METHODS FOR LIPID ANALYSIS
An Annotated Bibliography

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METHODS FOR LIPID ANALYSIS

Since the early 1930's the field of lipid research has grown tremendously. In conjunction with the expansion of our knowledge of lipids, many methods for the determination of these constituents have been published at a pace which makes it difficult for the investigator to keep abreast of developments. This bibliography was compiled in order that references to the many methods might be brought together and made more readily available.

No effort has been made to include papers wherein the method of choice is "standard" or only slightly modified, or those referred to in the "New Methods" section of the Journal of Lipid Research. Of the methods published before 1930, only the classical ones have been included. The papers are listed alphabetically by first author. Chemical Abstract numbers are included for the less common journals.

The author wishes to thank Dr. Charles G. Wilber and Mr. Paul F. Robinson of the Army Chemical Center for their many helpful suggestions.

Key to Symbols:

★ Separation and purification
● Glycerides, fatty acids and total lipids
▲ Phospholipids
■ Cholesterol

Abell, L. L., B. B. Levy, B. B. Brodie, and F. E. Kendall
1952. A simplified method for the estimation of total cholesterol in serum and demonstration of its specificity. ■ Journal of Biological Chemistry, 195: 357-366.

Serum is treated with alcoholic KOH to hydrolyze the cholesterol esters, the freed cholesterol is extracted into petroleum ether, and an aliquot of the extract is used for determination of cholesterol by the Liebermann-Burchard color reaction.

It is shown by counter-current distribution that only about 2% of the material determined as cholesterol by this method is other material. The Schoenheimer-Sperry method produced the same results.

Abu-Nasr, A. M. and R. T. Holman
1954. Highly unsaturated fatty acids. III. Isolation of methyl eicosapentaenoate, ethyl docosapentaenoate, ★ and ethyl docosahexaenoate from ● cod liver oil esters by chromatography. Journal of the American Oil Chemists Society, 31: 41-45. Chemical Abstracts, 48:4229h (1954).

Fatty acids from cod liver oil were separated on a Darco G-60 charcoal-Hyflo Supercel column using isopropanol as solvent and methyl behenate as displacer.

Abu-Nasr, A. M. and R. T. Holman
1955. Isomerization of polyunsaturated fatty acids and their esters by sodium amide in liquid ammonia. Journal of ● the American Oil Chemists Society, 32: 414-418. Chemical Abstracts, 49:12857d (1955).

Polyenoic acids were isomerized by using sodium amide in liquid ammonia, and determined by ultraviolet and infrared spectrophotometry.

Ahrens, E. H., Jr. and L. C. Craig
1952. Separation of the higher fatty acids. ★ Journal of Biological Chemistry, 195: ● 299-310.

Countercurrent distribution was used to separate a synthetic mixture of lauric, myristic, palmitic, and stearic acids; a synthetic mixture of oleic, linoleic, and linolenic acids; and an unknown mixture of

the fatty acids obtained by hydrolysis of pig mesenteric fat. Separations of saturated or unsaturated groups was good, but overlapping occurred in mixtures containing both saturated and unsaturated acids.

Ahrens, E. H., Jr.

1955. Application of countercurrent distribution for the study of lipids. Proceedings of the International Conference on Biochemical Problems of Lipids, 2nd Ghent.

★

A discussion of the applicability and use of counter-current distribution for lipid studies.

Albers, R. W. and O. H. Lowry

1955. Flourometric determination of 0.1 to 10 micrograms of cholesterol. Analytical Chemistry, 27: 1829-1831.

■

The flourescence produced by cholesterol in a trichloroethane, sulfuric acid, and acetic acid solution is used to measure the cholesterol extracted from 1 to 25 μg. of brain or other tissue. A stable, sensitive and precise method.

Allen, R. J. L.

1940. The estimation of phosphorus. Biochemical Journal, 34: 858-865.

▲

A modification of the Fiske-Subbarow reaction which uses amidol-sodium bisulfite as reducing agent.

Anderson, D. M. W.

1959. Applications of infrared spectroscopy: the identification and determination of gas-chromatographic fractions. Analyst, 84: 50-55.

Methods are described for collecting the components of mixtures separated by gas chromatography, and identification and quantitative determination of the components by vapor-phase infra-red spectroscopy.

Appleton, H. D., B. N. Ladu, B. B. Levy, J. M. Steele, and B. B. Brodie

1953. A chemical method for the determination of free choline in plasma. Journal of Biological Chemistry, 205: 803-813.

The method involves extraction of choline into acetone, evaporation of the acetone, and removal of interfering matter with butanol. After precipitation of choline as enneaiodide, the precipitate is dissolved in ethylene dichloride and measured spectrophotometrically at 365 mμ. As little as 5 μg. of choline is estimated. The method appears to be specific when used for plasma choline, but material other than choline is also assayed when the method is used for urine or tissue analyses. Adsorbed iodine does not interfere.

Arcus, A. C.

1959. Nephelometric detection of lipides in chromatographic column effluents. Analytical Chemistry, 31: 1618-1620.

A method is described for the rough estimation of the quantity of lipid material in effluents from chromatographic columns. The lipid material is precipitated from a methanolic solution with water, and the suspension is measured by nephelometry. The method is not specific, and is only applicable to material soluble in MeOH and insoluble in MeOH-H_2O (1:2).

Armbruster, O. and U. Beiss

1957. Paper chromatography of the phosphatides. Naturwissenschaften, 44: 420-421. Chemical Abstracts, 52:10268f (1958).

★
▲

A study of various paper-solvent combinations with regard to their suitability for separation of phosphatides. Data concerning those combinations found to be acceptable are given.

Artom, C.

1941. Cephalins, choline-containing phospholipids, and total phospholipids in normal human plasma. Journal of Biological Chemistry, 139: 65-70.

★
▲

A comparison of methods of extraction and purification of lipids. The methods compared were:

1. Extraction with cold alcohol followed by continuous hot alcohol extraction.

2. Precipitation with ammonium sulfate of the plasma proteins at pH 3, followed by continuous extraction with hot alcohol.

3. Folch and Van Slyke method (Proceed-

ings of the Society of Experimental Biology and Medicine, 41: 514, 1939) of precipitation with "colloidal iron" and magnesium sulfate.

The values that were obtained indicated that extraction of the phospholipids by all the above methods was probably complete, but washing procedures yield final products of slightly different compositions. The Folch and Van Slyke procedure was found to be reliable for routine analyses.

Artom, C.

1945. A quantitative method for ethanolamine and serine as a basis for the determination of phosphatidyl ethanolamine and phosphatidyl serine in tissues. Journal of Biological Chemistry, 157: 585-594.

▲

The method is based on the reaction of an ethanolamine-serine mixture with alkaline periodate before and after ethanolamine is removed by adsorption on Permutit. The NH_3 produced by the reaction is steam distilled and determined. Conditions for use and limitations of the method are discussed.

Ashley, B. D. and U. Westphal

1955. Separation of small quantities of saturated higher fatty acids by reversed-phase paper chromatography. Archives of Biochemistry and Biophysics, 56: 1-10.

★ ●

Fatty acids were separated on paraffin oil- or latex-coated paper with aqueous methanol saturated with cyclohexane or trimethylpentane, and aqueous methanol saturated with paraffin oil, respectively. Spots were detected by spraying with bromothymol blue, or immersing in a solution of lead acetate and developing the spots as lead sulfide or rhodizonate. Separates 10-50 μg. of C_{12}-C_{24} acids.

Awe, W. and B. Grote

1958. Paper chromatography of thiocyanogen derivatives of fatty acids. Fette, Seifen, Anstrichmittel, 60: 806-809. Chemical Abstracts, 53: 3737h (1959).

★ ●

Fatty acid cyanogen derivatives are separated by paper chromatography using an acetic acid-undecane solvent system. The separated derivatives are located by spraying the dried chromatogram with an $FeNH_4(SO_4)_2$ or $FeCl_3$ solution in an ammonia atmosphere.

Axelrod, J., J. Reichenthal, and B. B. Brodie

1953. The direct determination of phosphatidyl ethanolamine and phosphatidyl serine in plasma and red blood cells. Journal of Biological Chemistry, 204: 903-911.

▲

Ethanolamine and serine are determined by spectrophotometric measurement of their colored dinitroflourobenzene derivatives.

It was found that sensitivity of the Fiske and Subbarow phosphorus determination (Journal of Biological Chemistry, 66: 375, 1925) was increased 4-fold by heating 10 minutes in a boiling water bath after addition of reagents.

Ayers, C. W.

1956. Estimation of the higher fatty acids C_7-C_{18}. Analytica Chimica Acta 15: 77-83.

●

Methods are described for estimation of the C_7-C_{18} fatty acids as their copper or cobalt soaps. The soaps are dissolved in chloroform and the optical density of the solution is measured at 675 mμ. for copper or 525 mμ. for cobalt.

Ballance, P. E. and W. M. Crombie

1958. Paper chromatography of saturated and unsaturated fatty acids. Biochemical Journal, 69: 632-640.

★ ●

Methods and solvent systems are described for reversed-phase chromatography of fatty acids on paper impregnated with paraffin oil, castor oil, or polythene. Data on the separation of over 40 fatty acids are given.

Bargeton, D., M. E. Tricand-Redel, and P. Gros

1959. Comparison of results from three methods for the determination of serum cholesterol. Revue francaise d'etudes cliniques et biologiques, 4: 326-334. Chemical Abstracts, 53: 18143e (1959).

■

A comparison of the Schoenheimer-Sperry (Journal of Biological Chemistry, 106: 145, 1934), Grigaut (Compte rendu hebdomadaire des séances et mémoires de la Société de biologie, 112: 34, 1933), Machebouf and

Delsal (Bulletin de la Société de chimie biologique, 24: 296, 1942), and Abell (Journal of Biological Chemistry, 195: 351, 1952) methods for cholesterol determination. The S-S and Abell methods were found to be superior to the other two methods with regard to the standard error inherent in the method.

Barron, E. J. and D. J. Hanahan

1958. Observations on the silicic acid chromatography of the neutral lipids of rat liver, beef liver, and yeast. Journal of Biological Chemistry, 231: 493-503.

★

Pigments and hydrocarbons were eluted from a column of silicic acid with hexane, sterol esters with 15% benzene in hexane, triglycerides and free fatty acids with 5% ether in hexane, free sterols with 15-20% ether in hexane, diglycerides with 30% ether in hexane, and monoglycerides with 90-100% ether in hexane.

Barry, G. T., Y. Sato, and L. C. Craig

1951. Distribution studies. XIII. Separation and estimation of the higher normal fatty acids. Journal of Biological Chemistry, 188: 299-306.

★ ●

C_5 to C_{18} normal fatty acids were separated by counter-current distribution in isopropyl ether and 1 M potassium phosphate and estimated by titration.

Bartlett, G. R.

1959. Phosphorus assay in column chromatography. Journal of Biological Chemistry, 234: 466-468.

▲

A modified Fiske-Subbarow method which uses heating of the phosphorus reaction mixture in H_2SO_4 to produce an increase in light absorption at 830 mμ. This method permits greater latitude in reagent concentrations. The color developed is stable for 24 hours.

Bauer, F. C., Jr. and E. F. Hirsch

1949. A new method for the colorimetric determination of the total esterified fatty acids in human sera. Archives of Biochemistry, 20: 242-250.

●

Fatty acid esters are converted to hydroxamic acids and determined as colored ferric salts. A discussion of the procedure and sources of error is included.

Beadle, G. W.

1944. An inositolless mutant strain of neurospora and its use in bioassays. Journal of Biological Chemistry, 156: 683-689.

▲

A method is described which is suitable for use with concentrations of inositol between 5 and 30 μg. in 20 ml. of medium. A mutant strain of Neurospora crassa which requires inositol for growth is used in the bioassay.

