mitted to the Bleaching Committee as soon as its reorganization is effected.

11. Editor of Analytical Methods. V. C. Mehlenbacher, who performed the exceedingly difficult task of revising our Analytical Methods in a highly creditable manner, and who has served for several years most efficiently as editor of our Methods has found it necessary to resign from that important post. He has earned well the appreciation and gratitude of the Society for these outstanding services. The Uniform Methods Committee, recognizing the need for a permanent editorship for Analytical Methods in order to achieve uniformity in style and format, recommended that the president appoint yearly an editor of Analytical Methods, the person selected to be approved by the Governing Board. This recommendation had the unanimous support of the Uniform Methods Committee and was approved by the Society. It has been offered to the President and Governing Board.

It is a pleasure to add that these conclusions were reached in a discussion in which all members of the Uniform Methods Committee participated and that their decision on every question was unanimous.

Some of our technical committees have made progress reports with no recommendations for changes or additions to analytical methods. Such reports should not be interpreted as inactivity on the part of those committees. For example, the Cellulose Yield Committee has reported cooperative analytical work of its usual high quality and is planning to continue with a minimum of four samples during the coming year. Other committees are engaged in investigations whose value will become apparent when their experiments have progressed to a point where they can be reported with clear significance. It is our belief that no professional society has more active technical committees, composed of more able members, and engaged in more useful scientific effort.

J. T. R. ANDREWS, chairman.

M. M. DURKEE	R. R. KING
J. J. GANUCHEAU	T. C. LAW
T. H. HOPPER	L. B. PARSONS

Comparative Composition of Soybean and Corn Phosphatides[1]

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SOYBEAN "lecithin" and corn "lecithin" are the only plant phosphatides of commerce. The two are processed in a similar manner; they are similar in many of their properties, and one might expect them to be similar in composition. However very little has been published on the composition of corn phosphatides (6). Because of reports of certain superior properties of corn phosphatides, it seemed desirable that a comparison should be made between the phosphatides obtained from corn oil and those obtained from soybean oil. For this purpose, a sample of corn phosphatides has been prepared and fractionated in a manner similar to that previously used for soybean phosphatides (7). This paper reports those results and compares them with those obtained for the soybean phosphatides.

Analytical Methods. The analytical procedures used in this work were similar to those previously described for soybean phosphatides. Total nitrogen was determined by the Micro-Kjeldahl procedure. Phosphorus determinations reported in Table I were obtained gravimetrically as ammonium phosphomolybdate. Owing to the smaller size of the samples obtained from countercurrent distribution (Figures 1, 2, 3), phosphorus in this instance was measured spectrophotometrically by a modification of Burmaster's method for total phosphorus (2). Choline was measured by the reineckate method of Glick (5). Sugar was determined after hydrolyzing the phosphatides for approximately seven hours with 0.6 N H_2SO_4 on a steam bath. The hydrolyzate was filtered and neutralized, and sugar was measured by the method of Somogyi (8). Sugar was calculated as galactose in order to be comparable with the previous analysis of soybean phosphatides although other sugars are known to be present (9).

Inositol was estimated by a modification of the Atkin, Schultz, Williams, and Frey (1) microbiological method for the assay of pyridoxin. Samples were hydrolyzed with 20% HCl at 120°C. for 16 hours prior to assay.

Preparation of Fractions. The crude phosphatides used in this work were prepared from material obtained in the commercial degumming of corn oil. The gums were collected at the centrifuges and refrigerated immediately thereafter. This material was dissolved in ether and ether insoluble substances allowed to settle out. After drying the solution with sodium sulfate, the ether was removed under vacuum. The residue was extracted repeatedly with acetone, first stirring by hand, and, after the product became waxy, with the aid of a Waring Blendor. Analytical data on this crude acetone insoluble phosphatide preparation are shown in Table I.

The crude phosphatide preparation was subsequently separated into alcohol-soluble and alcohol-insoluble fractions in the same manner previously used with soybean phosphatides. A 150-gram portion of the phosphatides was extracted with six successive 300-ml. portions of absolute alcohol in a Waring Blendor. The alcohol-soluble portion was further fractionated by removing the solvent under vacuum at 50°C. and by again adding absolute alcohol. The alcohol was evaporated from the clear supernatant solution leaving 38.8 g., which are designated in Table I as Fraction I. A small portion, 1.6 g., which did not redissolve in the alcohol, is designated as Fraction II. Since this fraction is quite small, no

[1] Presented at fall meeting of American Oil Chemists' Society, October 31, November 1 and 2, 1949, in Chicago, Illinois.

[2] One of the laboratories of the Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Department of Agriculture. Report of a study made under the Research and Marketing Act of 1946.

