Russian Journal of General Chemistry, Vol. 71, No. 6, 2001, pp. 873–880. Translated from Zhurnal Obshchei Khimii, Vol. 71, No. 6, 2001, pp. 930–937.

New Approach to the Synthesis of Phosphamide Models of Cationic Phosphatidyl Cholines

D. A. Predvoditilev, S. V. Suvorkin, and E. E. Nifant'ev

Moscow Pedagogical State University, Moscow, Russia

Received January 25, 2000

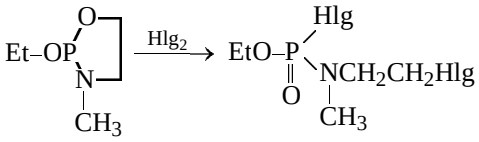
Abstract — A promising approach to the synthesis of phosphamide lipid constructions of cationic type on the basis of cyclophosphites and -amidophosphites is considered. The latter easily undergo oxidative decyclization under the action ofchlorine or bromine. The resulting halophosphates and amidohalophosphates take part in various reactions finally leading to models of cationic phosphatidyl cholines which are among priority objects of modern phospholipid chemistry.

Synthesis of phosphatidyl cholines and their analogs and models is of great scientific importance and, therefore, it has attracted considerable interest [1, 2]. Important contribution in the progress of this area of reasearch has been made by recent proposals concerning the use of trivalent phosphorus compounds and phosphocyclic systems as basic starting materials [3, 4]. In the present work we have combined these two lines, aiming at elaborating phospholipid syntheses on the basis of cyclophosphites and -amidophosphites of various classes. Our attention to phosphamide systems is explained by their availability, high reactivity, as well as prospective use of phosphamide glycerides in medical and biological studies [5–8]. The latter is due to the fact that phospholipids containing a phosphamide function are to a certain extent isosteric to phosphates and much differ from them in polarity [4]. Therefore, enzymologic comparison of glycerol-derived phosphates and phosphoramidates may prove very useful for study of general regularities of lipid metabolism as well as for design of new bioregulators. This circumstance stimulates investigations on phosphamide lipids for characterization of enzymes, such as phosphohydrolases [9, 10].

The method is based on the reaction of alkylene phosphites and dialkylamidophosphites with halogens. Note that analogous reactions involving phosphorus(III) cyclic systems have already been used for preparing glycerophospholipids. Hence, phosphatidyl cholines have been prepared by the reaction of halogens (Br_2, Cl_2) with glycerocyclophosphites [11–13], and models of phosphamide glycerophospholipids, by the reaction of the same halogens with glycerocycloamidophosphites [6].

In the present work we offer a new approach to application of this reaction in phospholipid synthesis. It includes synthesis of new phosphorylating reagents, unsymmetrical halophosphates, by halogenation of the simplest alkylene phosphites and amidophosphites and subsequent reaction of the products with glycerol derivatives. An advantage of this new approach with inverted sequence of formation of the phosphoglycerol unit in phosphoglycerol molecules is that the glycerol fragment is included in phospholipid molecules in the final stage of the synthesis and that it opens up possibilities for synthesis of a broad range of various analogs phospholipids. It is important that many of such analogs are difficult to prepare, if any, by previously reported methods.

In the first stage we explored reactions of cyclic phosphites and amidophosphites with halogens. 2-Ethoxy-3-methyl-1,3,2-oxazaphospholane (**I**) was chosen as the ester substrate.



I **II, III**

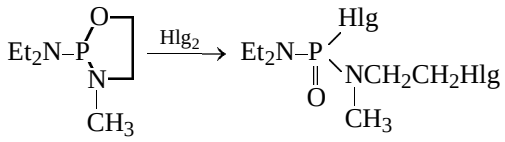
Hlg = Br (**II**), Cl (**III**).

The reactions of compound **I** with halogens were carried out in chloroform at –20°C for 10 min.

Note that this reaction is regioselective: Only an endocyclic ester bond is cleaved. This is evidently explained by the strain of the phospholane ring, resulting from the increase in the coordination number of phosphorus, produced by the reaction of the latter with halogen.

1070-3632/01/7106-0873 $25.00 © 2001 MAIK "Nauka/Interperiodica"

Under analogous conditions bromine and chlorine reacted with 2-diethylamino-3-methyl-1,3,2-oxazaphospholane (**IV**).

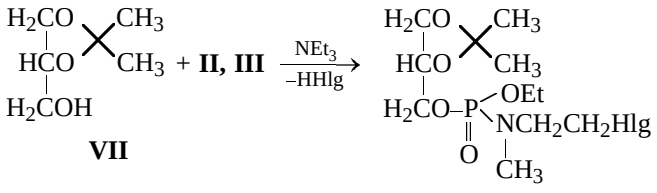


IV **V, VI**

Hlg = Br (**V**), Cl (**VI**).

Halides **II, II, V**, and **VI** were isolated by vacuum distillation in 68–91% yields. Their individuality and structure were confirmed by ^{31}P and ^{1}H NMR spectra. The ^{31}P NMR spectra showed singlets at δ 6.3, 16.7, 20.4, and 6.3 ppm. The ^{1}H NMR spectra contained a characteristic doublet of protons of the *N*-methyl group at δ 2.6–2.7 ppm ($^{3}J_{PH}$ 14 Hz) and a multiplet of methylene protons of the $N(CH_3)CH_2$ group at δ 3.3 ppm. The spectra of amidoesters **II** and **III** displayed signals of methyl and methylene protons of the *O*-ethyl group at δ 1.3 and 4.2 ppm respectively, while the spectra of diamides **V** and **VI**, a triplet of methyl protons and a multiplet of methylene protons of the *N*-ethyl groups at δ 1.1 and 3.1 ppm. In addition, the spectra of bromophosphates **II** and **V** contained multiplets at δ 3.43 ppm due to protons of the bromomethyl groups, while the spectra of chlorophosphates **III** and **VI**, triplets due to protons of the chloromethyl groups at δ 3.6 ppm ($^{3}J_{HH}$ 6 Hz).

Amidohalophosphates **II and III** were also involved in reaction with 1,2-*O*-isopropylideneglycerol (**VII**) in the presence of equimolar amount of triethylamine. The reaction gave glyceroamidophosphates **VIII, IX**.

