New synthetic phosphinate analogues of lecithin

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ABSTRACT  The chemical syntheses of two new, completely nonhydrolyzable phosphinate analogues of lecithin are described. These have the structures ROCH₂CH(OR')CH₂P(O)(O⁻)CH₂CH₂N(CH₃)₃ and ROCH₂CH(OR')(OR')CH₂P(O)⁻(O⁻)CH₂CH₂CH₃N(CH₃)₃, where R = C₁₈H₃₇ and R' = C₁₄H₂₉; each is thus isosteric with lecithin on either side of the phosphorus function. The infrared spectra of these compounds undergo unexpected changes under mild acid, base, or adsorptive treatment. These are discussed and compared with related lecithin analogues, including the simple phosphinate Cl₇H₁₇P(O)(O⁻)CH₂CH₂N(CH₃)₃, whose synthesis is also reported.

SUPPLEMENTARY KEY WORDS  isosteric  nonhydrolyzable  ethers  infrared spectra

THE PHOSPHINIC ACID (C—P—C) group contains two carbon–phosphorus bonds, and therefore a lecithin analogue containing this moiety would contain no phosphorus ester groups. Since in the absence of special structural features the two carbon–phosphorus bonds of a phosphinic acid are hydrolytically stable, a phosphinate-containing lecithin analogue can also be expected not to contain hydrolyzable phosphorus functions.

No examples of phospholipids containing the phosphinic acid group have as yet been isolated from natural sources. The chemical synthesis of one such lecithin analogue has, however, recently been reported from this laboratory (1, 2). This substance, a 2,3-dialkoxypropyl-[2'- (trimethylammonium)ethyl]phosphinate (I), is a compound in which ether groups substitute for the lecithin ester groups and which thus contains no hydrolyzable groups at any position. Compared with natural phosphate lecithin, however, this phosphinate analogue contains one less atom between phosphorus and nitrogen and between phosphorus and the long-chain functional groups.

The two phosphate-containing lecithin analogues whose synthesis is reported below represent structures isosteric (—CH₂— in place of —O—) with the natural lecithins on either side of the phosphorus atom. 3,4-Dioctadecyloxybutyl[2'- (trimethylammonium)ethyl]phosphinate (II) is isosteric at the "glycerol" chain, while 2-hexadecyloxy-3-octadecyloxypropyl[3'- (trimethylammonium)propyl]phosphinate (III) shows a similar structural feature at the nitrogenous base portion of the molecule.

PHOSPHINATE ANALOGS OF LECITHIN

Abbreviations: TLC, thin-layer chromatography.
2-Hexadecoxy-3-octadecoxypropyl[3'-(trimethylammonium)-propyl]phosphinate

Octadecyl[2'-(trimethylammonium)ethyl]phosphinate

The synthesis of the analogue II was very similar to that of the previously reported 2-hexadecoxy-3-octadecoxypropyl[2'-(trimethylammonium)ethyl]phosphinate (1, 2), while the synthesis of the 3-carbon base analogue III followed a quite different route, starting from 2-octadecoxy-3-octadecoxypropyl(allyl)phosphinate. The synthetic schemes are outlined below.

Both diether-phosphinate lecithin analogues proved to be hygroscopic (though not deliquescent) and each appeared to form several hydrates, so that analyses for the hydrates of the compositions reported could be obtained only by careful drying under specified conditions. This hydrophilicity was also shown by the much greater ease with which these analogues formed almost clear dispersions by sonication in water compared to the corresponding phosphonate and phosphate analogues.
The infrared spectra of these and similar compounds showed a number of unexpected transformations when they were exposed to mild acidic, basic, or adsorptive conditions.

MATERIALS AND METHODS

2-Hexadecoxy-3-octadecoxypropylphosphorylcholine, mp 193.5-194.5°C (dec) (3), and 2-hexadecoxy-3-octadecoxypropyl[2'-(trimethylammonium)ethyl]phosphinate, mp 202-203°C (dec) (1, 2), were synthesized as previously described. These substances were used only for comparison of their infrared spectra (see below).