Beattie, F. J. R.

1936. A colorimetric method for the determination of choline and acetylcholine in small amounts. Biochemical Journal, 30: 1554-1559.

Choline and acetylcholine are precipitated as their reineckates. The reineckate is dissolved in acetone and measured colorimetrically.

Beerthuis, R. K., G. Dijkstra, J. G. Keppler, and J. H. Recourt

1959. Gas-liquid chromatographic analysis of higher fatty acids and fatty-acid methyl esters. Annals of the New York Academy of Science, 72(art. 13): 616-632.

★ ●

Fatty acids were separated by gas chromatography and the separated acids at the exit of the column were combusted over CuO at 1000° and the evolved CO_2 was measured katharometrically.

Benham, G. H. and L. Klee

1950. An improved method for the determination of iodine numbers. Journal of the American Oil Chemists Society, 27: 127-129. Chemical Abstracts, 44:5119c (1950).

A modification of the method of Rosenmund and Kuhnhenn (Ber. 56: 1262, 1923). The use of Hg acetate permits a determination to be done in 1 minute.

Berenblum, I. and E. Chain

▲ 1938. An improved method for the

colorimetric determination of phosphate. Biochemical Journal, 32: 295-298.

Organic material is digested with $HClO_4$ and the phosphorus is converted to phosphomolybdate. The phosphomolybdate is extracted into isobutyl alcohol, reduced with $SnCl_2$, and the color is read colorimetrically. The method is not subject to most of the interferences which affect the methods of Fiske and Subbarow and Kuttner and Cohen. The standard method will determine 1-100μg. of phosphorus, and the micro method 0.1-10μg.

Bergmann, W.

1940. The splitting of digitonides. ■ Journal of Biological Chemistry, 132: 471-472.

Digitonides are split with dry pyridine, the pyridine is distilled off, and the digitonin is extracted with ether. The residue is steroid in yield of over 90%.

Bergström, S. and K. Pääbo.

1954. A method for the separation of saturated and monounsaturated fatty ★ ● acids through hydroxylation. Acta chemica scandinavia, 8: 1486-1487.

The fatty acids are hydroxylated with performic acid, methylated, and separated by elution from a silicic acid column.

Berk, L. C., N. Kretchmer, R. T. Holman, and G. O. Burr

1950. Microdetermination of unsaturated fatty acids by alkali isomeriza- ● tion. Analytical Chemistry, 22:718-720.

Aqueous alkali and high temperature and pressure are used for isomerization of unsaturated fatty acids. The fatty acids are then determined by ultraviolet spectrophotometry. A discussion of the method and data for linoleic, linolenic, and arachidonic acids are presented.

Bertram, S. H. and R. Rutgers

1938. The estimation of glycerol and and some other hydroxylated compounds. Recueil des travaux chimiques des Pays-Bas et de la Belgique, 57: 681-687. Chemical Abstracts, 32:7857[5] (1938).

The copper complex formed with glycerol in alkaline solution is acidified, KI is added, and the excess is titrated with thiosulfate.

Best, M., E. J. VanLoon, J. D. Wathem, and A. J. Seger

1954. Comparison of serum cholesterol methods. American Journal of Medi- ■ cine, 16: 601.

Serum cholesterol was determined by the methods of Schoenheimer-Sperry (Journal of Biological Chemistry, 106: 745, 1934); Abell, et al (Journal of Biological Chemistry, 195: 357, 1952); Gershberg-Forbes (Journal of Laboratory and Clinical Medicine, 27: 1439, 1942); modified Pearson, et al (Analytical Chemistry, 25: 813, 1953); and Zlatkis, Zak, and Boyle (Journal of Laboratory and Clinical Medicine, 41: 486, 1953). The Abell, et al, and Schoenheimer-Sperry methods agreed closely. When compared to the Abell method, the values of the Gershberg-Forbes, Pearson, and Zlatkis methods were higher by 15, 35, and 90 mg. percent, respectively. Since an alcohol-acetone extract of serum yields similar results by the Schoenheimer-Sperry and Zlatkis methods while the Zlatkis method yields values of 90 mg. percent higher when used directly on the same serum, it is suggested that the Zlatkis method measures a non-extractable lipid of protein as well as cholesterol.

Bevan, T. H., G. I. Gregory, T. Malkin, and A. G. Poole

1951. Chromatographic separation of choline-containing phospholipids from ★ ▲ phospholipid mixtures. Journal of the Chemical Society, 841-842.

Phospholipids were separated by chromatography on cellulose columns or on paper using a chloroform, water, and ethanol mixture as solvent. Ethanolamine- and serine-containing phospholipids were located on the developed chromatogram with ninhydrin, and choline-containing phospholipids were located with phosphomolybdic acid-stannous chloride reagent. Choline phospholipids were obtained uncontaminated.

Beveridge, J. M. R. and S. E. Johnson
1949. The determination of phospholipid phosphorus. Canadian Journal of Research, 27E: 159-163.

▲ Phospholipid is digested with H_2SO_4 and color is developed with molybdate-hydrazine sulfate reagent (Boltz and Mellon, Industrial and Engineering Chemistry, Analytical Edition, 19: 873, 1947) and read at 830 mμ. Accuracy of the method is within 1% on a 20 μg. sample of phosphorus.

Black, S.
1949. A microanalytical method for the volatile fatty acids. Archives of Biochemistry, 23: 347-359.

● The volatile fatty acids are determined by microdiffusion from an acidified solution into an alkaline solution, and microtitration of the excess alkali. Range: 0.2-2.0 μequivalents; error: < 0.03 μequivalent.

Blankenhorn, D. H. and E. H. Ahrens, Jr.
1955. Extraction, isolation, and identification of hydrolytic products of triglyceride digestion in man. Journal of Biological Chemistry, 212: 69-81.

★ ● A description of methods for extraction of lipids from intestinal contents and separation into fatty acid, bile acid, and mono-, di-, and triglyceride fractions.

Bligh, E. G. and W. J. Dyer
1959. A rapid method of total lipid extraction and purification. Canadian Journal of Biochemistry and Physiology, 37: 911-917.

★ ● A simplification of the method of Folch, et al, (Journal of Biological Chemistry, 191: 833, 1951). Lipids are extracted by homogenization of tissue with $CHCl_3$-MeOH in such proportions that a miscible system is formed with the water in the tissue. The homogenate is separated into a chloroform layer containing the lipids and a methanolic layer containing the non-lipid material by addition of chloroform and water. Extraction of the lipids is essentially complete.

Bloor, W. R.
1916. The determination of cholesterol in blood. Journal of Biological Chemistry, 24: 227-231.

■ Blood is extracted with alcohol-ether (3:1), the extract is evaporated to dryness and the residue is extracted with chloroform. Cholesterol is then determined colorimetrically using the Liebermann-Burchard color reaction.

Bloor, W. R.
1928. The determination of small amounts of lipid in blood plasma. Journal of Biological Chemistry, 77: 53-73.

● Nicloux reagent ($Ag_2Cr_2O_7$ in H_2SO_4) is used for oxidation of the extracted lipid, and the excess dichromate is titrated with thiosulfate.

Bloor, W. R.
1929. The oxidative determination of phospholipid (lecithin and cephalin) in blood and tissues. Journal of Biological Chemistry, 82: 273-286.

▲ The phospholipids are separated by precipitation with acetone-$MgCl_2$ and determined by oxidation with Nicloux reagent and titration of excess dichromate with thiosulfate.

Bloor, W. R.
1947. A colorimetric procedure for determination of fatty acids. Journal of Biological Chemistry, 170: 671-674.

● Fatty acids are determined by colorimetric measurement of the color change produced by oxidation with Nicloux reagent. A special colorimeter is required.

Böhm, P., S. Dauber, and L. Baumeister
1954. Neuraminic acid, its occurrence and its determination in serum. Klinische Wochenschrift, 32: 289-292. Chemical Abstracts, 48:7674e (1954).

Serum protein is precipitated with trichloracetic acid, centrifuged, and washed with water. The precipitate is treated with Bial reagent and heated, and the red-violet reaction product of neuraminic acid is extracted with amyl alcohol and read at 570 mμ.

Böhm, P. and G. Richarz

1954. Determination of inositol in phosphatides. Zeitschrift für physiologische Chemie, 298: 110-120. Chemical Abstracts, 49:4462d (1955).

▲

Lipid material is hydrolyzed with HCl and the hydrolysate is chromatographed on paper. The separated inositol is extracted with water, oxidized with periodate, and measured iodometrically.

Boldingh, J.

1950. Fatty acid analysis by partition chromatography. Recueil des travaux chimiques des Pays-Bas et de la Belgique, 69: 247-261 (in English). Chemical Abstracts, 44:6348h (1950).

★ ●

A method is described for separation of C_6-C_{18} saturated n-fatty acids from their mixtures and hydroxy fatty acids from saturated n-fatty acids by chromatography on a rubber column.

Boldingh, J.

1953. Separation of fatty acids by chromatography. Koninkl. Vlaam. Acad. Wetenschap., Letter. en Schone Kunsten Belg., Kl. Wetenschap., Intern. Colloquim Biochem. Problem Lipiden, Brussels, p. 64-81 (in English). Chemical Abstracts, 49:2097h (1955).

★ ●

Fatty acids were separated on a column of rubber powder swollen with peanut oil, with acetone-water-peanut oil as mobile phase.

Borgström, B.

1952. Investigation on lipid separation methods. Separation of phospholipids from neutral fat and fatty acids. Acta physiologica Scandinavica, 25: 101-110.

★ ● ▲

Acetone-$MgCl_2$ precipitation gives good separation of phospholipids, with about 1% of the phospholipids dissolving in the solution. MgO chromatography gives phospholipid-free neutral fat, but poorer purity of phospholipid, and cannot separate free fatty acids from choline-containing phospholipids. Non-choline-containing phospholipids are not recoverable unchanged. Silicic acid chromatography is slower than acetone-$MgCl_2$ precipitation, but gives best all-around results; neutral fat and free fatty acids are eluted quantitatively with $CHCl_3$, and phospholipids with MeOH.

Borgström, B.

1952. Investigation of lipid separation methods. Separation of cholesterol esters, glycerides, and free fatty acids. Acta physiologica Scandinavica, 25: 111-119.

★ ● ■

Silicic acid chromatography is used for separation of cholesterol esters from glycerides and free fatty acids. Free fatty acids are separated from glycerides in absence of lower glycerides by extraction of the acids from a petroleum ether solution with alkaline 50% ethanol. In the presence of lower glycerides which would be extracted into the alcoholic solution, IRA-400 ion exchanger is used to separate the free fatty acids from the glycerides.

Borgström, B.

1954. Investigation on lipid separation methods. III. Separation of tri-, di-, 1-mono-, and 2-monoglycerides. Acta physiologica Scandinavica, 30: 231-239.

★ ●

In the method described, tri-, di-, and monoglycerides are separated from their mixtures by chromatography on silicic acid (2-monoglycerides are partially isomerized). 1-mono- and 2-monoglycerides are separated from tri- and diglycerides by partition chromatography with heptane and 80% aqueous ethanol as phases. After oxidation of 1-monoglycerides with periodic acid, the 2-monoglycerides can be isolated by chromatography on silicic acid.

Boyd, E. M.

1931. Low phospholipid values in dog plasma. Journal of Biological Chemistry, 91: 1-12.

▲

A modification of Bloor's oxidation method (Journal of Biological Chemistry, 82: 273, 1929) for use in determination of low phospholipid levels. Essentially the same phospholipid values were obtained with or without heat in the alcohol-ether extraction of plasma. Substitution of ethyl ether for petroleum ether in extraction of the alcohol-

ether residue gave more consistent and slightly higher phospholipid values.

