-77-4; 1-hexadecyl-2-methoxybenzene, 22165-09-9; *p*-bromoanisole, 104-92-7; tetradecyl bromide, 112-71-0; 4-methoxy-1-tetradecylbenzene, 22165-05-5; *m*-bromoanisole, 2398-37-0; 3-methoxy-1-tetradecylbenzene, 22161-96-2; soketal, 22323-83-7.