TABLE I

Analysis of Phosphatide Preparations

Preparation	Per cent of total weight	Nitrogen	Phosphorus	Choline nitrogen	Inositol	Sugar	Molar ratio	Molar ratio
		%	%	%	%	%	*P/N*	*P/Inositol*
Crude phosphatides	100	1.30	3.10	0.39	4.27	3.7	1.08	.....
Fraction I	35	1.90	2.93	.85	0.14	1.78	.70	.....
Fraction II	1	.....	.....	.....	.....	.....	.....	.....
Fraction III	9	0.98	2.49	.49	0.56	1.91	1.15	.....
Fraction IV	55	1.10	3.25	.098	8.0	5.61	1.34	2.37

analytical work was done on it. The alcohol-insoluble portion was extracted four times in the Blendor with absolute alcohol warmed to 50°. This alcohol solution was evaporated under vacuum at 50°, leaving 10.4 g. of residue designated as Fraction III. The alcohol-insoluble material remaining weighed 78.8 g. and is designated as Fraction IV. Analytical data on these fractions are also given in Table I.

Phosphatide fractions were protected from oxidation by keeping them under carbon dioxide as much as possible during their preparation and by storing them at —16°C. in a vacuum desiccator.

Countercurrent Distribution. In order to compare distribution patterns of the corn phosphatides with those of the corresponding fractions from soybeans, Fraction I and Fraction IV were further fractionated by countercurrent distribution. The Craig apparatus used was a large preparative model. Each tube has a capacity of 84 ml. for each solvent layer which permitted the use of a larger sample and at the same time a more dilute solution than was possible in work with the soybean phosphatides. This large apparatus has a glass plate at each end which facilitates the operation by permitting observation of the solvent layers (4). The solvent pairs employed were the same as those used previously, hexane and 90% methanol for the alcohol-soluble fraction and hexane and 95% methanol for the alcohol-insoluble fraction.

For the distribution of the alcohol-soluble fraction, 3.026 g. of Fraction I was dissolved in 70 ml. of saturated hexane. In order to minimize difficulties with emulsions and changes in the interface level, the sample was carried through the first three distributions in separatory funnels. The solvent layers were adjusted to 84 ml. and transferred to the Craig apparatus. The fractionation was continued until a total of 28 distributions were applied. After the distribution was completed, the solutions were removed from each tube. Measurement of the volume of the lower layers indicated that there had been no significant change in the interface level. The fractions were evaporated to dryness and left in a vacuum desiccator until constant weights were obtained. They were then dissolved in chloroform. Phosphorus, total nitrogen, choline nitrogen, and sugar were determined on aliquots. The weights of the fractions in milligrams and the analytical results in millimoles are shown plotted against tube number in Figure 1. Non-choline nitrogen, the difference between total nitrogen and choline nitrogen, is also shown in this figure.

The alcohol-insoluble fraction was distributed in a manner similar to that described for the alcohol soluble fraction, a 3.012-g. portion of Fraction IV being used. It was found necessary to wait from 15 to 30 minutes after each shaking to allow emulsions to break in tubes 1 and 2. The solvent layers in all other tubes separated rapidly. The fractionation was continued until a total of 24 distributions were applied. After the fractions were dried to constant weight and dissolved in chloroform, phosphorus, total nitrogen, inositol, and sugar were determined on aliquots. The weights of the fractions in milligrams and the analytical results in millimoles are shown plotted against tube number in Figures 2 and 3.

Discussion. The lecithin from corn phosphatides, like that from soybean, is found almost entirely in the alcohol-soluble fractions as is shown by the choline nitrogen figures of Table I. Also similarly to the soybean phosphatides, the alcohol soluble fractions contain non-choline nitrogen. However the molar phosphorus to nitrogen ratio of Fraction I is only .70 in contrast to a ratio of nearly one for the corresponding fraction from soybean phosphatides.

In the countercurrent distribution of Fraction I the highest concentrations of both choline nitrogen and non-choline nitrogen are found in tube 0. Both fall off to the right, the non-choline nitrogen more rapidly than the choline nitrogen, especially in tubes 1 and 2. If it is assumed that all the choline occurs as a constituent of lecithin, then the lecithin in tube 0 accounts for 90% of the phosphorus in this tube. Thus there remains only one mole of phosphorus for every 17 moles of non-choline nitrogen. This leads to the conclusion that a nitrogen containing compound other than the phosphatides is present and that it is concentrated in tube 0 and adjacent tubes.