1,2-O-Dihexadecylglycerophosphorylcholine was obtained from Schwarz/Mann, Orangeburg, N.Y. Infrared spectra were obtained on KBr pellets or chloroform solutions, using a Perkin-Elmer 337 infrared spectrometer. Elemental microanalyses were performed by Schwarzkopf Microanalytical Laboratory, Woodside, N.Y. Thin-layer chromatography in all cases was performed on 5 × 20 cm Silica Gel G-coated plates, using unlined cylindrical tanks.

Synthetic Route

The syntheses of the phosphinate analogues II and III are outlined in the scheme above. It should be noted that both 2- and 3-carbon base analogues were prepared via the versatile allyl intermediates. In the first case, a 3,4-dialkoxybutyl bromide (4) on reaction with diisopropyl allylphosphonite gave the isopropyl 3,4-dialkoxybutyl(allyl)phosphinate, the allyl group of which was oxidatively cleaved by osmate-periodate. Borohydride reduction, mesylation, and reaction with aqueous diisopropyl allylphosphonite, which was quaternized with methyl iodide. After conversion of the iodide salt to the acetate to avoid the formation of hydriodic acid during acid hydrolysis, reaction with hydrochloric acid removed the isopropyl ester group to give the lecithin analogue II as a sesquihydrate.

For the synthesis of analogue III, hydroboration of isopropyl 2-octadecoxy-3-octadecoxypropyl(allyl)phosphinate with di-sec-isoamylborane, then oxidation with hydrogen peroxide in a homogeneous buffered solution, gave the 3'-hydroxypropyl phosphinate. Mesylation, reaction with trimethylamine, and, finally, acid hydrolysis gave the lecithin analogue III as the monohydrate.

EXPERIMENTAL

1. DL-3,4-Dioctadecoxybutyl[2'-(trimethylammonium)-ethyl]phosphinate (II)

DL-Isopropyl 3,4-Dioctadecoxybutyl(allyl)phosphinate. 3,4-Dioctadecoxybutyl bromide (10.0 g, 0.015 mole [4]) and freshly distilled diisopropyl allylphosphonite (5 g, 0.035 mole [5]) containing a trace of hydroquinone were heated under nitrogen at 120 ± 2°C for 40 hr. The cooled mixture in ether was washed with dilute HCl and water. It was then dried over MgSO₄, filtered, and the solvent was evaporated. To the residue was added acetonitrile at 10°C, and the precipitated product was filtered, washed with cold acetonitrile, and dried in vacuo. The yield of crude product was 9.8 g.

Crude isopropyl 3,4-dioctadecoxybutyl(allyl)phosphinate (9.0 g) in 100 ml of methylene chloride was applied to a column (50 mm I.D.) containing Mallinckrodt Silica AR CC-7 (450 g), previously activated by heating at 110°C for 16 hr and washing with 1 liter of methanol and 2.5 liters of methylene chloride. The column was eluted with methylene chloride (1 liter) and 1 liter each of 3%, 5%, and 6% ethanol in methylene chloride, all of which eluted only low-polarity impurities. The product was eluted with 8 and 10% ethanol in methylene chloride (1 liter each), successively. The product obtained by evaporation of the solvent weighed 5.5 g (61% yield). When chromatographed by TLC there were two essentially identical spots, Rₚ 0.47-0.49, when the Silica Gel G plates were developed in 8% ethanol in methylene chloride. Recrystallization of the product from chloroform–acetone and from absolute ethanol at 5°C yielded an analytically pure product, mp 35-36°C, presumably an approximately equal mixture of the two racemic diastereoisomers.

Analysis: C₄₆H₉₃O₄P (mol wt 741.22);
found: C, 74.24; H, 12.37; P, 3.89

The infrared spectrum showed a rather weak but sharp C–=O band at 1625 cm⁻¹ in addition to the P–O–C(955 cm⁻¹) absorptions.

2. DL-Isopropyl 3,4-Dioctadecoxybutyl(2'-acetaldho)phosphinate. The intermediate aldehyde, alcohol, mesylate, dimethylaminoethyl, and (trimethylammonium)ethyl isopropyl phosphonate esters were not isolated in analytically pure form before conversion to the phosphinate lecithin.