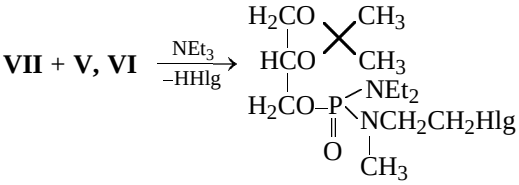


VII **VIII, IX**

Hlg = Br (**VIII**), Cl (**IX**).

In a similar way, 1,2-*O*-isopropylideneglycerol (**VII**) was reacted with halodiamides **V** and **VI** to obtain glyceroamidophosphates **X** and **XI**.

The reaction progress was controlled by ^{31}P NMR spectroscopy. Bromides **II** and **V** reacted with the glycerol derivative much more vigorously and in higher yields than chlorides **III** and **VI**. Hence, the



X, XI

Hlg = Br (**X**), Cl (**XI**).

reaction of 1,2-*O*-isopropylideneglycerol **VII** with bromides **II** and **V** was complete in several days at room temperature, and the yields of phosphoglycerols were 83 and 76%, respectively. At the same time, the reaction with chlorides **III** and **VI** could be accomplished only under heating at 80°C. Therewith, the yield of glyceroamidophosphate **IX** in the reaction with chloride **III** was 18% yield, whereas the yield of glyceroamidophosphate **XI** in the reaction with chloride **V** was as low as a few percent.[1]

Note that due to the high yield of bromides **II** and **V** in the reaction of cyclophosphites **I** and **IV** with bromine, these compounds can be reacted with 1,2-*O*-isopropylideneglycerol (**VII**) without isolation. In this case, *O*-ethyl glyceroamidophosphate **VIII** is higher (69% after two stages) than in the reaction of compound **VII** with individual phosphorylating agent **II** (56% after two stages).

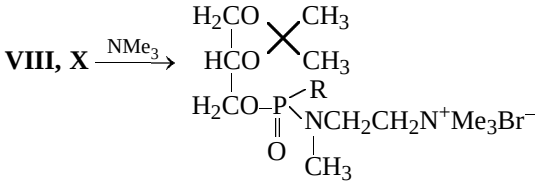
The ^{31}P NMR spectra of glycerophosphates **VIII**–**X** contain broadened singlets in expected regions (δ_P 10.1, 10.1, and 17.5 ppm, respectively). Inspite of the fact that compounds **VIII**–**X**, due to the presence of chiral phosphorus atoms and β-carbon atoms of the glycerol fragment, are mixtures of two pairs of diastereomers, we could not observe signals of individual diastereomers.

The ^{1}H NMR spectra of glycerophosphates **VII**–**X** compared with those of halides **II, III**, and **V** show two new singlets of methyl protons from the isopropylidene protective group at δ 1.3 and 1.4 ppm and multiplets of methylene and methine protons of the glycerol system at δ 3.8–4.3 ppm. Furthermore, the diastereomeric anisochronicity causes splitting of signals of the *O*-ethyl group in the spectra of glyceroamidophosphates **VIII** and **IX** and of methyl protons of the *N*-ethyl groups of glyceroamidophosphate **X** (Δδ 0.02 ppm), as well as complication of signals of the majority of other protons [14].

[1] Glyceroamidophosphates **IX** and **XI** can be prepared in good yields according to the procedure [6].

For the presented data on the relative reactivities of bromophosphates **II** and **V** and chlorophosphates **III** and **VI** toward glycerol derivative **VII** we can conclude that bromophosphates are more suitable for this reaction.

Quaternization of trimethylamine with glycerophosphobromoalkanes **VIII** and **X** gave phosphamide **XII** and phosphodiamide **XIII** models of cationic phosphocholines.



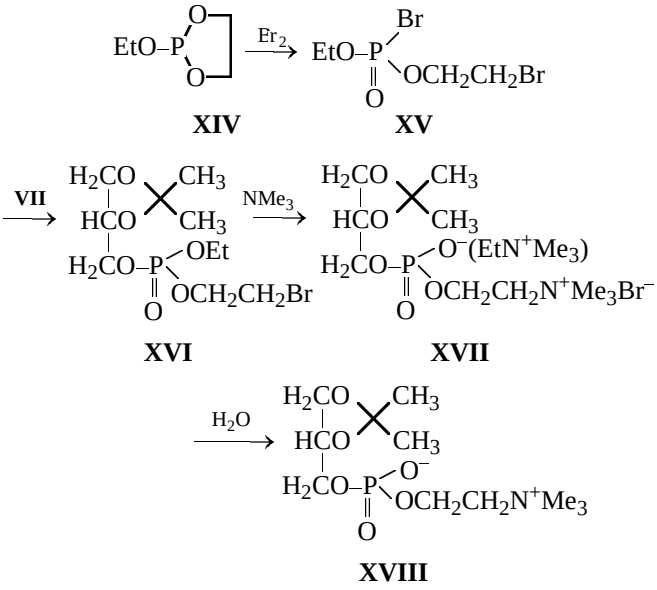
XII, XIII

R = OEt (**XII**), NEt_2 (**XIII**).

The reactions with trimethylamine were carried out in a sealed ampule in benzene solution at 80°C. The yields of glycerophosphocholines **XII** and **XIII** were 71 and 80%, respectively.

The ^{31}P NMR spectra of lipid betaines **XII** and **XIII** contained broadened singlets with chemical shifts close to those of the parent phosphates **VIII** and **X**. The ^{1}H NMR spectra of phospholipids **XII** and **XII** compared with those of glycerophosphobromoalkanes **VIII** and **X** showed a new singlet belonging to methyl protons of the ammonium group at δ 3.4 ppm and downfield shifted signals of β-methylene protons of the $CH_2CH_2N^+(CH_3)_3$ group (δ 3.5 ppm).

Further we studied reactions of alkylene phosphites and amidophosphites with halogens in relation to the lipid chemistry. The general scheme of synthesis of phospholipid models from cyclic phosphites was similar to that described above. With ethyl ethylene phosphite (**XIV**) as the starting compound, the final product was glycerophosphocholine **XVIII**.