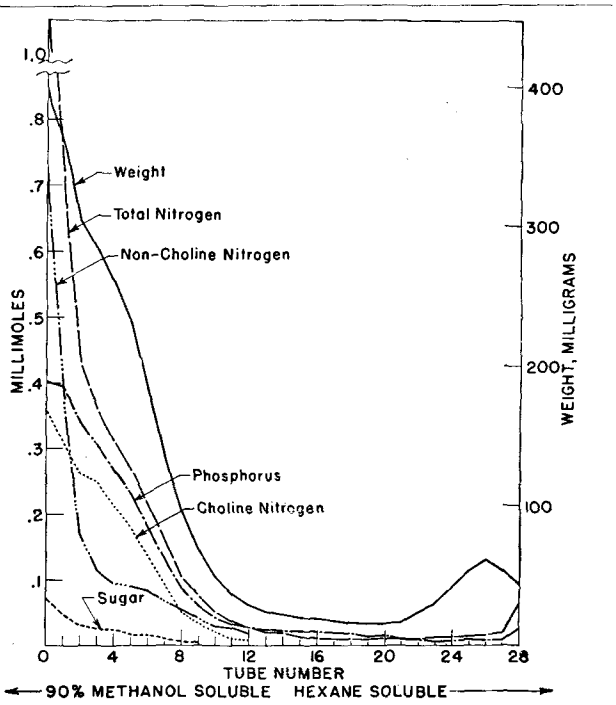

FIG. 1. Results of the countercurrent distribution of the alcohol-soluble fraction.

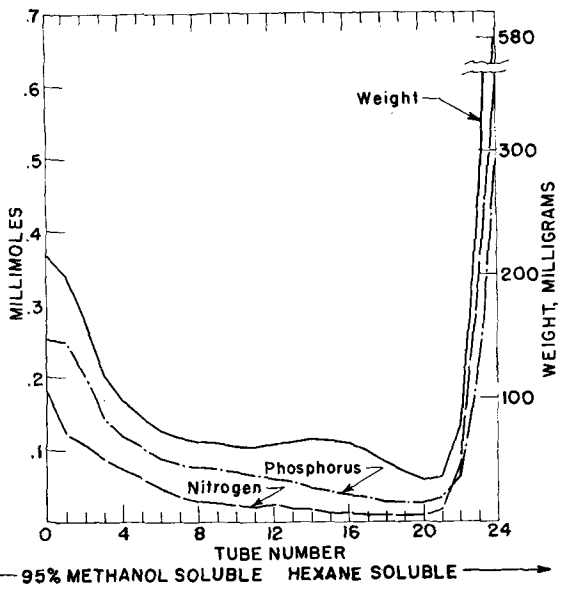

FIG. 2. Weight, nitrogen, and phosphorus results of the countercurrent distribution of the alcohol-insoluble fraction.

This conclusion is in agreement with the fact that ethanolamine nitrogen measured by the Burmaster (3) periodate oxidation method is much lower than non-choline nitrogen or than amino nitrogen measured by the Van Slyke method. In tubes 0 and 1 only about 8% of the non-choline nitrogen is accounted for by the Burmaster method; however in tubes 8 and 9 this figure is almost 30%. Further evidence of the small amount of ethanolamine is given by paper chromatograms of hydrolyzates of Fraction I, in which the ethanolamine spot was always very faint. Thus it appears that cephalin is present but in a much smaller amount than in soybean phosphatides.

Sugar is concentrated in the tubes at the left of the curve in a manner similar to the alcohol soluble soybean phosphatides. Even in tube 0 however there is only 3.2% sugar. Since the amount of sugar in all tubes on a molar basis is low, sugar can hardly be a component of a major phosphatide constituent.

The weight curve also has a small peak at tube 26 in the hexane soluble region. It is apparent from the curve the amounts of phosphorus and nitrogen in this and neighboring tubes are low. Tube 26 was also found to contain only 1.3% sugar. Because of the small amount of sample in these tubes the material was not further investigated.

Although the concentration of the sample in the tubes at the left edge of the curve would indicate a less satisfactory separation of the constituents than was the case with soybean phosphatides, interesting comparisons of the difference in composition between soybean and corn phosphatides are none the less possible. The alcohol-soluble fractions of corn phosphatides and soybean phosphatides are similar in that both contain lecithin and a small amount of sugar or sugar containing impurity. The corn phosphatide fraction contains a much smaller amount of cephalin than the corresponding soybean phosphatide fraction. The greatest difference seems to be the presence in the alcohol soluble corn phosphatides of a nitrogen containing compound in addition to the phosphatides. Upon partition between 95% methanol and hexane this compound is strongly concentrated in the methanol.