2 g of sodium metaperiodate in 43 ml of water and 500 ml of 95% ethanol were added to 1 g (0.00135 mole) of the purified allylphosphonite in 117 ml of absolute ethanol. While the mixture was stirred vigorously, a freshly prepared solution of osmium tetroxide (60 mg) in absolute ethanol (50 ml) was added. After stirring 2 hr at room temperature the mixture was evaporated in vacuo at 35°C to about 50 ml. The residue was extracted with chloroform and water, and after the chloroform layer was dried with MgSO₄ the solvent was removed in vacuo. The residue, which consisted of several spots on...
thin-layer plates, weighed 0.99 g and showed a strong, sharp aldehyde absorption at 1710 cm⁻¹.

**DL-Isopropyl 3,4-Diiodooctadecylbutyl(2'-hydroxyethyl)phosphinate.** Reduction of the aldehyde was accomplished by addition of sodium borohydride (450 mg) to a solution of the crude aldehyde (0.99 g) in absolute ethanol (65 ml); the mixture was stirred at room temperature for 16 hr. It was cooled to 5°C and 12 N HCl was added dropwise until no hydrogen was evolved. The mixture was evaporated to dryness in vacuo at 50°C. The crude alcohol (0.97 g) showed only a very slight absorption in the 1700 cm⁻¹ region.

The entire product in 20 ml of chloroform was applied to a column (20 mm i.d.) containing 100 g of SilicAR CC-7 (previously activated at 110°C and then washed with methanol and chloroform). The product was eluted in several fractions with 1 and 2% methanol in chloroform. The main fraction (0.70 g) was homogeneous (Rₚ 0.27) as judged by TLC in chloroform-methanol-58% aqueous ammonia 95:4:0.4.

**DL-Isopropyl 3,4-Diiodooctadecylbutyl(2'-mesyloxyethyl)-phosphinate.** The crude alcohol (0.70 g) in dry pyridine (12 ml) was cooled to 5°C while methanesulfonyl chloride (1.2 ml) was added dropwise during 5 min of vigorous stirring. After stirring at 5°C for 10 min more and at room temperature for 20 min, the mixture was extracted with ether (100 ml) and water (100 ml). The aqueous layer was reextracted with 2 × 50 ml of ether. The combined ether extracts were washed successively with 2 × 50 ml of water, 1 × 50 ml of 2 n H₂SO₄, 2 × 50 ml of water, 1 × 50 ml of 3 M Na₂CO₃, and finally with 1 × 100 ml of water. The ether solution was dried over MgSO₄, filtered, and evaporated in vacuo to give 0.75 g of the crude product. There was one main spot (Rₚ 0.79) by TLC (8% ethanol in methylene chloride). Bands at 1345 and 1163 cm⁻¹, which are characteristic of sulfonate esters, were observed.

**DL-Isopropyl 3,4-Diiodooctadecylbutyl(2'-trimethylammonium)ethylphosphinate Iodide.** A solution of di-sec-isooamyloborane was prepared by adding 12.0 g (0.17 mole) of 2-methyl-2-butenone to 100 ml of 0.85 M borane in tetrahydrofuran (1 M solution from Ventron Corp.) during 30 min of stirring under nitrogen at 18°C.

A solution of di-tert-isooamyloborane was prepared by adding 12.0 g (0.17 mole) of 2-methyl-2-butenone to 100 ml of 0.85 M borane in tetrahydrofuran (1 M solution from Ventron Corp.) during 30 min of stirring under nitrogen at 18°C.

To isopropyl 2-hexadecoxy-3-octadecoxypropyl(3'-hydroxypropyl)phosphinate. A solution of di-tert-isooamyloborane was prepared by adding 12.0 g (0.17 mole) of 2-methyl-2-butenone to 100 ml of 0.85 M borane in tetrahydrofuran (1 M solution from Ventron Corp.) during 30 min of stirring under nitrogen at 18°C.
this was slowly added an organic buffer prepared by dissolving 2.37 g of boric acid in a mixture of 38 ml of 25% methanolic tetrabutylammonium hydroxide and 190 ml of tetrahydrofuran (evolution of hydrogen). The mixture was cooled to 5°C, 15% aqueous hydrogen peroxide (50 ml) was added dropwise with stirring during 1 hr, and the mixture was stirred for 30 min more at room temperature. It was evaporated in vacuo at 50°C to half its volume, poured into a mixture of 2 liters of ice-water and 80 ml of 12 N HCl, and then kept overnight at room temperature. The mixture was filtered and the precipitate was washed thoroughly with water.