Boyd, E. M.
1933. A differential lipid analysis of blood plasma in normal young women by microoxidative methods. Journal of Biological Chemistry, 101: 323-336.

The oxidative procedure is recommended for determination of various lipids in the same extract as it eliminates the possibility of summation of errors by using different methods.

Bloor's method (Journal of Biological Chemistry, 77: 53, 1928) is used to determine total fatty acids after saponification of the lipids (99% recovery of known solutions and reproducibility of ± 2%), and modifications of Yasuda's method (Journal of Biological Chemistry, 92: 303, 1931) for oxidation of cholesterol digitonide (stock determinations reproducible ± 2%). Confirms observations that 1/2 hour heating is necessary for complete oxidation. Boyd's modifications (Journal of Biological Chemistry, 91: 1, 1931) of Bloor's method are used for phospholipid determination. Free cholesterol is estimated after removal of phospholipid. The method checks within experimental error with values from direct precipitation from alcohol-ether extract (96% recovery with ± 4.5% agreement on duplicates).

Boyd found difficulty in Okey's method (Journal of Biological Chemistry, 88: 367, 1930) for oxidative determination of cholesterol as the digitonide with respect to reproducibility and accuracy. Phospholipids are saponified with NaOH before determination of iodine numbers of phospholipid fatty acids, as direct determinations on whole phospholipid according to Yasuda (Journal of Biological Chemistry, 94: 401, 1931-2) gave erratic and variable results.

Boyd, E. M.
1936. The extraction of blood lipids. Journal of Biological Chemistry, 114: 223-234.

Effects of variables on extraction of blood lipids with Bloor's extract (alcohol-ether, 3:1) were studied.

It was found that when the extracts of blood are sufficiently diluted (at least 20:1) extraction is complete.

Boyd, E. M.
1936. The extraction of lipids from the red blood cells. Journal of Biological Chemistry, 115: 37-45.

The effects of variables on extraction of blood cell lipids were studied and are discussed.

Incompletely dried acetone used in $MgCl_2$ precipitation gave low phospholipid values.

Boyd, E. M.
1937. The storage of lipid extracts. Journal of Biological Chemistry, 121: 485-496.

A decrease of phospholipid during 1-3 months of storage was observed. After 3-6 months of storage a decrease in cholesterol esters, total cholesterol, and total lipid, increase in "neutral fat" and free cholesterol, and no appreciable change in total fatty acids was observed. Changes were generally unaffected by storage conditions of light, temperature (refrigerator or room), or solvent (EtOH or EtOH-Petr. ether). See also: Halliday (Journal of Nutrition, 16: 285, 1938).

Boyd, E. M.
1937. The oxidative micro-estimation of plasma total lipid. Journal of Laboratory and Clinical Medicine, 22: 956-959.

The factor of 3.25 is proposed in place of the factor of 3.7 used by Bloor for conversion to total lipid value of the total fatty acid-total cholesterol titration with potassium dichromate.

Boyd, E. M.
1938. The oxidative micro-estimation of blood lipids. American Journal of Clinical Pathology. Technical Supplement, 2: 77-90.

Lipids are extracted with alcohol-ether (3:1), oxidized with Nicloux reagent and $K_2Cr_2O_7$, and unreacted dichromate is titrated.

Boyd, G. S.
1954. The estimation of serum lipopro-

teins. A micromethod based on zone electrophoresis and cholesterol estimation. Biochemical Journal, 58: 680-685.

A method is described for the separation of serum lipoproteins by paper electrophoresis. Protein was located by staining with bromophenol blue. Cholesterol was extracted with acetone-ethanol (1:1) and estimated by the Liebermann-Burchard reaction.

Boyd, M. J. and M. A. Logan

1942. Colorimetric determination of serine. Journal of Biological Chemistry, 146: 279-287.

The formaldehyde formed by the action of periodate on serine is quantitatively distilled from the reaction mixture, condensed with 1, 8-dihydroxynaphthalene-3, 6-disulfonic acid, and measured colorimetrically. Required 1-5 mg. serine. Error 1-2%.

Bradbury, R. G.

1951. Microdetermination of glycerol. Mikrochemie (vereinigt mit Mikrochemica Acta), 38: 114-119. Chemical Abstracts, 45:6857c (1951).

Glycerol is freed from the fat by hydrolyzing with KOH and converted to isopropyliodide by reaction with HI in the presence of propionic acid. The I_2 freed during the reaction is collected in a solution of Br and sodium acetate in acetic acid, which oxidizes the I to IO_3^-. Excess Br is removed from the solution and I is freed and titrated with $Na_2S_2O_3$.

Bragdon, J. H.

1951. Colorimetric determination of blood lipides. Journal of Biological Chemistry, 190: 513-517.

A modification of Bloor's dichromate oxidation method (Journal of Biological Chemistry, 170: 671, 1947). $K_2Cr_2O_7$ in H_2SO_4 is used in place of $Ag_2Cr_2O_7$, as Ag may contain Cl which would interfere with colorimetric measurements. Better oxidation is obtained, as well as a straight-line curve of optical density.

Bragdon, J. H.

1956. Lipid nomenclature (recommendations regarding the reporting of serum lipids and lipoproteins). American society for the study of arteriosclerosis. Circulation Research, 4: 129.

The following are recommended:

1. The factor 1.67 be used to convert weight of cholesterol present in esterified form into weight of cholesterol ester. (Kelsey and Longenecker, Journal of Biological Chemistry, 139: 727, 1949).

2. The ratio of free cholesterol to total cholesterol be expressed as a decimal.

3. The factor 25 be used to convert lipid phosphorus to phospholipid.

4. The amount of phosphorus in a Bloor extract be considered the correct lipid phosphorus content of serum. (Van Slyke, Journal of Biological Chemistry, 200: 525, 1953).

5. Relative amounts of cholesterol and phospholipid be expressed as the ratio of total cholesterol to phospholipid, as a decimal. Values used should represent weights, not moles.

6. Triglycerides and fatty acids be reported as triglycerides, which predominate. The term "neutral fat" be avoided or specifically defined. Triglycerides be reported in milligrams. The term millequivalents be confined to values for fatty acids determined by titration when additional determinations are not available for the calculation of triglycerides.

Brand, F. C. and W. M. Sperry

1941. The determination of cerebrosides. Journal of Biological Chemistry, 141: 545-553.

A modification of the Miller and Van Slyke (Journal of Biological Chemistry, 114: 583, 1936) method for sugar determination. The cerebrosides are hydrolyzed, the freed sugars are oxidized with ferricyanide, and the ferrocyanide formed is titrated with ceric sulfate.

Brante, G.

1949. Studies on lipids in the nervous system with special reference to quantitative chemical determination. Acta

physiologica Scandinavica, 18 (suppl. 63): 1-189.

Methods are described for the determination of total lipids (gravimetric), phospholipids (modified Fiske-Subbarow), choline, ethanolamine, amino nitrogen, glycerol, hexose, inositol, and cholesterol.

Breusch, F. L. and E. Ulusoy

1946. Isolation and identification of fatty acids as bis-(p-dimethylaminophenyl)-ureides. Archives of Biochemistry, 11: 489-498.

★ ●

Free fatty acids are isolated from fat mixtures in approximately 80% yield by precipitation as ureides from ether solution with bis-(p-dimethylaminophenyl)-urea. The properties, melting points, and solubilities of the ureides of various fatty acids are given.

Brown, F.

1950. Separation of the lower fatty acids as anions by paper chromatography. Biochemical Journal, 47: 598-600.

★ ●

The lower fatty acids were separated by paper chromatography as their Na salts using n-butanol as developer in the presence of ammonia. The spots are revealed by spraying with a solution of bromothymol blue.

Brown, H. H., A. Zlatkis, B. Zak, and A. J. Boyle

1954. Rapid procedure for determination of free serum cholesterol. Analytical Chemistry, 26: 397-399.

■

$AlCl_3$ or $Al(OH)_3$ is used to gather digitonide precipitate and the color produced with the ferric chloride reagent of Zlatkis, Zak, and Boyle (Journal of Laboratory and Clinical Medicine, 41: 486, 1953) is measured colorimetrically.

Brown, M., D. A. Yeadon, L. A. Goldblatt, and J. W. Dieckert

1957. Chromatography of phospholipids and related compounds on glass paper impregnated with silicic acid. Analytical Chemistry, 29: 30-31.

★ ▲

A method for the separation and identification of phospholipids and some of their cleavage products is described. A phenol, ethyl ether, acetone, and water solution is used to develop the chromatogram on a silicic acid-impregnated glass paper. Charring with sulfuric acid is used to locate spots on the paper. (Ethanolamine and choline chloride are not detectable by this technique.) A modified Dragendorf reagent (Bregoff, et al, Journal of Biological Chemistry, 205: 565, 1953) or phosphomolybdic acid reagent (Levine and Chargaff, Journal of Biological Chemistry, 192: 465, 1952) is used for identification of quaternary ammonium compounds, and Ninhydrin reagent for detection of primary amino compounds.

Brückner, J.

1941. Estimation of cerebrosides. Zeitschrift für physiologische Chemie, 268: 163-170. Chemical Abstracts, 35:7436[7] (1941).

Cerebrosides are hydrolyzed with H_2SO_4, the fatty acids are removed with $CHCl_3$, and galactose is determined using the orcin reagent. Cerebrosides are calculated from the amount of galactose.

Buchanan, M. A.

1959. Paper chromatography of the saturated fatty acids. Analytical Chemistry, 31: 1616-1618.

★ ●

A method is described for chromatography of fatty acids on paper treated with mineral oil. The chromatogram is developed at 37°C with acetic acid - 88% formic acid - 30% hydrogen peroxide (6:1:1) for separation of the even-numbered straight-chain saturated fatty acids from lauric to lignoceric acid in the presence of large amounts of unsaturated acids. Mercuric acetate and s-diphenylcarbazide were used for locating spots on the chromatogram.

Bulliard, H., I. Grundland, and M. Maillet

1950. Histochemical detection of cell phosphatides. Compte rendu hebdomadaire des séances et mémoires de la Société de biologie, 144: 192-194. Chemical Abstracts, 44:10024i (1950).

▲

Phosphatides were detected in tissue sections by soaking the formalin-fixed tissue in $CdCl_2$, washing in acetone, and converting the Cd which had combined with the leci-

thin into CdS by exposure to H_2S.

Burmaster, C. F.
1946. Microdetermination of α and β glycerophosphates. Journal of Biological Chemistry, 164: 233-240.
▲
Results of colorimetric measurement of the orthophosphate produced by the reaction of periodate on α glycerophosphate followed by acid hydrolysis agree with those obtained by titration of the reduced periodate. Microcolorimetric methods suitable for the analysis of sodium glycerophosphate and phosphorus compounds in phospholipid hydrolysates, for inorganic phosphorus, α phosphorus, $(\alpha + \beta)$ phosphorus, and total phosphorus are described.

Burmaster, C. F.
1946. The microdetermination of serine and ethanolamine in phospholipide hydrolysates. Journal of Biological Chemistry, 165: 1-6.
▲
Ethanolamine and serine are determined by microdiffusion of the ammonia produced by periodate in a solution nearly saturated with potassium metaborate. Cephalin nitrogen can be measured by this method in the presence of all the known components of phospholipid acid hydrolysates. It is a more specific measure of cephalin than the HNO_2 method of Van Slyke (Journal of Biological Chemistry, 16: 121, 1913-14). No indication was found of the reaction of hydroxylamine with periodate to form NH_3 which Nicolet (Journal of the American Chemical Society, 61: 1615, 1939; Journal of Biological Chemistry, 139: 687, 1941) described.
Obtained a better yield than Ramsey (Biochemical Journal, 35: 39, 1941), since Ramsey's addition of K_2CO_3 to acid solution causes generation of heat and evolution of CO_2 and some destruction of NH_3 by the hot acid periodate.