Inositol containing compounds are present in corn as well as in soybean phosphatides. In both cases the inositides are strongly concentrated in the alcohol insoluble fraction. In this fraction however the phosphorus to nitrogen ratio is 1.34 and the phosphorus to inositol ratio is 2.37 while, in the same fraction from soybean phosphatides, both ratios approximated two.

The countercurrent distribution of the alcohol insoluble fraction between methanol and hexane indicated that at least two major types of components are present. Upon partition between the two solvents, one of these is more soluble in 95% methanol and the other is more soluble in hexane. In addition, there is a small peak in the weight curve occurring at tubes 14 and 15.

It will be noted that the peak at the right of the curve occurs in tube 24 rather than in tube 23 as published for soybean phosphatides. However in several subsequent distributions of this fraction of soybean phosphatides in separatory funnels this peak has always been found in tube 24. It is believed that in the published work the band was broadened and the peak was shifted to tube 23 by a movement of the solvent interface, which in turn was caused by the more concentrated phosphatide solution used.

In contrast to the distribution pattern for the alcohol-insoluble soybean phosphatides where sugar is concentrated in the methanol-soluble fractions, most of the sugar of the corn phosphatides is found in the hexane-soluble fractions. Tube 24 contains 16% sugar determined by Somogyi's copper reduction method and calculated as galactose while tube 0 contains only 3.8%. This is the reverse of the situation with soybean phosphatides in which the sugar was concentrated in the methanol-soluble fractions and tube 23 contained only 3.9%. The high sugar content of the hexane-soluble fractions suggests the presence of a sugar-containing phosphatide such as

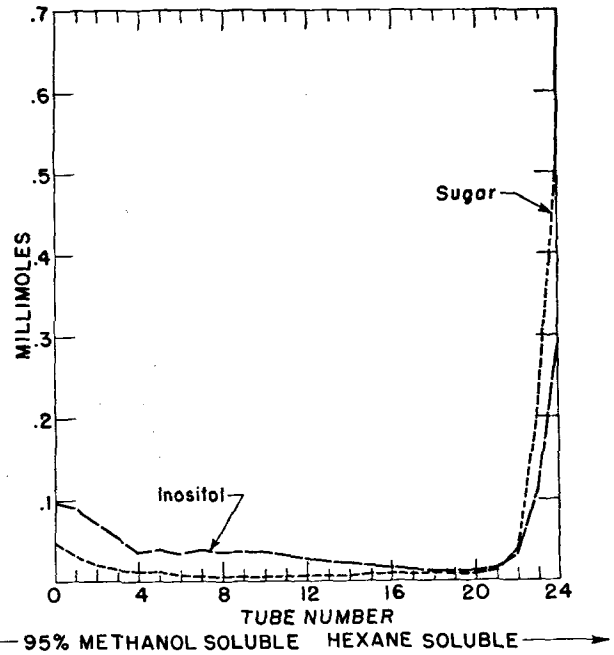

FIG. 3. Sugar and inositol results of the countercurrent distribution of the alcohol-insoluble fraction.

proposed by Woolley for lipositol (10). In soybean inositides by contrast sugar appeared only to be "carried along" rather than to be a component of the molecule.

The percentages of phosphorus, nitrogen, and inositol are greater in the fractions comprising the large peaks at each edge of the curve than in the center. For example, tube 0 contains 3.6% phosphorus, 1.2% nitrogen, and 8% inositol, and tube 24 contains 2.7% phosphorus, 1.5% nitrogen, and 9% inositol. In tube 15, which constitutes a small peak at the center of the weight curve, there is only 2.1% phosphorus, 0.27% nitrogen, and 5% inositol. In considering the inositol contents, it should be pointed out that the inositol was estimated by a microbiological assay. This procedure is highly specific for inositol, but the results are considered to be accurate to only about 10%.

In the work with soybean phosphatides it appeared that the phosphoinositides contained two moles of phosphorus for each mole of nitrogen and inositol. However the phosphorus to nitrogen and phosphorus to inositol ratios of the corn inositide fractions vary widely and somewhat irregularly. Since in most cases the ratios do not approximate whole numbers, it seems unlikely that any fraction contains a single pure compound. However it is observed that in the left hand portion of the curve phosphorus to nitrogen ratios are greater than one while in the right hand portion of the curve the ratio is one or slightly less than one. As can be seen from the curves in Figures 2 and 3, tubes 23 and 24 appear to contain a large amount of material with one mole of phosphorus for each mole of nitrogen and sugar.