The wet material was dissolved in chloroform and washed with water; the chloroform layer was dried over MgSO4, filtered, and evaporated in vacuo at 45°C. The crude alcohol (5.6 g) in chloroform (50 ml) was applied to a column 50 mm in diameter containing Mallinckrodt SilicAR CC-7 (activated at 105°C and by washing with methanol and then with chloroform). Chloroform (1 liter) and 2% absolute ethanol in methylene chloride eluted low-polarity impurities. The product was eluted with 4% and then 6% absolute ethanol in methylene chloride eluted homogeneous impurities. The product was eluted with 4% and then 6% absolute ethanol in methylene chloride eluted low-polarity impurities. The product was eluted with 4% and then 6% absolute ethanol in methylene chloride eluted low-polarity impurities. The product was eluted with 4% and then 6% absolute ethanol in methylene chloride. The product was eluted with 4% and then 6% absolute ethanol in methylene chloride eluted low-polarity impurities. The product was eluted with 4% and then 6% absolute ethanol in methylene chloride eluted low-polarity impurities. The product was eluted with 4% and then 6% absolute ethanol in methylene chloride eluted low-polarity impurities. The product was eluted with 4% and then 6% absolute ethanol in methylene chloride. The product was eluted with 4% and then 6% absolute ethanol in methylene chloride eluted low-polarity impurities. The product was eluted with 4% and then 6% absolute ethanol in methylene chloride. The product was eluted with 4% and then 6% absolute ethanol in methylene chloride eluted low-polarity impurities. The product was eluted with 4% and then 6% absolute ethanol in methylene chloride.

The methanesulfonate (1.28 g) was dissolved in 40 ml of methanolic trimethylamine and the solution was crystallized twice from acetone yielded a pure white product (1.1 g) which proved difficult to analyze because of its hygroscopicity. Redrying at 50°C just prior to analysis gave values correct for a monohydrate.

Analysis: C17H19NO7PS + H2O (mol wt 872.350)
- calculated: C, 64.71; H, 11.79; N, 1.60; S, 3.67
- found: C, 65.06; H, 11.73; N, 1.83; S, 3.75

If the compound was dried at 150°C in a nitrogen atmosphere just prior to analysis, the anhydrous material was formed.

Analysis: C17H18NO7PS (mol wt 854.335)
- calculated: C, 66.08; H, 11.80; N, 1.64; S, 3.75
- found: C, 65.91; H, 11.87; N, 1.83; S, 3.76

The sulfonic ester regions at 1175 and 1345 cm^-1 showed little absorption. After successive drying at 40°C, 60°C, 100°C, and 120°C in vacuo, the salt melted at 156-156.5°C.

DL-2-Hexadecoxy-3-octadecoxypargyl(3′-trimethylammonium)propylphosphinate (II). The methanesulfonate salt (800 mg, 0.917 mmole) was dissolved in a warm mixture of acetic acid (120 ml) and 6 N hydrochloric acid (12 ml), and the solution was kept at 80-85°C for 24 hr. Evaporation of the mixture to dryness at 50°C followed by removal of residual acid (in vacuo < 1 mm) for 20 min gave a white product. The material was dissolved in 2 ml of chloroform; 0.5 ml of pyridine was added, and the product was precipitated with cold acetonitrile. The precipitate was filtered and washed with cold acetonitrile and acetone and dried in vacuo. The product at this point weighed 700 mg (44% yield from the allylphosphinate); it was almost homogeneous (Rf 0.32, chloroform-methanol-water 65:25:4).