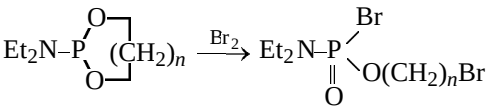
Let us dwell on some specific features of this sheme. It was found that dialkyl bromophosphate **XV** is much less stable in isolation and handling than the above-described amidobromophosphates **II** and **V** and amidochlorophosphates **III** and **VI**. Therefore, compound **XV** was reacted with 1,2-*O*-isopropylideneglycerol (**VII**) without isolation. The yield of glycerophosphate **XVI** after two stages was 82%. It is important that the reaction of glycerophosphate **XVI** with triethylamine involved, along with quaternization of the amine due to cleavage of the C–Br bond, elimination of the *O*-ethyl group. In this respect the reaction with glycerophosphate **XVI** differs from the similar reaction with glyceroamidophosphate **VIII**: In the latter reaction no elimination of the *O*-ethyl group is observed. The difference in these two similar reactions is determined by different electronic effects in the XP(O)OEt fragment of the ethoxy group and the amino group X.

Binary salt **XVII** was obtained in 72% yield. It was converted to glycerophosphocholine **XVIII** by treatment with aqueous methanol. The yield of glycerophosphocholine **XVIII** after chromatographic purification on a silica gel column was 83%. Physicochemical characteristics of compound **XVIII** were in agreement with published data [15].

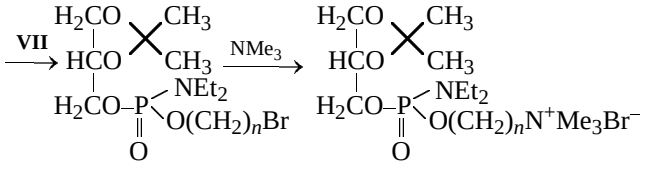
The individuality and structure of compounds **XV**–**XVIII** were proved by physicochemical methods (see Experimental). The ^{1}H NMR spectra of bromophosphate **XV** and dialkyl glycerophosphate **XVI** contain well-defined triplet and multiplet of methyl and methylene protons of the *O*-ethyl group at 1.3 and 4.1 ppm, as well as multiplets of methylene protons of the CH_2Br group at 3.5 ppm and the $PO\mathit{CH}_2CH_2Br$ group at 4.5 ppm. In the spectrum of binary salt **XVIII**, methyl protons from the ethyl group of the EtN^+Me_3 cation give a triplet at 1.36 ppm, and methylene protons of this cation appear as a multiplet at 3.69 ppm. Methyl protons of the CH_3N^+Et cation give a singlet at 2.98 ppm, while those of the $N^+Me_3Br^-$ group, a singlet at 3.44 ppm. The convertion of binary salt **XVII** to phosphocholine **XVIII** have produced expected spectral changes: The signals of the EtN^+Me_3 group have disapeared, and the N^+Me_3 methyl singlet has shifted to 3.51 ppm.

By treatment of ethylene and trimethylene amidophosphites **XIX** and **XX** with bromine with subsequent realization of the above-described general

scheme of synthesis of nitrogenous phospholipids we obtained the amide analog of cationic glycerophosphocholine **XXV** and its homo derivative **XXVI**.



XIX, XX **XXI, XXII**



XXIII, XXIV **XXV, XXVI**

The reaction and product isolation were carried out by the above-described procedures for amidophosphates **II, VIII**, and **XII**. The intermediate phosphates **XXI**–**XXIV** and the final glycerophosphocholines **XXV, XXVI** were obtained in 64–83% yields. Note the lower yield of propyl glycerophosphate **XXIV** (64%) compared with ethyl glycerophosphate **XXIII** (87%).

Comparison of the chemical properties of bromides and chlorides **II, III, V, VI, XV, XXI**, and **XXII** reveals some regular trends. The most stable to handling among bromides **II, V, XV, XXI**, and **XXII** is diamidobromophosphate **V**. It remains intact in an inert atmosphere at room temperature for 2 weeks. Alkyl amidobromophosphates **II, XXI**, and **XXII** are less stable and under the above-described conditions decompose within a week. Dialkyl bromophosphate **XV** proved to be the least stable. We failed to isolate it pure because on attempted high-vacuum distillation (10^{-4} mm) it began to decompose already at a bath temperature of 45°C. Note that at 5°C bromides **II, V, XXI**, and **XXII** can be stored for 1 month. Contrary to bromides **II, V, XV, XXI**, and **XXII**, chlorides **III** and **VI** are much more stable and can be handled in an inert atmosphere for several months.

The stability of chlorides **II, III, V, VI, XV, XXI**, and **XXII** on handling correlates with their reactivity toward 1,2-*O*-isopropylideneglycerol (**VII**). Thus, dialkyl bromophosphate **XV** is the most active. Its reaction is complete at room temperature in 10 h. The reaction of alkyl amidobromophoshates **II, XXI**, and **XXII** at this temperature takes 24 h to complete, and that of diamidobromophosphate **V**, 72 h. Chlorophosphates **III** and **VI** are the least reactive toward 1,2-*O*-isopropylideneglycerol (**VII**).

To complete the discussion of the new procedure for preparing complex models of glycerophospholipids, based on the preparation and use of new effective phosphorylating agents, bromophosphates, we can conclude that that this procedure is preparatively expedoient. It permits one to easily and effectively control hydrophilic surrounding of phosphorus by substitution of ester bonds by one or two phosphamide functions, as well as the number of methylene units in the hydrophilic part of the molecule. In this connetion the classical procedure of phosphorylation, widely used in phospholipid chemistry and based on the traditional chlorophosphates [1, 2], can be considerably improved by using corresponding bromophosphates for creation of the phosphorus entity. It is important that in this way we could obtain a series of model phospholipids with different types of exocyclic bond with the cationic choline residue, which can used to success in enzymological studies as carriers of biologically active compounds, as well as for solving other problems of physicochemical biology.

EXPERIMENTAL

The ^{1}H NMR spectra of compounds **II, III, V, VI, VIII**–**XIII**, and **XXI**–**XXVI** in $CDCl_3$ were measured on a Bruker WM-250 instrument (250 MHz) against internal TMS; the spectra were assigned on the basis of double magnetic resonance data. The ^{31}P–{^{1}H} NMR spectra of compounds **II, III, V, VI, VIII**–**XIII, XV**–**XVIII**, and **XXI**–**XXVI** in $CHCl_3$ were recorded on a Bruker WP-80 SY spectrometer (32.4 MHz) against external 85% phosphoric acid.