Summary

Corn phosphatides have been separated into alcohol-soluble and alcohol-insoluble fractions employing the same procedure previously used with soybean phosphatides. Alcohol-soluble and alcohol-insoluble portions have been fractionated by countercurrent distribution. The alcohol-soluble portion was found to contain lecithin and a small amount of cephalin. In addition to the phosphatides a nitrogen containing compound was concentrated in the 95% methanol-soluble fractions along with a small amount of sugar. As with the corresponding alcohol-insoluble fraction from soybean phosphatides, two major types of phosphoinositides are found to be present, those more soluble in hexane and those more soluble in 95% methanol. In contrast to soybean phosphatides most of the sugar is concentrated in the hexane-soluble fractions. The phosphorus to nitrogen and phosphorus to inositol ratios vary widely and do not approximate two as was the case in the soybean inositides. However the more hexane-soluble fractions seem to be made up largely of material with one mole of phosphorus for each mole of nitrogen and sugar.

Acknowledgment

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ABSTRACTS . . . . . Don Whyte, Editor

• Oils and Fats

R. A. Reiners, Abstractor

RAPID FAT DETERMINATION IN PLANT CONTROL OF CACAO PRODUCTS. F. X. Kobe (Rockwood and Co.). *Anal. Chem.* 22, 700 (1950). Finely ground samples are extracted with petroleum ether at room temperature and the solids separated from the miscella in a centrifuge. Results are comparable to those obtained by the A.O.A.C. method.

MICRODETERMINATION OF UNSATURATED FATTY ACIDS BY ALKALI ISOMERIZATION. L. C. Berk, N. Kretchmer, R. T. Holman, and G. O. Burr (Univ. of Minnesota). *Anal. Chem.* 22, 718 (1950). The ethylene glycol-potassium hydroxide reagent normally used for alkali isomerization is unsuitable for isomerization of small (100 microgram) samples of fat due to high background absorption. This difficulty is avoided by isomerization in aqueous potassium hydroxide at high temperatures (180°).

DETECTION OF SOLVENT RESIDUES IN EXTRACTED GROATS. W. Wodsak (Hyg. Inst. Hansestadt, Hamburg, Ger.). *Z. Lebensm.-Untersuch. U. -Forsch.* 90, 265-72(1950). Detection of residual solvent in commercial solvent extraction residues was possible by determining the fluorescence of the steam distillate of the isobutyl alcohol-treated sample or by determining the vapor pressure of the materials volatile under high vacuum. A 100-g. sample under vacuum was heated to 160°, and volatile material was condensed with liquid air. The vapor pressure of the condensate at 0° was determined. (*Chem. Abs.* 44, 6044)

DETERMINATION OF NEUTRAL FAT IN THE HIGHLY ACID FATS. F. Provvedi (Lab. Chim. Provinciale, Bergamo, Italy). *Olii Minerali, Grassi E Saponi, Colori E Vernici* 26, 69-72(1949). Dissolve 5 g. of the fat in 50 cc. of 95% ethanol, saponify, add H_2O to obtain a 50% alcohol solution, decompose the soap with H_2SO_4, dissolve the fatty acids in petroleum ether, evaporate the solvent, and weigh the residue. Determine the acid no. of the fatty acids (I_a) and that of the original fat (I_f), and calculate the % of the fatty acids by the formula: $X = 100\ I_f/I_a$. The neutral fat is: $100 - X$. (*Chem. Abs.* 44, 5119)

THE ESTIMATION OF HORSE-FAT IN ADMIXTURE WITH OTHER FATS. R. A. Dalley. *Analyst* 75, 336(1950). The method depends on the presence in horse-fat of ½% of linoleic acid and its relative scarcity in other animal fats. Satisfactory results were obtained on mixtures containing as little as 5-10% horse-fat admixed with pig, mutton, and beef fat.

QUALITATIVE REACTION FOR SESAME OIL IN OTHER FOOD OILS. N. E. Bührer (Inst. Biol., Curitiba, Brazil). *Arquiv. Biol. E. Tecnol., Inst. Biol. E Pasquisas Tecnol., Curitiba, Brazil* 3, 57-9(1948). Tests with pure and mixed almond and sunflower oils showed that the Villavecchia-Fabris test (0.1 ml. of a 2% solution of furfural in the presence of HCl) is reliable. (*Chem. Abs.* 44, 5492)

CHROMATOGRAPHIC DETERMINATION OF VOLATILE FATTY ACIDS IN SILAGE. E. Brouwer and H. J. Nijkamp (Landb. Hogeschool, Wageningen, Netherlands). *Chem. Weekblad* 46, 37-9(1950). Chromatographic examination of 10 samples of grass silage showed that formic acid, propionic acid, and some higher volatile fatty acid may be present besides acetic and butyric acids. In silages with low pH the contents of formic and