The material dissolved in warm ethanol-water-chloroform 7:3:1 was passed through a column (1 cm I.D.) of 20 g of 1:1 mixture of Amberlite 1R-120(H+) and 1R-45 (OH-) at 40-45°C. The column was washed thoroughly with the same solvent, and the combined eluates were evaporated in vacuo at 45°C. The residue was dehydrated by reevaporation three times with isopropanol. The product was recrystallized from boiling chloroform-acetone; the yield at this point was 510 mg, or 32% over-all yield from the allylphosphinate. The compound melted sharply with decomposition at 192-194°C. The material was very difficult to dry adequately for analysis.

Analysis: C27H40NO4P + H2O (mol wt 734.18)
- calculated: C, 70.35; H, 12.63; N, 1.91; P, 4.22
- found: C, 70.11; H, 12.83; N, 1.80; P, 3.91
The infrared spectrum is discussed below. The synthesis could also be accomplished if the initial hydroboronation of the allylphosphinate was performed with borane or 9-borabicyclo[3.3.1]nonane (6); in either case, however, the succeeding intermediates were much more difficult to purify than when distiamylborane was used.

3. Octadecyl[2'-[(trimethylammonium)ethyl]-phosphinate (IV)

The synthesis of this single long-chain hydrocarbon phosphinate analogue was very similar to that of the dioctadecoxybutyl analogue, except that none of the intermediates were purified extensively or characterized. Isopropyl octadecyl(allyl)phosphinate was prepared in 62% yield from 1-bromoocadecane and diisopropyl allylphosphonite according to the method used for the corresponding dioctadecoxybutyl compound (see above). The product, however, was somewhat soluble in acetonitrile at room temperature, so that precipitation was performed at -10°C. The material as isolated contained several components on TLC.

The crude allylphosphonate was oxidized by the periodate–osmate procedure as in the dioctadecoxybutyl series discussed above. The crude isopropyl octadecyl(2-acetaldo)phosphinate showed a very strong aldehyde band at 1710 cm⁻¹. Reduction as above with sodium borohydride yielded isopropyl octadecyl(2-hydroxyethyl)phosphinate, as evidenced by loss of the aldehyde absorption. The mesylate was similarly prepared as described above for the dioctadecoxybutyl series. It showed strong bands at 1345 and 1120 cm⁻¹, characteristic of sulfonate esters.

Treatment of the sulfonate ester with aqueous dimethylamine, washing the isolated crude product with base to free the tertiary amine, and allowing the latter to react with methyl iodide in the dark for several days as above gave isopropyl octadecyl[2'-[(trimethylammonium)ethyl]phosphinate iodide, isolable as a precipitate from cool ether at 10–15°C.

The methiodide was hydrolyzed directly, without conversion to the acetate salt, by hydrochloric acid in acetic acid solution at 80°C for 17 hr. The product was isolated as for the above two lecithin analogues, but with precipitation by acetonitrile at -15°C. The product in chloroform was purified by adsorption on SilicAR CC-7 (50 g per 150 mg of product) in a 20-mm (i.d.) column. Chloroform (200 ml), and 5, 15, and 25% methanol in chloroform (150 ml each), eluted impurities, which were present only in small quantities. 200 ml of methanol–chloroform 1:5 eluted a small quantity of an intermediate fraction containing both the desired product and impurities, and the pure product was eluted with 180 ml of chloroform–methanol–water 65:25:4. For analysis the product was recrystallized twice from chloroform–acetone.

Before chromatography the yield of product was 30% over-all from the allylphosphinate; about 25% of the applied material was lost during the chromatographic purification. The pure product melted at 212–213°C with decomposition. Although the product appeared to be hygroscopic, careful drying at 100°C in a nitrogen stream and in vacuo gave a correct analysis for an anhydrous product.

Analysis: C₃₅H₆₁NO₅P (mol wt 403.63);
calculated: C, 68.44; H, 12.49; N, 3.47; P, 7.62
found: C, 68.51; H, 12.49; N, 3.61; P, 7.35

The infrared spectrum of this substance is discussed below.