Column chromatography was carried out on a 15-mm column of Silica gel L 100-200 μm. Thin-layer chromatography was carried out on Silufol UV-254 plates; eluents 3:1 benzene–dioxane (A), 3:1 hexane–dioxane (B), 3:1 chloroform–methanol (C), or 4:1 methanol– water (D); development in iodine vapor or by calcination at 250–300°C. The melting points were measured in a sealed capillary, heating rate 1 deg/min.

All experiments with trivalent phosphorus compounds were carried out under dry argon. The solvents were dried according to standard procedures. 2-Ethoxy-3-methyl-1,3,2-oxazaphospholane (**I**) and 2-(dimethylamino)-3-methyl-1,3,2-oxazaphospholane **IV** were prepared according to [16], 2-ethoxy-1,3,2-dioxaphospholane (**XIV**), according to [17], 2-(diethylamino)-1,3,2-dioxaphospholane (**XIX**), and 2-(diethylamino)-1,3,2-dioxaphosphorinane (**XX**), according to [18]. Physicochemical characteristics of the products agreed well with published data.

Ethyl [(2-bromoethyl)methylamido]bromophosphate (II). To a solution of 0.3 g of oxazaphos-

pholane **I** in 3 ml of chloroform, a solution of 0.32 g of bromine in 1 ml of chloroform was added dropwise with vigorous stirring at –30°C. After 10 min, the solvent was removed in a vacuum. Amidophosphate **II** was isolated pure by vacuum distillation (10^{-4} mm) at a bath temperature of 48–50°C. Yield 0.42 g (68%), n_D^{20} 1.5122, R_f 0.81 (A), 0.67 (B), 0.86 (C). ^{1}H NMR spectrum, δ, ppm: 1.25 t (3H, OCH_2CH_3, $^3J_{HH}$ 6.22 Hz), 2.62 d (3H, NCH_3, $^3J_{PH}$ 9.81 Hz), 3.37–3.43 m (4H, NCH_2CH_2Br), 4.15 m (2H, OCH_2CH_3). ^{31}P NMR spectrum, δ_P, ppm: 6.3 s. Found, %: C 19.31; H 3.90; P 9.96. $C_5H_{12}Br_2NO_2P$. Calculated, %: C 19.44, H 3.90; P 10.03.

Ethyl [(2-chloroethyl)methylamido]chlorophosphate (III). Through a solution of 0.3 g of oxazaphospholane **I** in 3 ml of chloroform, dry chlorine was passed at –30°C until a stable green coloration of the solution appeared (ca. 10 min). The solvent and excess chlorine were removed in a vacuum. Amidophosphate **III** was isolated pure by vacuum distillation (10^{-4} mm) at a bath temperature of 44–56°C. Yield 0.39 g (88%), n_D^{20} 1.4670, R_f 0.82 (A), 0.36 (B), 0.88 (C). ^{1}H NMR spectrum, δ, ppm: 1.42 t (3H, OCH_2CH_3, $^3J_{HH}$ 7.4), 2.80 d (3H, NCH_3, $^3J_{PH}$ 13.69 Hz), 3.39 m (2H, NCH_3), 3.64 t (2H, CH_2Cl, $^3J_{HH}$ 7.22 Hz), 4.23 m (2H, OCH_2CH_3). ^{31}P NMR spectrum, δ_P, ppm: 16.7 s. Found, %: C 27.35; H 5.52; P 14.03. $C_5H_{12}Cl_2NO_2P$. Calculated, %: C 27.29; H 5.50, P 14.08.

[(2-Bromoethyl)methylamido](diethylamido)phosphoric bromide (V) was obtained similarly to amidophosphate **II** from 0.35 g of phospholane **IV** and 0.32 g of bromine. Diamidophosphate **V** was isolated pure by vacuum distillation (10^{-4} mm) at a bath temperature of 78–80°C. Yield 0.56 g (84%), n_D^{20} 1.5203, R_f 0.77 (A), 0.63 (B), 0.81 (C). ^{1}H NMR spectrum, δ, ppm: 1.09 t [6H, $N(CH_2CH_3)_2$, $^3J_{HH}$ 7.09 Hz], 2.65 d (3H, NCH_3, $^3J_{PH}$ 13.56 Hz), 3.12 q [4H, $N(CH_2CH_3)_2$, $^3J_{HH}$ 7.09 Hz], 3.28 m [2H, $N(CH_3)CH_2$], 3.43 m (2H, CH_2Br). ^{31}P NMR spectrum, δ_P, ppm: 20.4 s. Found, %: C 24.86; H 5.12; P 9.07. $C_7H_{17}Br_2N_2OP$. Calculated, %: C 25.02; H 5.10; P 9.22.

[(2-Chloroethyl)methylamido](diethylamido)phosphoric chloride (VI) was obtained analogously to amidophosphate **III** from 0.35 g of phospholane **IV.** Diamidophosphate **VI** was isolated pure by vacuum distillation (10^{-4} mm) at a bath temperature of 73–75°C. Yield 0.45 g (91%), n_D^{20} 1.4812, R_f 0.76 (A), 0.47 (B), 0.91 (C). ^{1}H NMR spectrum, δ, ppm: 1.07 t [6H, $N(CH_2CH_3)_2$, $^3J_{HH}$ 7.11 Hz], 2.66 d (3H, NCH_3, $^3J_{PH}$ 12.6 Hz), 3.04 m [4H, $N(CH_2CH_3)_2$], 3.34 m [2H, $N(CH_3)CH_2$], 3.56 t (2H, CH_2Cl, $^3J_{HH}$ 6.21 Hz). ^{31}P NMR spectrum, δ, ppm: 27.9 s. Found, %: C 33.89; H 6.90; P 12.48. $C_7H_{17}Cl_2N_2OP$. Calculated, %: C 34.02; H 6.94; P 12.53.