Burness, A. T. H. and H. K. King
1958. Detection of fatty acids on paper chromatograms by means of ninhydrin. Biochemical Journal, 68: 32P.
●
Ethylamine or ammonia is used in the developing solvent, and the salts formed with the fatty acids are detected by spraying the developed chromatogram with ninhydrin.

Cahn, T., J. Houget, and R. Agid
1948. The determination of glycerophosphoric acid. Application to the case of the phosphatides. Bulletin. Société chimique de France, pp 666-668. Chemical Abstracts, 42: 8860d (1948).
▲
Phosphatides are saponified, unsaponifiables are removed, the solution is acidified, the fatty acid is removed, glycerophosphoric acid is precipitated as the Ba salt, and P is determined. Unsaponifiable matter, fatty acids, and glycerophosphoric acid may all be determined on a 0.5 to 1.0 g. sample by this method.

Candela, A., P. Capella, and G. Jacini
1956. Researches on phosphatidic acids. III. Olii minerali, grassi e saponi, colori e vernici, 33: 99-101. Chemical Abstracts, 50: 16887g (1956).
▲
Choline, in 65.5% yield, was obtained by treatment of egg lecithin with carrot enzymes and a buffer, acidification of the solution with HCl, and extraction with Et_2O.

Cannon, J. A., K. T. Zilch, and H. J. Dutton
1952. Countercurrent distribution of methyl esters of higher fatty acids. Analytical Chemistry, 24: 1530-1532.
★ ●
The methyl esters of the higher fatty acids were separated by countercurrent distribution in a pentane-hexane vs nitroethane-nitromethane solvent system. Applicability of the method is discussed.

Carayon-Gentil, A., and E. Cortegiana
1942. Phosphoaminolipides of brain tissue. Fractionation and estimation of choline-containing substances. Bulletin de la Société de chimie biologique, 24: 89-96. Chemical Abstracts, 38: 6312[5] (1944).
★ ▲
Free choline, lecithin and cephalin, and sphingomyelin are obtained from brain tissue by selective extraction with Me_2CO, petroleum ether, and MeOH-$CHCl_3$ (3:1), respectively. The extracts are hydrolyzed with boiling alcoholic HCl and choline is estimated by acetylation and estimation by

action on leech muscle of the acetylcholine formed.

Cardini, C. E. and M. E. Serantes
1943. Methods of extracting sphingomyelin. Anales de farmacia y bioquimica. Buenos Aires, 14: 123-132. Chemical Abstracts, 38:4630^7 (1944).
★ ▲
Initial extraction of material by Bloor's method (Journal of Biological Chemistry, 82: 273, 1929), followed by reextraction of the tissue by the method of Thannhauser (Journal of Biological Chemistry, 129: 709, 1939) is recommended.

Carlson, L. A. and L. B. Wadström
1958. A colorimetric method of determining unesterified fatty acids in plasma. Scandinavian Journal of Clinical and Laboratory Investigation, 10: 407-414.
●
The lipids are extracted with chloroform-methanol (2:1). Phospholipids are separated on silicic acid, and fatty acids on Amberlite IRA-400 resin. The methylated fatty acids are determined colorimetrically as the hydroxyamic acids. Two milliliters of plasma may be used for determination of unesterified fatty acids, glycerides, phospholipids, and cholesterol. The method is sensitive to amounts of fatty acid from 0.5 to 4.0 μequivalents. With 2.0 μequivalent, recovery was 97.6% and error $\pm$ 2.5%.

Carlson, L. A. and L. B. Wadström
1959. Determination of glycerides in blood serum. Clinica Chimica Acta, 4: 197-205.
●
Glycerides are separated from phospholipids by silicic acid chromatography and saponified. The glycerol is then determined by periodic acid oxidation and estimation of the formaldehyde formed.

Carpenter, K. J., A. Gostis, and D. M. Hegsted
1957. Estimation of total cholesterol in serum by a micro method. Clinical Chemistry, 3: 233-238.
■
An adaptation of the method of Albers and Lowry (Analytical Chemistry, 27: 1829, 1955) for use in determination of blood serum cholesterol. H_2SO_4 is added to a 1, 1, 2-trichloroethane-acetic anhydride solution of cholesterol, and the flouresence which develops is measured.

Cavanaugh, D. J. and D. Glick
1952. Determination of cholesterol in microgram quantities of tissue. Analytical Chemistry, 24: 1839-1841.
■
A microadaptation of the Sperry-Webb method (Journal of Biological Chemistry, 187: 97, 1950), for use in analysis of cholesterol in microtome sections of tissue. Error of the method ranges from 1.5 to 4.5% depending on sample size.

Chapman, R. A. and K. MacKay
1949. The estimation of peroxides in fats and oils by the ferric thiocyanate method. Journal of the American Oil Chemists Society, 26: 360-363. Chemical Abstracts, 43:6840h (1950).
●
It was found, in agreement with the findings of Lea (Journal of the Society of Chemical Industry, 64: 106, 1945), that atmospheric oxygen interferes with the determination of peroxide by the ferric thiocyanate method. Exclusion of air and use of nitrogen atmosphere markedly reduced the peroxide values determined by the ferric thiocyanate method.

Charlampous, F. C. and P. Abrahams
1957. Biochemical studies on inositol. I. Isolation of myo-inositol from yeast and its quantitative enzymatic estimation. Journal of Biological Chemistry, 225: 575-583.
Methods for the extraction, purification, and crystallization of myo-inositol from yeast cells are described. A method is described for the colorimetric estimation of inositol using the reduction of 2, 6-dichlorophenolindophenol by inositol dehydrogenase in the presence of inositol.

Chargaff, E.
1942. A study of Lipoproteins. Journal of Biological Chemistry, 142: 291-504.
★ ▲
Ether extraction and dialysis were used to remove "lipovitellin" from egg yolk, and from it removed the phosphophatides.

Ethanolamine and choline in the phosphatide fraction were separated as ethanolamine 3,5-diiodosalicylate and choline chloride-6 $HgCl_2$ double salt.

Chargaff, E., C. Levine, and C. Green
1948. Technique for the demonstration by chromatography of nitrogenous lipide constituents, sulfur-containing amino acids, and reducing sugars. Journal of Biological Chemistry, 175: 67-71.

★ ▲

HCl hydrolysis and paper chromatography were used to separate components. Tested for choline with phosphomolybdic acid, ethanolamine and serine with ninhydrin, and reducing sugars with m-phenylenediamine dihydrochloride.

Chen, P. S., Jr., T. Y. Toribara, and H. Warner
1956. Microdetermination of phosphorus. Analytical Chemistry, 28: 1756-1758.

▲

A modification of the method of Ammon and Hinsberg (Zeitschrift für physiologische Chemie, 239: 207, 1936) for use in determining as little as 0.15 μg. of phosphorus in blood and urine. Ascorbic acid is used to reduce phosphomolybdate. Effects of variables are given. Results are identical with Fiske-Subbarow method. The method is suitable for lipid phosphorus determination.

Chiamori, N. and R. J. Henry
1959. Study of the ferric chloride method for determination of total cholesterol and cholesterol esters. American Journal of Clinical Pathology, 31: 305-309.

■

$FeCl_3$-acetic acid reagent is used to precipitate serum proteins, and the cholesterol color is developed with H_2SO_4. Bromine interference can be avoided by removing it with an ion-exchange resin.

Traces of digitonide interfered with the determination of cholesterol by the method of Pearson (Analytical Chemistry, 25: 813, 1953), using p-toluenesulfonic acid. Excess silver iodate will interfere with the color reaction (Rice and Lukasiewicz, Clinical Chemistry, 3: 160, 1957).

Christl, H.
1953. Determination of acetal phosphatide in tissues. Zeitschrift für physiologische Chemie, 293: 83-88. Chemical Abstracts, 49: 13346d (1955).

▲

A modification of the method of Feulgen (Zeitschrift für physiologische Chemie, 287: 90, 1951) for use in the determination of tissue acetal phosphatides.

Clayton, M. M., P. A. Adams, G. B. Mahoney, S. W. Randall, and E. T. Schwartz
1959. Micro methods for the determination of chylomicron counts, fatty esters, lipid phosphorus, and cholesterol in blood serum. Clinical Chemistry, 5: 426-444.

● ▲ ■

Procedures for determination of lipid components on 75 μl. samples of blood serum are given. After alcohol-ether extraction, the fatty esters are determined by colorimetric measurement of the ferric hydroxamate complex color developed with acidified ferric perchlorate (Hill, Industrial and Engineering Chemistry, Analytical Edition, 18: 317, 1946, Ibid. 19: 932, 1947, and Bauer and Hirsch, Archives of Biochemistry, 20: 242, 1949).

Phospholipids are oxidized with H_2SO_4 and H_2O_2 and determined by the Fiske-Subbarow colorimetric method. Cholesterol esters are hydrolyzed with benzyltrimethylammonium hydroxide and total cholesterol is determined by color development with H_2SO_4-acetic anhydride.

Cole, P. G., G. H. Lathe, and C. R. J. Ruthven
1953. The application of counter-current methods to the fractionation of lipid material from brain. Biochemical Journal, 54: 449-457.

★ ▲

A water-methanol-carbon tetrachloride solvent system is described for use in counter-current fractionation of brain lipids, and the behavior of the lecithin, cephalin, and sphingomyelin-cerebroside fractions is described. Variations of the solvent system and its uses are discussed.

Coleman, C. M. and G. Middlebrook

1957. Interface enrichment of methylene blue by fatty acids with micro-analytic applications. Science, 126: 163.

A heptane solution of fatty acids is enriched by methylene blue when in contact with an aqueous solution of the dye. Spectrophotometric measurement of the enriched heptane solution is used for determination of the amount of fatty acids present.

Collins, F. D. and L. W. Wheeldon

1958. Studies on phospholipids. 4. Determination of ethanolamine and serine. Biochemical Journal, 70: 46-49.

After extraction and dinitrophenylation, the lipids are hydrolyzed in ethanolic HCl. The hydrolysate is dried and dissolved in a solution of dimethylformamide-ethanol-n-butylamine, and tetramethylammonium hydroxide is added. The ethanolamine and serine values are then determined by differential color measurement at 393 mμ and 500 mμ.

Collins, F. D.

1959. Studies on phospholipids. 5. The separation of dinitrophenylated and methylated phospholipids by counter-current distribution. Biochemical Journal, 72: 281-287.

A method is described for the separation of phospholipids from sheep and rat brain lipids by counter-current distribution of their dinitrophenyl and methyl derivatives in several solvent systems.

Colman, D. M. and A. F. McPhee

1956. An improved method for determination of total serum cholesterol. American Journal of Clinical Pathology, 26: 181-186.

Direct saponification of the serum sample, extraction of the cholesterol, precipitation with digitonide, and color development with a modified Liebermann-Burchard reagent are all carried out in the same tube. Accuracy and precision are good.

Corcoran, G. B.

1956. Chromatographic separation and determination of straight-chain saturated monocarboxylic acids C_1 through C_{10} and dicarboxylic acids C_{11} through C_{16}.

The acids were separated by chromatography on a glycine-buffered column of silicic acid using a 1-butanol-chloroform mobile phase. Accuracy is ± 1%.

Cormier, M. and P. Jouan

1957. Separation of lipides by chromatography on paper impregnated with silicic acid. Bulletin de la Société de chimie biologique, 39: 1321-1327. Chemical Abstracts, 53:11491a (1959).

A method for the separation of lipids by chromatography on silicic acid-impregnated filter paper.

Cormier, M. and P. Jouan

1958. Separation of the total lipides of blood serum by chromatography on paper. Bulletin de la Société de chimie biologique, 40: 171-176. Chemical Abstracts, 53:18146b (1959).