**Infrared Spectra of Lecithin Analogues**

A curious and unexpected phenomenon was observed (2) in the infrared spectrum of the previously reported phosphinate lecithin analogue, 2-hexadecoxy-3-octadecoxypropyl[2'-[(trimethylammonium)ethyl]phosphinate. As ordinarily obtained after acid hydrolysis, the compound does not possess any strong absorptions around 1000–1050 cm⁻¹, and the P → O absorption centered around 1280 cm⁻¹ is rather broad and shallow and merges with the broader and much stronger absorption at 1190–1120 cm⁻¹. When this compound in any of a variety of solvents (chloroform–methanol mixtures, aqueous tetrahydrofuran, chloroform–ethanol–water, etc.) was adsorbed and then eluted from silicic acid, or passed through ion-exchange resins or even Sephadex LH-20, the reisolated compound showed dramatically different infrared absorptions in the regions noted. The strong absorption around 1100 cm⁻¹ became much weaker and was now bracketed by two much stronger and moderately sharp absorptions at 1040 and 1160 cm⁻¹.

We have now found that this change is more simply effected by warming the material in aqueous pyridine for a few hours or in tetrahydrofuran–aqueous ammonia for several hours at room temperature. Furthermore, it was freely reversible; by allowing material containing these new peaks to stand overnight in acetic acid containing a little hydrochloric acid, removing the acids in vacuo, adding pyridine, and reprecipitating with cold acetonitrile, the original material was obtained again.

The same transformation was observed for the dialkoxybutyl analogue II. The infrared spectrum (Fig. 1, A) of this compound was not altered after redissolving it in warm chloroform–pyridine 3:1, adding 5 volumes of cold acetonitrile, and reisolating the reprecipitated product by filtration. Nonetheless, the following treatment is an example of a procedure which effected the
transformation: The lecithin analogue II (unrecrystal-
ized; 125 mg) in 15 ml of methylene chloride-methanol
19:1 was applied to a 20-mm (i.d.) column containing
20 g of SilicAR CC-4 previously washed with the same
solvent. Methylene chloride–methanol 19:1 (40 ml) and
methylene chloride–absolute ethanol 9:1 (100 ml)
eluted only trace impurities. Chloroform–methanol–
water 65:25:4 (120 ml in 10-ml fractions) eluted, suc-
cessively, 14 mg of impurities, 14 mg of unchanged ana-
logue, and 90 mg of material of altered infrared spectrum
(Fig. 1, B).

Alternative procedures for producing the form of spec-
trum B, Fig. 1, were aqueous ammonia (30°C for 3 hr
in tetrahydrofuran–58% ammonia 9:1) and aqueous
pyridine (50°C overnight in tetrahydrofuran–pyridine–
water 5:3:2), as mentioned above. In either case the
product was isolated by evaporation of solvents in vacuo.
Passage of the spectrum A form of analogue II in chloro-
form–ethanol–water 1:7:3 through an equimolar mixture
of Amberlite 1R-120 (H+) and 1R-45 (free base),
followed by elution with the same solvent, evaporation
in vacuo, reevaporation with isopropanol, and drying
of the product gave the spectrum B form.

Spectra of this and the other analogues studied were
initially taken in KBr pellets. The essential spectral dif-
fences between two forms of the same analogue were,
however, preserved when the spectra were examined in
chloroform solution.

Approximately the same effects were seen in the case of
2-hexadecoxy-3-octadecoxypropyl[3′-(trimethylam-
monium)propyl]phosphinate (analogue III). The spec-
tra without and with peaks are seen in Fig. 2, C and D,
respectively. Some indication was observed that the
transformation may not be quite as facile with the 3-
carbon base analogue as with the above 2-carbon base
compounds, since fractions which were intermediate in
spectral characteristics between the two forms were ob-
tained from a silica column. Nevertheless, the trans-
formation between the two forms took place quite
readily.

The elemental analysis given above for this lecithin
analogue was obtained on the silica-treated form, which
showed peaks at 1045 and 1150 cm⁻¹. This compound,
on reconversion to the spectrum C form, nonetheless
had the same elemental composition.