1,2-*O*-Isopropylideneglycero-3-[ethyl [(2-bromoethyl)methylamido]phosphate] (VIII). To a solution of 0.52 g of bromide **II** in 2 ml of benzene, a solution of 0.26 g of 1,2-*O*-isopropylideneglycerol (**VII**) and 0.2 g of triethylamine in 1 ml of benzene was added dropwise with stirring. The reaction mixture was kept at room temperature for 24 h. The triethylamine hydrobromide was filtered off, the solvent was removed in a vacuum, and the residue was applied to a silica gel column filled with benzene and eluted with 150 ml of a 5 : 1 benzene–dioxane mixture. The solvents were removed in a vacuum, and the product was kept in a vacuum (1 mm) for 3 h at 40°C. Yield 0.6 g (83%), n_D^{20} 1.4676, R_f 0.53 (A), 0.36 (B), 0.86 (C). ^{1}H NMR spectrum, δ, ppm: 1.33 t (3H, OCH_2CH_3, $^3J_{HH}$ 6.83 Hz), 1.36 s (3H), 1.42 s [3H, $C(CH_3)_2$], 2.73 d (3H, NCH_3, $^3J_{PH}$ 9.81 Hz), 3.41 m [2H, $N(CH_3)CH_2$], 3.44 m (2H, CH_2Br), 3.83 m (1H), 3.96 m (1H) (CH_2CHCH_2OP), 4.03 m (2H, $POCH_2 \cdot CH_3$, $^3J_{PH}$ 7.68 Hz), 4.07 m (2H, CH_2CHCH_2OP), 4.31 m (1H, CH_2CHCH_2OP, $^3J_{HH}$ 5.55 Hz). ^{31}P NMR spectrum, δ_P, ppm: 10.06 br.s. Found, %: C 36.53; H 6.40; P 8.56. $C_{11}H_{23}BrNO_5P$. Calculated, %: C 36.68; H 6.44; P 8.60.

1,2-*O*-Isopropylideneglycero-3-[ethyl [(2-chloroethyl)methylamido]phosphate] (IX). To a solution of 0.44 g of chloride **III** in 2 ml of benzene, a solution of 0.26 g of 1,2-*O*-isopropylideneglycerol (**VII**) and 0.2 g of triethylamine in 1 ml of benzene was added dropwise. The reaction mixture was kept for 12 h at 80°C. The triethylamine hydrochloride was filtered off, the solvent was removed in a vacuum, and the residue was applied to a silica gel column (10 g) filled with benzene and eluted with 250 ml of a 7 : 1 benzene–dioxane mixture. The solvents were removed in a vacuum, and the product was kept in a vacuum (1 mm) for 3 h at 40°C. Yield 0.11 g (18%); n_D^{20} 1.4530 (n_D^{20} 1.4529 [6]); R_f 0.45 (A), 0.18 (B), 0.84 (C). ^{1}H NMR spectrum, δ, ppm: 1.12 t (3H, $OCH_2 \cdot CH_3$), 1.39 s (3H), 1.50 s (3H) [$C(CH_3)_2$], 2.58 d (3H, NCH_3), 3.22 m [2H, $N(CH_3)CH_2$], 3.42 t (2H, CH_2Cl), 3.80 m (1H), 3.92 m (1H) (CH_2CHCH_2OP), 4.00–4.12 m (4H, $CH_2CHCH_2OPOCH_2CH_3$), 4.20 m (1H, CH_2CHCH_2OP). ^{31}P NMR spectrum, δ_P, ppm: 10.08 br.s. Found, %: C 41.77; H 7.26; P 9.75. $C_{11}H_{23}ClNO_5P$. Calculated, %: C 41.84; H 7.34; P 9.81.

1,2-*O*-Isopropylideneglycero-3-[(2-bromoethyl)-methylamido](diethylamido)phosphate] (X) was prepared analogously to glyceroamidophosphate **VIII**

from 0.67 g of bromide **V**, 0.26 g of 1,2-*O*-isopropylideneglycerol (**VII**) and 0.2 g of triethylamine for 72 h. Glycerophosphate **X** was isolated pure by vacuum distillation (10^{-4} mm) at a bath temperature of 120°C. Yield 0.59 g (76%), n_D^{20} 1.4674, R_f 0.37 (A), 0.25 (B), 0.90 (C). ^{1}H NMR spectrum, δ, ppm: 1.02 t [6H, $N(CH_2CH_3)_2$], 1.27 s (3H), 1.33 s (3H) [$C(CH_3)_2$], 2.60 d (3H, NCH_3, $^3J_{PH}$ 9.61 Hz), 2.95 m [4H, $N(CH_2CH_3)_2$, $^3J_{PH}$ 6.96 Hz], 3.30 m [2H, $N(CH_3)CH_2$], 3.34 m (2H, CH_2Br), 3.74 m (1H), 3.85 m (1H) (CH_2CHCH_2OP), 3.98 m (2H, CH_2·$CHCH_2OP$), 4.22 m (1H, CH_2CHCH_2OP, $^3J_{HH}$ 5.6 Hz). ^{31}P NMR spectrum, δ_P, ppm: 17.5 br.s. Found, %: C 40.13; H 7.20; P 7.82. $C_{13}H_{28}BrN_2O_4P$. Calculated, %: C 40.32; H 7.29; P 7.99.

1,2-*O*-Isopropylideneglycero-3-[(2-chloroethyl)-methylamido](diethylamido)phosphate] (XI) was prepared analogously to glyceroamidophosphate **IX** from 0.49 g of chloride **VI**, 0.26 g of 1,2-*O*-isopropylideneglycerol (**VII**), and 0.2 g of triethylamine for 24 h. Yield 0.02 g (3%), R_f 0.40 (A), 0.34 (B), 0.81 (C). ^{31}P NMR spectrum, δ_P, ppm: 17.6 br.s.