A method is described for the separation of serum lipids by chromatography on silicic acid impregnated paper. The phospholipids are first separated by chromatography with a Et_2O-petroleum ether-Me_2CO(2:100:50) solvent mixture, and the glycerides, steroids, and cholesterol are then separated by using Et_2O-petroleum ether-heptane (4:50:50).

Cormier, M., P. Jouan, and L. Girre

1959. Separation of lipides by chromatography on paper impregnated with silicic acid. II. Possibilities and limitations of the method. Bulletin de la Société de chimie biologique, 41: 1037-1046. Chemical Abstracts, 54:4736e (1960).

Lipids were separated into steroid, triglyceride, and mono- and diglyceride groups by chromatography on SiO_2-impregnated paper using an ether-petroleum ether-heptane (8:50:50) solvent mixture. The applications of the method are discussed.

Corner, M.

1959. Rapid microdetermination of organically bound halogens, arsenic, phosphorus, and boron. Analyst, 84: 41-46.

The sample is combusted in O (Schöniger, Helvetica chimica acta, 39: 650, 1956), and phosphorus is precipitated as quinoline molybdophosphate and titrated.

Cotte, J. and E. Kahane

1950. The determination of choline as trimethylamine. Bulletin. Société chimique de France, 639-648. Chemical Abstracts, 45:1641e (1951).

The method consists of oxidation of choline by $KMnO_4$ in the presence of $Na_2S_4O_6$ and measurement of the formaldehyde formed.

Courchaine, A. J., W. H. Miller, and D. B. Stein, Jr.

1959. Rapid semimicro procedure for estimating free and total cholesterol. Clinical Chemistry, 5: 609-614.

A modification of the method of Zlatkis, Zak, and Boyle (Journal of Laboratory and Clinical Medicine, 41: 486, 1953). Phosphoric acid is used as solvent for ferric chloride, giving a stable color reagent. The method compares well with methods of Schoenheimer-Sperry (Journal of Biological Chemistry, 106: 745, 1934) and Abell, et al (Journal of Biological Chemistry, 195: 357, 1952).

Craig, B. M. and N. L. Murtz

1958. The separation of saturated and unsaturated fatty acid esters by gas-liquid chromatography. Canadian Journal of Chemistry, 36: 1297-1301.

Fatty acid methyl esters are separated according to chain length on a silicone grease column and according to chain length and degree of unsaturation on a plasticizer column using a firebrick support and helium as the carrier gas.

Crawford, N.

1959. Determination of serum α and β lipoprotein cholesterol. Clinica Chimica Acta, 4: 494-502. Chemical Abstracts, 53: 20208h (1959).

Lipoproteins are separated by paper electrophoresis and their cholesterol is estimated using $FeCl_3$.

Cremer, H. D.

1949. Tetrahydrofuran as extraction agent for lipide determination in blood and serum. Klinische Wochenschrift, 27: 755. Chemical Abstracts, 44: 8986d (1950).

A method is described which is similar to Bloor's method, but which uses hot tetrahydrofuran for extraction of blood or serum lipids. The method gives higher values than Bloor's method.

Crombie, W. M. L., R. Comber, and S. G. Boatman

1955. The estimation of unsaturated fatty acids by reversed-phase partition chromatography. Biochemical Journal, 59: 309-316.

Unsaturated fatty acids were separated on paraffin-impregnated kieselguhr using acetone-water mixtures for elution. Effects of isomerism and functional groups on the rates of elution are discussed.

Davenport, J. B.

1955. Column partition chromatography of the fatty hydroxamic acids. Chemistry and Industry (London) 705-706.

Fatty acid hydroxamates were separated on a cellulose column and identified by their melting points.

Dawson, R. M. C.

1954. A note on the estimation of sphingomyelin in nervous tissue. Biochemical Journal, 56: 621-625.

Lower values for sphingomyelin were obtained by the method of Schmidt, et al (Journal of Biological Chemistry, 166: 505, 1946) when the lipids in brain tissue were precipitated with trichloracetic acid prior to solvent extraction. The cause is believed to be the presence of phosphorus-containing materials which become insoluble in trichloracetic acid after alkaline hydrolysis and are therefore estimated as sphingomyelin.

DeLorenzi, F. and R. Aldrovandi

1952. Volumetric determination of phosphorous in phosphates, glycerophosphates, and lecithin with Complexon III. Farmaco, scienza e tecnica (Pavia) 7:

309-312. Chemical Abstracts, 46: 11036b (1952).

Phospholipid is precipitated with a measured amount of $MgCl_2 \cdot 6H_2O$ in the presence of NH_4 salt and NH_3, and excess Mg is titrated with Complexon III.

Delsal, J. L.

1944. New procedure for extraction of serum lipides with methylal. Application to microdetermination of total cholesterol, phosphoaminolipides and proteins. Bulletin de la Société de chimie biologique, 26: 99-105. Chemical Abstracts, 39:3560^9 (1945).

★

Extraction of serum cholesterol and lipids with methylal was found to be complete.

Delsal, J. L.

1946. Colorimetric microdetermination of free cholesterol. Bulletin de la Societé de chimie biologique, 28: 441-445. Chemical Abstracts, 41:4188a (1947).

■

A method is described for the determination of free cholesterol by precipitation with natigen and measurement by the Liebermann-Burchard reaction.

Delsal, J. L.

1950. Extraction of cholesterol from blood serum by chloroform in the presence of anhydrous sodium sulfate or plaster of paris. Compte rendu hebdomadaire des séances et mémoires de la Société de biologie, 144: 67-69. Chemical Abstracts, 44:10024f (1950).

★ ■

Cholesterol is not completely extracted from serum which is dehydrated with anhydrous sodium sulfate unless the serum is first hydrolyzed with NaOH.

Delsal, J. L. and Mme. DeMont-Argant

1950. Accord between the colorimetric and gravimetric (by natigin or digitonin) determinations of cholesterol in normal and pathological human serum. Anomaly observed with certain lipoid nephrosis serums. Compte rendu hebdomadaire des séances et mémoires de la Société de biologie, 144: 87-89. Chemical Abstracts, 44:10024g (1950).

■

Cholesterol values obtained by colorimetric microdetermination after saponification usually agree with those obtained by gravimetric determination.

Delsal, J. L.

1954. Fractionation of the lipides of blood serum by organic solvents. Bulletin de la Société de chimie biologique, 36: 1329-1334. Chemical Abstracts, 49:7628a (1955).

★

A methylal-methanol (4:1) mixture is used to precipitate protein and extract the lipids from a serum sample. Petroleum ether and water are added to the extract to cause the extract to separate into two phases. Sterides and triglycerides concentrate in the upper phase, and phosphoaminolipides in the lower.

Devis, R.

1944. Acidimetric determination of total fat acids in very small quantities of biological fluids and tissues. Bulletin de la Société de chimie biologique, 26: 232-238. Chemical Abstracts, 40:1190^7 (1946).

●

Lipids are hydrolyzed and acidified to free the fatty acids. The acids are extracted with benzene and titrated with KOH.

Dieckert, J. W. and R. Reiser

1956. A paper chromatographic procedure for separating 1-mono-, 1,3-di-, and triglycerides, cholesterol, and cholesterol esters. Journal of the American Oil Chemists Society, 33: 123-126. Chemical Abstracts, 50:7012g (1956).

★ ● ■

1-mono, 1,3-di-, and triglycerides, cholesterol, and cholesterol esters were separated by chromatography on silicic acid-impregnated glass fiber paper using ethyl ether-isooctane mixtures as solvents. Spots on the developed chromatogram were detected by spraying with 50% aqueous H_2SO_4 and gentle heating to detect cholesterol-containing lipids, followed by stronger heating to detect the glycerides. Degree of unsaturation of the glyceride fatty acids had no detectable effect on the R_f values of the triglycerides.

The glass paper was specially washed in MeOH and ethyl ether to remove impurities.

The dichromate-H_2SO_4 reagent used earlier by Fillerup and Mead (Proceedings of the Society for Experimental Biology and Medicine, 83: 574, 1953) gave yellow-orange background color, and did not permit differentiation between cholesterol- and non-cholesterol-containing lipids.

Dieckert, J. W. and R. Reiser
1956. Paper chromatography of phospholipides on silicic acid impregnated glass fiber filter paper. Journal of the American Oil Chemists Society, 33: 535-537.

★ ▲

Phospholipids were separated from their mixtures by chromatography on glass paper impregnated with silicic acid using 1:1 methanol-ethyl ether as solvent.

Dijkstra, G., J. G. Keppler, and J. A. Schols
1955. Gas-liquid partition chromatography. Recueil des travaux chimiques des Pays-Bas et de la Belgique, 73: 805-812. Chemical Abstracts, 50: 1528f (1956).

★ ●

A silicone grease- or paraffin-coated column of celite was used to separate mixtures of medium chain fatty acids, alcohols, and aldehydes. Other gas chromatographic systems for identification and determination of aliphatic materials are described.

Dittmer, J. C., J. L. Feminella, and D. J. Hanahan
1958. A study of the quantitative estimation of ethanolamine and serine in the phospholipids. Journal of Biological Chemistry, 233: 862-867.

▲

The phospholipids are hydrolyzed with 6 N HCl, and the free amines are separated by ion exchange. Ethanolamine and serine are estimated as NH_3 released by periodate oxidation of the amines.

Dole, V. P.
1956. A relation between non-esterified fatty acids in plasma and the metabolism of glucose. Journal of Clinical Investigation, 35: 150-154.

●

Non-esterified fatty acids are extracted with iso-propyl alcohol-heptane-1 N H_2SO_4 and titrated with 0.018 N NaOH. The method measures fatty acids in 1 cc. of plasma to ± 3% in replicate analyses.

Drekter, I. J., A. Bernhard, and J. S. Leopold
1935. The extraction of cholesterol from blood serum. Journal of Biological Chemistry, 110: 541-549.

■

It was found that alcohol extracts all of the cholesterol from blood serum, while ether only extracts a small part of it. Part of the cholesterol is presumably bound to protein which is denatured by the alcohol, thus allowing complete extraction with alcohol.

Drekter, I. J., A. E. Sobel, and S. Natelson
1936. Fractionation of cholesterol in blood by precipitation as pyridine cholesteryl sulfate and cholesterol digitonide. Journal of Biological Chemistry, 115: 391-399.

■

It was found that the pyridine sulfate method (Sobel, Journal of Biological Chemistry, 115: 381, 1936) gave free cholesterol values from 6 to 10% of the total, while for the cholesterol digitonide method the value was 25 to 35%. The theory is advanced that the digitonin method may give higher values due to splitting of loosely bound cholesterol or the precipitation of some form of combined cholesterol other than fatty acid esters.

Ducet, G. and E. Kahane
1946. Biochemistry of choline and its derivatives. XX. Identification and chemical determination of choline in biological substances. Bulletin de la Société de chimie biologique, 28: 794-805. Chemical Abstracts, 41 4188b (1947).

After decomposition of the tissue with HNO_3, choline is freed by hydrolysis and precipitated by I_2KI or phosphotungstic acid and determined in the precipitate.

Ducet, G.
1948. Separation and determination of the water-soluble forms of choline. Analytica Chimica Acta, 2: 839-843. (in French). Chemical Abstracts, 43: 7998b (1949).

★

Free choline is adsorbed on silica gel,

and the combined water-soluble choline in the effluent is hydrolyzed and determined separately as the periodide.

Duffie, M. J. and J. L. Guravich
1959. A comparison of the methods of Bloor and Schoenheimer-Sperry in the estimation of cholesterol in serum. American Journal of Clinical Pathology, 32: 92-96.

■ Serum cholesterol determinations by the Bloor (Journal of Biological Chemistry, 24: 227, 1916) and the Sperry and Webb modification (Journal of Biological Chemistry, 187: 97, 1950) of the Schoenheimer-Sperry method were done in parallel. The Bloor method yielded values which were 1.044 times as large as comparable values by the S-S method. The Bloor method appeared to be as accurate, and was faster than the S-S method.