Analysis: C₁₅H₃₉NO₄P + H₂O (mol wt 734.18);
calculated: C, 70.35; H, 12.63; N, 1.91; P, 4.22
found: C, 70.11; H, 12.83; N, 1.80; P, 3.91

Some differences in the melting points were observed;
form C melted with decomposition at 204–206°C, while
form D melted with decomposition at 192–194°C. The
TLC behavior of both was identical; the two forms
could not be separated from a mixture.

The possibility was considered that the ether groups
were in some way involved in these changes, since the
region around 1100 cm⁻¹, which shows considerable dif-
ference between the two forms, is in a range where ether
absorptions occur. For this reason the hydrocarbon

Fig. 1. A, spectrum of 3,4-dioctadecoxybutyl[2′-(trimethylammonium)ethyl]phosphinate (II) after acidic
treatment; B, spectrum after basic treatment. Conditions are given in text.
analogue octadecyl[2′-(trimethylammonium)ethyl]phosphate was synthesized. The changes in its infrared spectrum were studied after its isolation under the different conditions outlined above. Essentially the same spectral alterations were observed as with the ether-containing phosphate analogues. The two spectra of the hydrocarbon analogue are shown in Fig. 3, E and F.

A commercial diether phosphate lecithin, 2,3-dihexadecylglycerylphosphorylcholine, showed some variation in its absorption peaks at 970 and 1245 cm⁻¹ after the acid treatment; a sharper peak at 1265 cm⁻¹ appeared while the 970 cm⁻¹ absorption became much weaker. These changes, however, are much less pronounced than those seen in the phosphinate analogues.
Intermediate in structure between the phosphinate and phosphate analogues is the phosphonate analogue (2-hexadecyloxy-3-octadecyloxypropylphosphonylcholine (3). Its acid-induced spectral transformation was also more or less intermediate between the phosphinate and phosphate analogues. The strong absorption at 1210 cm⁻¹ virtually disappeared after acid treatment, while the complex medium-width absorption at 1060–1100 cm⁻¹ was replaced by a broader absorption in the same region with three distinct peaks at 1055, 1097, and 1125 cm⁻¹. The original sharp, strong peak at 970 cm⁻¹ became stronger but broader. As in the case of all the lecithin-type structures studied, these alterations were completely reversible.

Various explanations were considered for these transformations, which at least in the case of the phosphinates were remarkable in degree. The most obvious possibility was that the acid-treated compounds were simply the free phosphinic acid–chloride salt forms. This seems unlikely in view of the careful removal of excess hydrochloric acid at 20 mm and then below 1 mm, followed by addition of excess pyridine to a concentrated chloroform solution of the products before isolation by precipitation with acetone or acetonitrile. Moreover, the spectral shifts observed are not in accord with the changes seen in simple acid–salt interconversions of phospholipids, which have been studied in detail by Abramson, Norton, and Katzman (7). These authors stress that P–OH absorptions occur not only in the 2700 cm⁻¹ region but also at 1000–1040 cm⁻¹ and perhaps at 1250–1220 cm⁻¹. In the salt forms, the P–O⁻ absorption occurs at 1110–1090 cm⁻¹. In our compounds the single most remarkable change is the appearance of a strong, rather sharp band at about 1040 cm⁻¹, where little absorption was seen before, after treatment with weak bases (which should ionize any P–OH groups present) or adsorbents. In other words, the spectral shift in this region is just opposite to that which would be predicted if simple ionization were taking place. Differences between the two forms in the 2700 cm⁻¹ region, moreover, are very minor, while variations in the 1250–1220 cm⁻¹ region are not consistent from compound to compound (but in compound II are again opposite to the predicted variation).

We tentatively suggest that these forms represent different conformational structures, possibly involving different orientations of hydrogen-bonded hydration water. Conceivably, one form may be a quasi-cyclic structure analogous to the gauche form proposed by Sundaralingam (8) for phosphate lecithins, and the other may be a more or less “trans” or open-chain structure. Differences in hydrogen bonding is suggested because the greatest changes occur in the absorption region of P → O, whose position is known to be very susceptible to shifts produced by hydrogen bonding (9). Further physical studies are of course necessary to clarify these alterations.

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