1,2-*O*-Isopropylideneglycero-3-[ethyl [2-(dimethylammonio)ethylamido]phosphate] bromide (XII) A sealed ampule with a solution of 0.47 g of amidophosphate **VIII** and 0.77 g of trimethylamine in 2 ml of anhydrous benzene was kept at 80°C for 6 h. The solvent was removed in a vacuum, and the residue was dissolved in chloroform and precipitated with diethyl ether. The resulting viscous oil was washed with diethyl ether (2 × 2 ml) and kept for 3 h in a vacuum (10^{-4} mm) over P_2O_5 for 40°C. Yield 0.39 g (71%), R_f 0.23 (C), 0.52 (D). ^{1}H NMR spectrum, δ, ppm: 1.21 t (3H, OCH_2CH_3, $^3J_{HH}$ 6.83 Hz), 1.23 s (3H), 1.30 s (3H) [$C(CH_3)_2$], 2.68 d (3H, $PNCH_3$, $^3J_{PH}$ 9.82 Hz), 3.42 s [9H, $N^+(CH_3)_3$], 3.52 m [2H, $CH_2N^+(CH_3)_3$], 3.68 m [2H, $PN(CH_3)CH_2$], 3.92 m (6H, $CH_2CHCH_2OPOCH_2$), 4.19 m (1H, CH_2CHCH_2OP). ^{31}P NMR spectrum, δ_P, ppm: 10.2 br.s. Found, %: C 40.01; H 7.74; P 7.28. C_{14}·$H_{32}BrN_2O_5P$. Calculated, %: C 40.10; H 7.69; P 7.39.

1,2-*O*-Isopropylideneglycero-3-[(diethylamido)-[2-(dimethylammonio)ethylamido]phosphate] bromide (XIII) was prepared analogously to bromomethylate **XII** from 0.5 g of amidophosphate **X** and 0.77 g of trimethylamine. Yield 0.46 g (80%). R_f 0.19 (C), 0.54 (D). ^{1}H NMR spectrum, δ, ppm: 1.12 t [6H, $N(CH_2CH_3)_2$, $^3J_{HH}$ 7.26 Hz], 1.34 s (3H), 1.42 s (3H) [$C(CH_3)_3$], 2.78 d (3H, $PNCH_3$, $^3J_{PH}$ 10.25 Hz), 3.04 m [4H, $N(CH_2CH_3O_2]$], 3.55 s [9H, $N^+(CH_3)_3$], 3.80 m [4H, $PN(CH_3)CH_2CH_2$], 4.04 m (4H, CH_2·$CHCH_2OP$), 4.33 m (1H, CH_2CHCH_2OP). ^{31}P NMR spectrum, δ_P, ppm: 18.3 br.s. Found, %: C 42.93; H 8.44; P 6.88. $C_{16}H_{37}BrN_3O_4P$. Calculated, %: C 43.05; H 8.36; P 6.94.

(2-Bromoethyl) ethyl bromophosphate (XV) was prepared analogously to amidophosphate **II** from 0.27 g of phospholane **XIV** and 0.32 g of bromine. R_f 0.83 (A), 0.63 (B), 0.86 (C). ^{1}H NMR spectrum, δ, ppm: 1.39 t (3H, $POCH_2CH_3$), 3.54 t (2H, CH_2Br), 4.15–4.50 m (4H, CH_2OPOCH_2). ^{31}P NMR spectrum, δ_P, ppm: –7.67 s.

1,2-*O*-Isopropylideneglycero-3-[(2-bromoethyl) ethyl phosphate] (XVI) was obtained analogously to glyceroamidophosphate **VIII** from bromide **XV** (prepared from 0.27 g of phospholane **XIV** and 0.32 g of bromine), 0.26 g of 1,2-*O*-isopropylideneglycerol (**VII**), and 0.2 g of triethylamine in 10 h. Yield 0.57 g (82%), n_D^{20} 1.4490, R_f 0.63(A), 0.43(B), 0.91(C). ^{1}H NMR spectrum, δ, ppm: 1.31 t (3H, OCH_2CH_3, $^3J_{HH}$ 7.04 Hz), 1.32 s (3H), 1.39 s (3H) [$C(CH_3)_3$], 3.51 t (2H, CH_2Br, $^3J_{HH}$ 6.24 Hz), 3.79 m (1H), 4.04 m (1H) (CH_2CHCH_2OP), 4.09 m (2H, CH_2CHCH_2OP), 4.31 m (3H, $CH_2CHCH_2OPOCH_2$). ^{31}P NMR spectrum, δ_P, ppm: –1.43 br.s. Found, %: C 34.42; H 5.76; P 8.88. $C_{10}H_{20}BrO_6P$. Calculated, %: C 34.59; H 5.81; P 8.92.

1,2-*O*-Isopropylideneglycero-3-[ethyltrimethylammonium [2-(dimethylammonio)ethyl]amidophosphate] bromide (XVII) was prepared analogously to bromomethylate **XII** from 0.45 g of amidophosphate **XVI** and 0.77 g of trimethylamine. Yield 0.44 g (73%). R_f 0.27 (C), 0.60 (D). ^{1}H NMR spectrum, δ, ppm: 1.29 s (3H), 1.36 s (3H) [$C(CH_3)_2$], 1.36 t (3H, $N^+CH_2CH_3$, $^3J_{HH}$ 7.5 Hz), 2.98 s [9H, $CH_3CH_2N^+$·$(CH_3)_3$], 3.44 s [9H, $OCH_2CH_2N^+(CH_3)_3$], 3.51 m (2H, $OCH_2CH_2N^+$), 3.69 m (2H, $N^+CH_2CH_3$), 3.81 m (1H), 3.90 m (1H) (CH_2CHCH_2OP), 4.03 m (2H, CH_2CHCH_2OP), 4.26 (2H, $POCH_2CH_2$, $^3J_{PH}$ 6.4 Hz), 4.49 m (1H, CH_2CHCH_2OP), 4.26 m (2H, $POCH_2$·CH_2, $^3J_{PH}$ 6.4 Hz), 4.49 m (1H, CH_2CHCH_2OP). ^{31}P NMR spectrum, δ_P, ppm: –0.6 br.s. Found, %: C 41.07; H 8.20; P 6.57. $C_{16}H_{38}BrN_2O_6P$. Calculated, %: C 41.29; H 8.23; P 6.66.