Dutton, H. J. and C. L. Reinbold
1948. Adsorption analysis of lipides. III. Synthetic mixtures of ethyl stearate, oleate, linoleate, and linolenate. Journal of the American Oil Chemists Society, 25: 120-124. Chemical Abstracts, 42:3973f (1948).

★ ● Details are given of a study of adsorption analysis of binary mixtures of ethyl stearate, oleate, linoleate, and linolenate on alumina columns.

Edman, P. V.
1942. A micromethod for the estimation of cerebrosides in nerve tissue. Journal of Biological Chemistry, 143: 219-221.

The carbazole reaction of Dische (Mikrochemie, 8: 4, 1930) is used for the quantitative estimation of 0.2 to 0.6 mg. of cerebrosides by colorimetry. After hydrolysis of the cerebrosides, the fatty acids are removed by virtue of their insolubility in water, and glycerol is extracted into ethyl acetate. Noll's (Zeitschrift für physiologische Chemie, 27: 370, 1899), Kimmelsteil's (Biochemische Zeitschrift, 212: 359, 1929), and Kirk's (Journal of Biological Chemistry, 123: 613, 1938) methods for estimation of galactose set free from the cerebrosides by hydrolysis are criticized for their lack of specificity in reduction methods.

Edman, P. V. and S. E. G. Aquist
1945. A micromethod for the estimation of phosphatidyl ethanolamine in nerve tissue. Acta physiologica Scandinavica, 10: 144-149.

▲ Phosphatidyl ethanolamine is estimated as ethanolamine by hydrolysis of an alcoholic tissue extract, distillation of the ethanolamine, and colorimetric measurement of the blue color developed with sodium hypochlorite and phenol.

Edsgaard, J.
1948. On the determination of the phosphatide content of serum. Acta physiologica Scandinavica, 16: 171-178.

▲ An investigation of methods for the determination of serum phospholipid. It was found that absolute ethanol alone is as effective for the extraction of serum phospholipids as alcohol-ether (3:1) or alcohol-acetone (1:1). Temperature and time of extraction are not critical as long as 1:20 ratio of serum to solvent is maintained.

Edsgaard, J.
1948. On the colorimetric determination of phosphorus with "amidol". Acta physiologica Scandinavica, 16: 179-182.

▲ A discussion of the effects of variables in the determination of phosphorus with "amidol" (diaminophend hydrochloride) and molybdate. Acidity and temperature of color development were found to be important, but age of the reagents had little effect.

Ellis, G. and L. A. Maynard
1937. The determination of phospholipids in bovine blood. Journal of Biological Chemistry, 118: 701-709.

▲ The values for Bloor's oxidative technique (Journal of Biological Chemistry, 82: 273, 1929) were found to be variable and also 50% or more below values obtained by the lipid phosphorus method (method not stated). The values for phospholipid found in the final moist ether (or chloroform) solution after the purification steps of Bloor, however,

were in good agreement whether obtained by the oxidative procedure or by multiplying the phosphorus present by the factor 25. It was found that the temperature of evaporation of the alcohol-ether extract affects the solubility of the phospholipids in petroleum ether; therefore the use of a vacuum and temperature of less than 50° is recommended for the evaporation. $CHCl_3$ dissolves the phospholipid-$MgCl_2$ precipitate as well as moist ether, giving more consistent results and requiring less time for solution. Alcohol-ether extracts of plasma, when stored for several months gave ratios of lipid phosphorus, phospholipid, and phospholipid fatty acid no lower than extracts used a few days after preparation. See also: Boyd (Journal of Biological Chemistry, 121: 485, 1937) and Halliday (Journal of Nutrition, 16: 285, 1958).

Engel, R. W.

1942. Modified methods for the chemical and biological determination of choline. Journal of Biological Chemistry, 144: 701-710.

Choline was extracted from biological material by continuous extraction with methanol, which is claimed to be the most effective solvent (obtained recoveries within 1.5% of theoretical). (Brante (Acta physiologica Scandinavia, 8: supplement 63, 1949) states that ethanol-ether (3:1) is just as effective as methanol). The extract was concentrated and hydrolyzed with $Ba(OH)_2$ at 100° C. (100° was used since the temperature of 80° suggested by Jacobi (Journal of Biological Chemistry, 138: 571, 1941) failed to give consistent results.) Choline was determined as the reineckate by colorimetry. The incidence of kidney hemmorhage in rats was also used as an indication of the quantity of choline and choline-like materials in biological test diets, and showed sensitivity to a variation of 10% of the materials.

Engel, R. W., W. D. Salmon, and C. J. Ackerman

1954. Chemical estimation of choline. In Methods of Biochemical Analysis, D. Glick, Editor, New York, Interscience Publishing, Inc., Vol. 1, pp. 265-286.

A study and discussion of various methods for the estimation of choline in tissues.

Entenman, C., A. Taurog, and I. L. Chaikoff

1944. The determination of choline in phospholipids. Journal of Biological Chemistry, 155: 13-18.

The phospholipids were hydrolyzed with $Ba(OH)_2$ and the choline was precipitated as the reineckate and read colorimetrically. Accuracy ± 3% using choline chloride.

1.2 N HCl was used as solvent for ammonium reineckate, making it possible to use a more concentrated reineckate solution. Precipitation is complete in 30 minutes.

Entenman, C. and I. L. Chaikoff

1945. On the determination of choline in the liver and plasma of the dog. Journal of Biological Chemistry, 160: 377-385.

Various extracts and fractions of plasma and liver were analyzed for choline by the methods of Glick (Journal of Biological Chemistry, 156: 643, 1944) and of Entenman, et al (Journal of Biological Chemistry, 155: 13, 1944). Identical choline values were found for the phospholipid fraction isolated from either liver or plasma when the precipitations of choline reineckate were carried out in the presence of 1.2 N HCl (Entenman, et al) or at a pH between 8 and 9 (Glick). Both methods gave values in agreement on analyses of both alcohol-ether and methanol extracts of plasma, but Glick's method gave lower values for an alcohol-ether extract of liver.

Erickson, B. N., I. Avrin, D. M. Teague, and H. H. Williams

1940. Micromethods for the determination of sphingomyelin and choline. Applications for the estimation of the phospholipid partition (sphingomyelin, lecithin, and cephalin) in blood and urine. Journal of Biological Chemistry, 135: 671-684.

A procedure is described in which sphingomyelin is precipitated as the reineckate and calculated from the phosphorus content of the reineckate. Choline is determined, after

hydrolysis of the phospholipids, by precipitation as the enneaiodide, conversion of the iodide to iodate with bromine, and titration with sodium thiosulfate. Comparison of $Ba(OH)_2$ and methanolic HCl hydrolysis techniques showed nearly identical values with phospholipids of approximately 25% sphingomyelin content. With those of over 50% sphingomyelin, the methanolic HCl method gave values 10% higher.

Fairbairn, D.

1945. Free fatty acids in animal tissues. ● Journal of Biological Chemistry, 157: 645-650.

A rapid loss of phospholipids occurs shortly after tissue is removed due to autolysis (approximately 8% in a few minutes in whole cat liver; approximately 15% in ground liver). Homogenization in alcohol or fast-freezing decreases autolysis.

Farvarger, P. and J. Gerlach

1958. A quantitative evaluation of Twitchell's method for the separation of saturated and unsaturated fatty acids. ★ ● Archives des sciences (Geneva), 11: 539-542. Chemical Abstracts, 53: 19075h (1959).

Twitchell's lead salt operation method (Journal of Industrial and Engineering Chemistry, 13: 806, 1921) was used to separate the fatty acids of the mouse into saturated and unsaturated fractions. Recoveries were followed by addition of C^{14} labelled fatty acids. Recoveries ranged from 84.2 to 95.7%.

Faure, M.

1950. Method for purification of lecithins. ★ ▲ Bulletin de la Société de chimie biologique, 32: 503-508. Chemical Abstracts, 45:695h (1951).

Lecithin was purified by precipitation with $CdCl_2$, removal of Cd, precipitation from ether with Me_2CO, extraction with ether, evaporation, and chromatography of the ether solution on alumina.

Feichtmeir, T. V. and J. Bergerman

■ 1953. Indirect colorimetric determination of cholesterol. American Journal of Clinical Pathology, 23: 599-602.

Cholesterol is precipitated as the digitonide, and the digitonide is quantitatively determined by measurement of the color produced with anthrone reagent.

Feulgen, R. and H. Grunberg

1939. Estimation of plasmal (plasmalogen) in lipoid mixtures and in organs. ▲ Zeitschrift für physiologische Chemie, 257: 161-172. Chemical Abstracts, 33:3407[9] (1939).

Plasmalogens in lipid mixtures were determined by photometric measurement of the color produced with fuchsin-sulfurous acid reagent.

Feulgen, R., W. Boguth, and G. Anderson

1951. Determination of acetal phosphatide (plasmalogen) in serum with regard to the Waelsch effect. ▲ Zeitschrift für physiologische Chemie, 287: 90-108. Chemical Abstracts, 48:4621g (1954).

Acetal phosphatide of serum is determined by hydrolysis with HCl in an acetic acid solution, neutralization with NaOH, and development of color with SO_2-reduced fuchsin.

Fillerup, D. L. and J. F. Mead

1953. Chromatographic separation of the plasma lipids. ★ ● Proceedings of the Society for Experimental Biology and Medicine, 83: 574-577.

Plasma lipids were separated into groups by chromatography on silicic acid with ethyl ether-petroleum ether and methanol-ethyl ether as eluting solvents.

Fink, K. and R. M. Fink

1949. Application of filter paper partition chromatography to quantitative analysis of volatile and non-volatile ★ ● organic acids. Proceedings of the Society for Experimental Biology and Medicine, 70: 654-656.

The C_1-C_8 acids are chromatographed on filter paper as their potassium hydroxamates and located on the developed chromatogram by spraying with ferric chloride solution.

Fiske, C. H. and Y. Subbarow

▲ 1925. The colorimetric determination of

phosphorus. Journal of Biological Chemistry, 66: 375-400.

Aminonaphtholsulfonic acid is used for reduction of phosphomolybdic acid in the presence of bisulfite in place of the hydroquinone used in earlier methods. Effects of interfering substances and variations in reagent concentrations on the development of color are discussed.

The method is suitable for colorimetric determination of inorganic and total phosphate in a variety of biological materials.

Fitelson, J.

1950. A new oxidation method for the determination of saturated fatty acids. Journal of the American Oil Chemists Society, 27: 1-8. Chemical Abstracts, 44:2259f (1950).

Saturated fatty acids are purified by chromatography on alumina after oxidation of the unsaturated acids with performic acid and extraction of the oxidation products with petroleum ether.

Fleckenstein, A., E. Gerlach, and J. Janke

1953. Rapid test for identification of easy and difficultly hydrolyzable phosphoric esters in paper chromatogram. Naturwissenschaften, 40: 462. Chemical Abstracts, 48:8124g (1954).

The chromatogram is sprayed with molybdate reagent and exposed to H_2S, which turns spots containing P to a blue color. Exposing the paper to NH_3 vapor causes the color to disappear from the spots caused by phosphoric esters which are difficult to hydrolyze, while the spots from inorganic P or readily hydrolyzed esters remain blue.

Fleury, P. and R. Paris

1933. A comparison of the action of periodic acid on the α and β glycerophosphoric acids. Compte rendu, 196: 1416-1418. Chemical Abstracts, 27: 3915 (1933).

A confirmation of earlier findings that oxidation by HIO_4 of a compound having several alcoholic groups does not take place unless these groups are adjacent.