1,2-*O*-Isopropylideneglycero-3-phosphocholine (XVIII). Binary salt **XVII**, 0.3 g, was dissolved in 1 ml of a 9 : 1 methanol–water system, and the solution was applied to a silica gel column (10 g) filled with chloroform and eluted with 250 ml of a 5 : 1 chloroform–methanol mixture. The solvent was removed in a vacuum, and the residue was kept in a vacuum (10^{-4} mm) for 3 h at 90°C to give the anhydrous salt [15]. Yield 0.16 g (83%), mp 172–173°C (partial melting at 165–166°C) {published data [15]: 173–175°C (partial melting at 165–167 °C) [15]}. R_f 0.34 (C), 0.67 (D). ^{1}H NMR spectrum, δ, ppm: 1.31 s

(3H), 1.45 s (3H) {C(CH$_3$)$_2$], 3.51 s [9H, N$^+$(CH$_3$)$_3$], 3.76 m (1H), 3.92 m (1H) (C*H*$_2$CHCH$_2$OP), 3.98 m (2H, CH$_2$CHC*H*$_2$OPOC*H*$_2$), 4.26 m (1H, CH$_2$C*H*·CH$_2$OP). ^{31}P NMR spectrum, δ_P, ppm: –0.2 br.s. Found, %: C 44.27; H 7.99; P 10.32. $C_{11}H_{24}NO_6P$. Calculated, %: C 44.44; H 8.14; P 10.42.

2-Bromoethyl (diethylamido)bromophosphate (XXI) was prepared analogously to amidophosphate **II** from 0.33 g of phospholane **XIX** and 0.32 g of bromine and isolated pure by vacuum distillation (10^{-4} mm) at a bath temperature of 58–60°C. Yield 0.5 g (77%), n_D^{20} 1.4965. R_f 0.79 (A), 0.84 (B), 0.86 (C). ^{1}H NMR spectrum, δ, ppm: 1.15 t [6H, N(CH$_2$C·*H*$_3$)$_2$, $^3J_{HH}$ 7.12 Hz], 3.13 ppm [4H, N(C*H*$_2$)CH$_3$)$_2$], 3.54 m (2H, CH$_2$Br, $^3J_{HH}$ 6.11 Hz), 4.37 m (2H, OCH$_2$). ^{31}P NMR spectrum, δ_P, ppm: 5.1 s. Found, %: C 22.19; H 4.33; P 9.53. $C_6H_{14}Br_2NO_2P$. Calculated, %: C 22.31; H 4.37; P 9.59.

3-Bromoethyl (diethylamido)bromophosphate (XXII) was prepared analogously to amidophosphate **II** from 0.35 g of phosphorinane **XX** and 0.32 g of bromine and isolated pure by vacuum distillation (10^{-4} mm) at a bath temperature of 66–68°C. Yield 0.47 g (70%), n_D^{20} 1.4992, R_f 0.57 (A), 0.55 (B), 0.93 (C). ^{1}H NMR spectrum, δ, ppm: 1.20 t (6H, N(CH$_2$·C*H*$_3$)$_2$), $^3J_{HH}$ 7.26 Hz], 2.29 m (2H, C*H*$_2$CH$_2$Br), 3.17 m [4H, N(C*H*$_2$CH$_3$)$_2$, $^3J_{PH}$ 13.23 Hz], 3.50 t (2H, CH$_2$Br, $^3J_{HH}$ 6.4 Hz), 4.30 m (2H, OCH$_2$, $^3J_{PH}$ 5.12 Hz). ^{31}P NMR spectrum, δ_P, ppm: 5.6 s. Found, %: C 24.81; H 4.70; P 9.22. $C_7H_{16}Br_2NO_2P$. Calculated, %: C 24.95; H 4.79; P 9.19.

1,2-*O*-Isopropylideneglycero-3-[(2-bromoethyl) (diethylamido)phosphate] (XXIII) was prepared analogously to glyceroamidophosphate **VIII** from 0.65 g of bromide **V**, 0.26 g of 1,2-*O*-isopropylideneglycerol (**VII**), and 0.2 g of triethylamine in 24 h. Yield 0.65 g (87%), n_D^{20} 1.4584. R_f 0.42(A), 0.27(B), 0.76(C). ^{1}H NMR spectrum, δ, ppm: 1.09 t [6H, N(CH$_2$C*H*$_3$)$_2$, $^3J_{HH}$ 7.09 Hz], 1.32 s (3H), 1.39 s (3H) [C(CH$_3$)$_2$], 3.06 m [4H, N(C*H*$_2$CH$_3$)$_2$], 3.50 t (2H, CH$_2$Br, $^3J_{HH}$ 6.20 Hz), 3.79 m (1H), 3.92 m (1H) (C*H*$_2$CHCH$_2$OP), 3.98 m (2H, CH$_2$CHC*H*$_2$OP), 4.18 m (2H, POC*H*$_2$CH$_2$Br), 4.28 m (1H, CH$_2$C*H*·CH$_2$OP). ^{31}P NMR spectrum, δ_P, ppm: 10.3 br.s. Found, %: C 38.44; H 6.70; P 8.15. $C_{12}H_{25}BrNO_5P$. Calculated, %: C 38.51; H 6.73; P 8.28.

1,2-*O*-Isopropylideneglycero-3-[(3-bromopropyl) (diethylamido)phosphate] (XXIV) was obtained similarly to glyceroamidophosphate **VIII** from 0.67 g of bromide **V**, 0.26 g of 1,2-*O*-isopropylideneglycerol (**VII**), and 0.2 g of triethylamine in 24 h. Yield 0.5 g (64%), n_D^{20} 1.4660, R_f 0.45 (A), 0.49 (B), 0.94 (C). ^{1}H NMR spectrum, δ, ppm: 1.11 t [6H, N(CH$_2$C*H*$_3$)$_2$, $^3J_{HH}$ 7.25 Hz], 1.37 s (3H), 1.44 s (3H) [C)CH$_3$)$_2$], 2.20 m (2H, C*H*$_2$CH$_2$Br), 3.08 m [4H, N(C*H*$_2$CH$_3$0$_2$], 3.50 t (2H, CH$_2$Br), 3.59 m (1H), 3.73 m (1H) (C*H*$_2$·CHCH$_2$OP), 3.77 m (2H, CH$_2$CHC*H*$_2$OP), 4.04 m (2H, POC*H*$_2$CH$_2$Br), 4.22 m (1H, CH$_2$C*H*CH$_2$OP). ^{31}P NMR spectrum, δ_P, ppm: 10.7 br.s. Found, %: C 40.40; H 6.93; P 7.85. $C_{13}H_{27}BrNO_5P$. Calculated, %: C 40.22; H 7.01; P 7.98.

1,2-*O*-Isopropylideneglycero-3-[[2-(dimethylammonio)ethyl] (diethylmido)phosphate] bromide (XXV) was prepared analogously to bromomethylate **XII** from 0.49 g of amidophosphate **XXIII** and 0.77 g of trimethylamine. Yield 0.47 g (83%), R_f 0.23 (C), 0.63 (D). ^{1}H NMR spectrum, δ, ppm: 1.09 t [6H, N(CH$_2$C*H*$_3$)$_2$, $^3J_{HH}$ 6.92 Hz], 1.33 s (3H), 1.40 s (3H) [C(CH$_3$)$_2$], 3.01 m [4H, N(C*H*$_2$CH$_3$)$_2$], 3.54 s [9H, N$^+$(CH$_3$)$_3$], 3.74 m [2H, C*H*$_2$N$^+$(CH$_3$)$_3$], 3.80–4.30 m (6H, C*H*$_2$CHC*H*$_2$OPOC*H*$_2$), 4.41 m (1H, CH$_2$C*H*·CH$_2$OP). ^{31}P NMR spectrum, δ_P, ppm: 10.8 br.s. Found, %: C 41.42; H 7.89; P 7.12. $C_{15}N_{34}BrN_2O_5P$. Calculated, %: C 41.57; H 7.91; P 7.15.

1,2-*O*-Isopropylideneglycero-3-[[3-(dimethylammonio)propyl](diethylmido)phosphate] bromide (XXVI) was prepared similarly to bromomethylate **XII** from 0.5 g of amidophosphate **XXIV** and 0.77 g of trimethylamine. Yield 0.5 g (86%). R_f 0.28 (C), 0.60 (D). ^{1}H NMR spectrum, δ, ppm: 1.08 t [6H, N(CH$_2$C*H*$_3$)$_2$, $^3J_{HH}$ 6.83 Hz], 1.32 s (3H), 1.39 s (3H) [C(CH$_3$0$_2$], 2.25 m (2H, POCH$_2$C*H*$_2$), 3.04 m [4H, N(C*H*$_2$CH$_3$)$_3$], 3.47 s [9H, N$^+$(CH$_3$)$_3$], 3.68 m [2H, C*H*$_2$N$^+$(CH$_3$)$_3$], 3.74 m (1H), 3.87 m (1H) (C*H*$_2$·CHCH$_2$OP), 3.87 m (2H, CH$_2$CHC*H*$_2$OP), 4.06 m (2H, POC*H*$_2$CH$_2$), 4.30 m (1H, CH$_2$C*H*CH$_2$OP). ^{31}P NMR spectrum, δ_P, ppm: 11.0 br.s. Found, %: C 42.82; H 8.07; P 6.85. $C_{16}H_{36}BrN_2O_5P$. Calculated, %: C 42.96; H 8.11; P 6.92.

REFERENCES

1. Evstigneeva, R.P., Zvonkova, E.N., Serebrennikova, G.A., and Shvets, V.I., *Khimiya lipidov* (Lipid Chemistry), Moscow: Khimiya, 1983.
2. Paltauf, F. and Hermetter, A., *Prog. Lipid Res.*, 1994, vol. 93, no. 3, pp. 239–328.
3. Nifant'ev, E.E. and Predvoditelev, D.A., *Usp. Khim.*, 1994, vol. 63, no. 1, pp. 73–92.
4. Predvoditelev, D.A. and Nifant'ev, E.E., *Zh. Obshch. Khim.*, 1995, vol. 31, no. 12, pp. 1761–1785.
5. Malenkovskaya, M.A., Predvoditelev, D.A., and Nifant'ev, E.E., *Zh. Obshch. Khim.*, 1997, vol. 67, no. 6, pp. 944–947.
6. Nifant'ev, E.E., Predvoditelev, D.A., Malenkov-

skaya, M.A., and Suvorkin, S.V., *Zh. Obshch. Khim.*, 1998, vol. 68, no. 2, pp. 257–263.

7. Nifant'ev, E.E., Predvoditelev, D.A., Suvorkin, S.V., Malenkovskaya, M.A., and Bel'skii V.K., *Zh. Obshch. Khim.*, 1999, vol. 69, no. 3, pp. 383–389.
8. Chen, H., Chen, R., and Li, P., *Phosphorus, Sulfur Silicon*, 1997, vol. 122, pp. 1–7.
9. Marfin, S.F., Josey, J.A., Wong, Y., and Dean, D.W., *J. Org. Chem.*, 1994, vol. 59, no. 12, pp. 4805–4820.
10. Marfin, S.F., Wong, Y., and Wagman, A.S., *J. Org. Chem.*, 1994, vol. 59, no. 12, pp. 4821–4831.
11. Erukulia, R.K., Byun, H., and Bittman, R., *Tetrahedron*, 1994, vol. 35, no. 32, pp. 5783–5784.
12. Erukulia, R.K., Byun, H., and Bittman, R., *J. Org. Chem.*, 1995, vol. 60, no. 23, pp. 7706–7708.
13. Byun, H. and Bittman, R., *J. Org. Chem.*, 1996, vol. 61, no. 24, pp. 8706–8708.
14. Nifant'ev, E.E., Predvoditelev, D.A., Grachev, M.K., and Shin V.A., *Dokl. Akad. Nauk SSSR*, 1977, vol. 235, no. 3, pp. 595–598.
15. Nifant'ev, E.E., Predvoditelev, D.A., and Alarkon, Kh.Kh., *Zh. Org. Khim.*, 1978, vol. 14, no. 1, pp. 67–71.
16. Mukayama, T. and Kodaira, Y., *Bull. Chem. Soc. Jpn.*, 1966, vol. 39, no. 6, pp. 1297–1301.
17. Cason, J., Baxter, W.N., and DeAcetis, W., *J. Org. Chim.*, 1959, vol. 24, no. 1, p. 247.
18. Lucas, H.J., Mitchel, F.N., and Scully, C.N., *J. Am. Chem. Soc.*, 1950, vol. 72, no. 2, p. 549.