Fleury, P. and H. Guitard

1948. Determination of choline in lecithin. Annales pharmaceutiques francaises, 6: 252-254. Chemical Abstracts, 43:5344h (1949).

Choline is determined colorimetrically as the reineckate.

Fleury, P., J. Courtois, and M. Grandchamp

1949. Estimation of mixtures of colamine and serine. Biochimica et biophysica Acta, 3: 336-340. (In French).

Ethanolamine and serine are estimated by determination of the NH_3 liberated and the amount of periodic acid reduced during 48 hours of standing at room temperature.

Folch, J. and D. D. van Slyke.

1939. Nitrogenous contaminants in petroleum extracts of plasma lipids. Journal of Biological Chemistry, 129: 539-546.

Urea is the primary N-containing contaminant of petroleum extracts of plasma lipids. Although urea is insoluble in petroleum ether, it is dissolved in a lipid extract of petroleum ether, presumably due to the solvent effect of the lipids.

Folch, J. and D. D. van Slyke

1939. Preparation of blood lipid extracts free from non-lipid extractives. Proceedings of the Society for Experimental Biology and Medicine, 41: 514-515.

Proteins and lipids are precipitated together with colloidal iron and washed with H_2O, and the lipids are extracted with alcohol-ether. Brante (Acta physiologica Scandinavica, 18 (supplement 63): 1, 1949), and Sperry (Journal of Biological Chemistry, 170: 675, 1947) found that a considerable amount of lipid material is lost by this method of purification.

Folch, J.

1941. Isolation of phosphatidyl serine from brain cephalin and identification of the serine component. Journal of Biological Chemistry, 139: 973-974.

Phosphatidyl serine is separated from brain cephalin by precipitation of the cephalin in alcohol-chloroform, and the

serine component is removed by 30 hour HCl hydrolysis and crystallization from absolute alcohol.

Folch, J.
1942. Brain cephalin, a mixture of phosphatides. Separation from it of phosphatidyl serine, phosphatidyl ethanolamine, and a fraction containing an inositol phosphatide. Journal of Biological Chemistry, 146: 35-43.
★ ▲

Phosphatidyl serine, phosphatidyl ethanolamine, and an inositol phosphatide were separated by their differences in solubility in chloroform and alcohol.

Folch, J. and D. W. Woolley
1942. Inositol, a constituent of brain phosphatide. Journal of Biological Chemistry, 142: 963-964.
▲

Inositol is separated by precipitation and HCl hydrolysis. Inositol is found in brain in combined form.

Folch, J.
1948. The chemical structure of phosphatidyl serine. Journal of Biological Chemistry, 174: 439-450.
★ ▲

A method is described for the isolation of phosphatidyl serine of at least 92% purity by means of precipitation, dialysis, and solvent recrystallization procedures.

Folch, J.
1949. Complete fractionation of brain cephalin; isolation from it of phosphatidyl serine, phosphatidyl ethanolamine, and diphosphoinositide. Journal of Biological Chemistry, 177: 497-504.
★ ▲

The cephalin fractions are separated by their different solubilities in organic solvents.

Folch, J., I. Ascoli, M. Lees, J. A. Meath, and F. N. LeBaron
1951. Preparation of lipide extracts from brain tissue. Journal of Biological Chemistry, 191: 833-841.
★

Lipid extracts are prepared by homogenizing brain tissue with $CHCl_3$-MeOH (2:1), filtering to remove insoluble matter, and washing with water. The method was compared with those of Brante (Acta physiologica Scandinavica, 18 (supplement 63): 1, 1949) and McKibbin and Taylor (Journal of Biological Chemistry, 178: 17, 1949). It was found that this method extracted more strandin and proteolipids than the other methods, but that the extraction of other lipids and removal of non-lipid contaminants from the extracts were essentially the same for all three methods.

Sperry (Journal of Biological Chemistry, 209: 377, 1954) found no significant difference in his own method and Folch's, but his final material was completely soluble in chloroform-methanol (2:1) while Folch's method left a small insoluble residue.

Folch, J., M. Lees, and G. H. Sloane-Stanley
1957. A simple method for the isolation and purification of total lipids from animal tissues. Journal of Biological Chemistry, 226: 497-509.
★ ●

A simplification of the procedure of Folch, et al (Journal of Biological Chemistry, 191: 833, 1951).

Tissue lipids are extracted with $CHCl_3$-MeOH (2:1) and the extract is purified by washing with aqueous salt solutions.

Forbes, J. C. and T. T. Atkinson, Jr.
1943. The separate determination of the fatty acid fraction and of the neutral fat plus sterol fraction in feces. Journal of Laboratory and Clinical Medicine, 28: 1507-1510.
●

The fat is extracted and is separated into free fatty acid and neutral fat fractions by precipitation of the free fatty acids as their Na soaps. The separate portions are then determined by oxidation with silver chromate and iodometric measurement of excess dichromate.

Foreman, H. D. and J. B. Brown
1944. Solubilities of the fatty acids in organic solvents at low temperatures. Oil and Soap, 21: 183-187. Chemical Abstracts, 38: 4821[8] (1944).
●

Data is given on the solubility in several organic solvents of various saturated and unsaturated fatty acids at temperatures

ranging from 10° to -70°.

Foulger, J. H.
1932. Two new color tests for hexoses. Journal of Biological Chemistry, 99: 207-211.

Urea or guanidine is used in a $SnCl_2$ - H_2SO_4 solution to give a color reaction with simple hexoses. The urea reagent differentiates between aldo- and keto-hexoses, and the guanidine reagent gives a distinct color for each sugar but does not distinguish between aldo- and keto-hexoses. Sensitivity varies from 0.02 to 0.5 mg. depending on the sugar tested.

Frampton, V. L., R. D. Maseles, and G. N. Martin
1948. Lipides of the cottonseed. III. Perchloric acid ashing of lipides for the determination of phosphorus. Journal of the American Oil Chemists Society, 25: 219-220. Chemical Abstracts, 42:5689f (1948).

▲

The presence of $HClO_4$ does not interfere with development of molybdenum blue color in the determination of phosphorus by the Fiske-Subbarow method.

Franzke, C. and G. Ittrich
1947. Determination of linoleic and linolenic acid by means of bromine addition. Fette, Seifen, Anstrichmittel, 59: 740-744. Chemical Abstracts: 53:18512i (1959).

★ ●

Linoleic and linolenic acids are separated as their bromine addition products by using the difference in their solubilities in petroleum ether.

Freeman, N. K., F. T. Lindgren, Y. C. Ng, and A. V. Nichols
1957. Serum lipide analysis by chromatography and infrared spectrophotometry. Journal of Biological Chemistry, 227: 449-464.

★ ●

Lipids are separated into three fractions on a silicic acid-Celite column, and cholesteryl esters, glycerides, total phosphatides, cholesterol, and free fatty acids are estimated by infrared absorption measurements. A 1 ml. sample of serum (5-10 mg. of total lipids) is used. A component of at least 0.5 mg. is estimated to ± 10%.

Fries, J., A. Holasek, and H. Lieb
1956. Detection of unsaturated fatty acids on a paper chromatogram. Mikrochimica Acta, pp 1722-1726. Chemical Abstracts, 51:4215b (1957).

●

Unsaturated fatty acids on a paper chromatogram are made visible by exposing the chromatogram to ozone and spraying with fuchsin-sulfurous acid.

Frisell, W. R., L. A. Meech, and C. G. MacKenzie
1954. A simplified photometric analysis for serine and formaldehyde. Journal of Biological Chemistry, 207: 709-716.

The formaldehyde formed by oxidation of serine with periodate is measured by direct photometry without distillation.

Fugger, J., J. A. Cannon, K. T. Zilch, and H. J. Dutton
1951. Analysis of fat acid oxidation products by countercurrent distribution methods. IV. Methyl linolenate. Journal of the American Oil Chemists Society, 28: 285-289. Chemical Abstracts, 45:10617i (1951).

★ ●

The oxidation products of methyl linolenate were fractionated by countercurrent distribution with aqueous ethanol and hexane as solvents.

Galloway, L. S., P. W. Nielson, E. B. Wilcox, and E. M. Lantz
1957. Micro-determination of cholesterol by use of 0.04 ml. of blood serum. Clinical Chemistry, 3: 226-232.

■

A micro-adaptation of the method of Sperry and Webb (Journal of Biological Chemistry, 187: 97, 1950).

Garton, G. A. and A. K. Lough
1957. Analysis of mixtures of higher saturated normal fatty acids: A comparison of reversed-phase partition chromatography and ester fractionation. Biochimica et Biophysica Acta, 23: 192-195.

★ ●

The odd-numbered n-fatty acids were

separated on a dichlorodimethylsilane-treated column of silicic acid. Results compared well with ester fractionation methods.

Garvin, J. E. and M. L. Karnovsky
1955. Nonaqueous titration of lipids, with particular reference to phosphatides and related compounds. Proceedings of the International Conference on Biochemical Problems of Lipids, 2nd Ghent, (Pub. 1956) pp. 14-16.

The use of 99% ethoxyethanol as a solvent for titration of small amounts of fatty acids is discussed.

Gertler, M. M., J. Kream, and O. Baturay
1954. Studies on the phosphatide content of human serum. Journal of Biological Chemistry, 207: 165-173.

Phosphatides were extracted from serum and hydrolyzed with 6 N HCl for 48 hours at 100°, and the free bases were separated by paper chromatography. Serine and ethanolamine were located on the chromatogram with ninhydrin and extracted with pyridine. The optical density of the pyridine solutions was determined spectrophotometrically at 580 mμ. Choline was treated on the paper with phosphomolybdic acid-stannous chloride and the spot areas were measured by planimetry.

Gibble, W. P., E. B. Kurtz, Jr., and A. E. Kelley
1956. A semi-micro procedure for the separation and degradation of long-chain fatty acids. Journal of the American Oil Chemists Society, 33: 66-68. Chemical Abstracts, 50:5309b (1956).

Total lipid is extracted with petroleum ether and saponified by refluxing for 6 hours with 12% alcoholic KOH. The fatty acids are freed with HCl and extracted with ether. The ether is evaporated and the fatty acids are dissolved in acetone and separated by low temperature crystallization. Unsaturated fatty acids are hydrogenated using PtO_2 in absolute ethanol. Saturated fatty acids are degraded using thionyl chloride, pyridine, and $AlCl_3$.

Glick, D.
1944. Concerning the reineckate method for the determination of choline. Journal of Biological Chemistry, 156: 643-651.

No significant difference in result was obtained by the purification of neutralized $Ba(OH)_2$ hydrolysates of wheat germ extracts by ether extraction or by adsorption of the choline on Permutit and elution with salt solution according to Horowitz and Beadle (Journal of Biological Chemistry, 150: 325, 1943). Choline reineckate was washed free of excess Reinecke salt with n-propanol.

Goddu, R. F., N. F. LeBlanc, and C. N. Wright
1955. Spectrophotometric determination of esters and anhydrides by hydroxamic acid reaction. Analytical Chemistry, 27: 1251-1255.

A method is described for determination of esters by conversion to their hydroxamic acids and spectrophotometric measurement of the color produced by the ferric complexes of the acids. The effects of variables are discussed.

Goodwin, J. F.
1959. Total, phospholipide, and labile phosphorus in serum and tissue employing chloric acid and n-phenyl-p-phenylenediamine. Proceedings of the Society for Experimental Biology and Medicine, 100: 217-219.

A colorimetric method is described for determination of the phosphorus fractions of serum. Chloric acid is used for digestion, and the phosphomolybdate complex is reduced with p-semidine. The method has the advantages of ease of digestion and color development, and stability of the color complex. It compares well with the Fiske-Subbarow method.

Gorbach, G. and A. Jurinka
1944. Micromethods for fats. IX. Higher saturated fatty acids by the Bertram method. Fette und Seifen, 51:171-173. Chemical Abstracts, 42:9204f (1948).

A description of micromodifications of Bertram's method (Zeitschrift für Untersuchung der Lebensmittel, 55: 179, 1928).

Gortner, W. A.

1945. An evaluation of micromethods for phospholipid. Journal of Biological Chemistry, 159: 97-100.

▲

Analyses of tissue by determination of lipid phosphorus, by oxidation of the fatty acids from acetone-insoluble lipids, and by direct oxidation of the intact phospholipid are compared.

When a phospholipid to phosphorus ratio of 24 is used, and a fatty acid recovery of 63% of the weight of the saponified phospholipid was assumed, the three methods gave comparable results. Phosphorus analysis, however, gave better agreement among replicate samples (error $\pm$ 0.8%) than either of the oxidative procedures studied (error $\pm$ 2.2% and $\pm$ 2.1%).

The author cautions against the use of conversion factors on mixtures of different types of phospholipid.

Gracian y Tous, J. and A. V. Pizarro

1947. Separation of higher fat acids by selective adsorption. Anales de fisica y quimica (Madrid), 42: 109-122. Chemical Abstracts, 41:5323i (1947).

★ ●

Alumina reacted chemically with oleic and stearic acids, making their separation on alumina difficult. Silicic acid was satisfactory as an adsorbent for the acids.

Graff, M. M. and E. L. Skau

1943. Colored chromatograms with higher fatty acids. Industrial and Engineering Chemistry, Analytical Edition, 15: 340-341

★ ●

Fatty acids were separated into zones by adsorption on magnesium oxide impregnated with phenol red. The column was sectioned after development and the fatty acids were recovered by dissolving the MgO in acid and extracting the fatty acids with ether. Saturated and unsaturated fatty acids of the same chain length and saturated fatty acids differing in chain length by 4 carbons were separable by the method.

Griswold, B. L., F. L. Humoller, and A. R. McIntyre

1951. Inorganic phosphates and phosphate esters in tissue extracts. Analytical Chemistry, 23: 192-194.

▲

A modification of the Fiske-Subbarow (Journal of Biological Chemistry, 66: 375, 1925) method for reduction of phosphomolybdic acid with aminonaphtholsulfonic acid which uses the heating in 1 N H_2SO_4 recommended by Boltz and Mellon (Analytical Chemistry, 19: 873, 1947) for color development. The higher acidity prevents color formation from reduction of free molybdic acid as is possible in the Fiske-Subbarow method.

Gurin, S. and D. B. Hood

1939. The identification and estimation of hexoses in polysaccharides and glycoproteins by the carbazole method. Journal of Biological Chemistry, 131: 211-223.

The carbazole reaction is used for quantitative identification of glucose, fructose, galactose, and mannose, or mixtures of these hexoses, by colorimetric measurement of the colors produced in sulfuric acid. Galactose and an equimolar glucose-mannose mixture give nearly identical values, so a supplementary test is necessary in these cases.

Hack, M. H.

1946. Some observations concerning sphingomyelin and sphingomyelin reineckate. Journal of Biological Chemistry, 166: 455-462.

▲

Reineckates prepared from crude extracts contained both glycerol and hexose, implying the presence of other lipids than sphingomyelin. The MeOH-soluble fraction of cephalin and the cerebrosides kerasin and phrenosin formed reineckates. Recovery of pure sphingomyelins as the reineckates was low.

Values obtained by isolation of the sphingomyelin were too low; and those obtained by the Reineckate method were too high, but there was general agreement in the two methods.

Hack, M. H.

1947. Estimation of the phospholipides in human blood. Journal of Biological Chemistry, 169: 137-143.

▲

Lipids are extracted from 15 ml. of blood

with $CHCl_3$MeOH (1:1) and total phospholipid is determined on an aliquot of the extract. After saponification with 1 N KOH for 16 hours at 37° and acidification, choline is precipitated as the reineckate and determined colorimetrically at 526 mμ. (Beer's Law did not hold at 327 mμ. under the conditions studied.) Sphingomyelin is calculated as the difference in total phosphorus and that freed by hydrolysis. Lecithin is calculated from the liberated choline, and cephalin is calculated as the liberated phosphorus minus lecithin.

Maximum precipitation of choline reineckate requires freshly prepared ammonium reineckate.

Hack, M. H.

1953. Analysis of lipids by spot tests on filter-disk chromatograms. Biochemical Journal, 54: 602-605.

▲ ■

Spot tests are given for detection of quantities of 10^{-2} μmoles or less of choline lipids, amine lipids, plasmalogens, phosphoric esters, cholesterol, glycolipids, and unsaturation on filter-disc chromatograms.

Hack, M. H.

1955. A method for the estimation of fatty acid esters. Archives of Biochemistry, 58: 19-23.

●

A micro modification of the method of Bauer and Hirsch (Archives of Biochemistry, 20: 242, 1949).

The color produced by reaction of the hydroxamic acids from fatty acid esters with ferric perchlorate is determined colorimetrically. Data on specificity and sensitivity are given. Estimates 0.2 to 3 μmoles of ester in a few minutes.

Halliday, N.

1938. Fatty livers in vitamin B_6 deficient rats. Journal of Nutrition, 16: 285-290.

●

Storage of lipid extracts at room temperature resulted in losses of total fatty acids and phospholipid fatty acid. No change occurred in total cholesterol and a definite drop occurred in all iodine numbers.

See also: Boyd (Journal of Biological Chemistry, 121: 485, 1937).

Halliday, N.

1939. The effect of formalin fixation on liver lipids. Journal of Biological Chemistry, 129: 65-69.

A drop in iodine value and phospholipid fatty acids occurred in lipids stored in formalin. There was little loss of total fatty acids during periods of up to three months, but longer storage caused a considerable loss.

Hamilton, J. G. and R. T. Holman

1954. Displacement analysis of lipids. X. Model mixtures of glycerides. Journal of the American Chemical Society, 76: 4107-4109.

★ ●

Glyceride mixtures were separated on a Darco G-60 charcoal-Hyflo Super-Cel column, using ethanol and benzene as eluting solvents.

Hanahan, D. J., M. B. Turner, and M. E. Jayko

1951. The isolation of egg phosphatidyl choline by an adsorption column technique. Journal of Biological Chemistry, 192: 623-628.

★ ▲

Phosphatidyl choline is eluted from an alumina column with 95% ethanol as eluent. Rechromatography of the purer fractions yields a product of 99-100% pure phosphatidyl choline.

Hanahan, D. J., J. C. Dittmer, and E. Warashina

1957. A column chromatographic separation of classes of phospholipids. Journal of Biological Chemistry, 228: 685-700.

★ ▲

A method is described for separation of the mixed phospholipids by elution with mixtures of chloroform and methanol from a single silicic acid column. Recovery of lipid phosphorus is 88-95%, with 90-95% of inositol-containing phospholipids being eluted in one fraction. Reproducibility is good.

The use of gradient elution caused smearing of fractions. Cotton, when used as support for the silicic acid, adsorbed approximately 20% of the phospholipids, particularly the phosphoinositides, which were difficult to remove from the cotton.

An almost complete reversal in the elution pattern and overlapping of the eluted components occurred when the Hyflo Super-Cel filter aid was heated to 110° before use.

Hirsh (Journal of Biological Chemistry, 233: 213, 1958) also found that abrupt change of solvent yielded better separations than gradient elution.

Handler, P.

1947. The determination of choline in biological material. Biological Symposia, 12: 361-372. Chemical Abstracts, 41:5117g (1947).

Choline-containing lipid is extracted from tissue with methanol and hydrolyzed with $Ba(OH)_2$. The choline is precipitated as the reineckate and the precipitate is dissolved in acetone and measured colorimetrically.

Handschumaker, E. and L. Linteris

1947. A modified method for the determination of monoglyceride in fats and oils by oxidation with periodic acid. ● Journal of the American Oil Chemists Society, 24: 143-145. Chemical Abstracts, 41:4659e (1947).

Monoglycerides are determined by periodic acid oxidation and iodometric measurement of excess periodic acid.

Hanel, H. K. and H. Dam

1955. Determination of small amounts of total cholesterol by the Tschugaeff reaction with a note on the determination of lathosterol. ■ Acta chemica scandinavia, 9: 677-682.

A method is described for simultaneous determination of cholesterol and lathosterol (Δ-7-cholesterol). After hydrolysis with KOH and extraction with petroleum ether, the cholesterol in chloroform solution is treated with $ZnCl_2$ in acetyl chloride and measured in a spectrophotometer at 528mμ. Lathosterol is read at 395 mμ., and gives almost no absorbance at 528 mμ.

Hansen, P. W. and H. Dam

1957. Paper chromatography and colori- ★ metric determination of free and ester- ■ ified cholesterol in very small amounts of blood. Acta chemica scandinavia, 11: 1658-1662.

Cholesterol is separated by paper chromatography from substances which might interfere with color development by the method of Zlatkis, Zak, and Boyle (Journal of Laboratory and Clinical Medicine, 41: 486, 1958). The method can determine as little as 2.5 μg. of cholesterol and its esters with accuracy of ±5%.

Harding, V. J. and C. E. Downs

1933. Notes on a Schaffer-Somogyi copper reagent. Journal of Biological Chemistry, 101: 487-492.

A modified Somogyi copper reagent (Journal of Biological Chemistry, 70: 599, 1926) is recommended as a good general sugar reagent. The reagent is prepared in two parts, and keeps for at least 3 months in the cold. Ammonium salts which interfere in the method are removed with $HgSO_4$ and $BaCO_3$.

Harris, W. D. and P. Popat

1954. Determination of the phosphorus content of lipids. Journal of the Amer- ▲ ican Oil Chemists Society, 31: 124-127. Chemical Abstracts, 48:6716g (1954).

A method is described for determination of lipid phosphorus using perchloric acid digestion and elon(p-methylaminophenolsulfate) as reducing agent for development of molybdate color. Effects of variables and disadvantages inherent in other methods are discussed.

Hartman, L.

1953. Rapid determination of glycerol by the potassium periodate method. Journal of Applied Chemistry, (London) 3: 308-311. Chemical Abstracts, 48: 79i (1954).

Complete oxidation of glycerol by KIO_4 takes place in 3 minutes when an excess of solid KIO_4 is used.

Hartman, L.

1954. Determination of fat peroxides in the presence of phospholipides. Jour- ● nal of the Science of Food and Agriculture, 5: 476-481. Chemical Abstracts,

49:2756i (1955).

Peroxides were determined by reduction with $FeCl_2$ in a benzene-methanol solution and determination of the Fe^{3+} produced by adding 2, 6-dichloroindophenol and $H_4P_2O_7$.

Hartman, L.

1955. Analysis of glycerol. Chemistry and Industry (London) pp. 1407-1408. Chemical Abstracts, 50:2370a (1956).

The glycerol sample is oxidized with $NaIO_4$, the excess $NaIO_4$ is reduced with propylene glycol, and the formic acid formed is measured by iodometric titration.

Hartman, L.

1956. Methanolysis of triglycerides. Journal of the American Oil Chemists Society, 33: 129, 1956. Chemical Abstracts, 50:6813i (1956).

Methyl esters were prepared by using sodium methoxide with ignited potassium carbonate as catalyst. This method was found to set free 90-95% of the total glycerol and cause the least degree of saponification of the various catalysts tested.

Hartmann, S. and J. Glavind

1948. A new sensitive method for the chemical determination of organic peroxides. Acta physiologica Scandinavica, 16 (supplement 53): 32-33.

The leuco base of 2, 6-dichlorophenolindophenol in acidified butanol is added to a xylene solution of fatty acids and the mixture is heated. The color developed by the reaction is measured spectrophotometrically and the peroxide value is calculated .

Hartmann, S. and J. Glavind

1948. A new sensitive method for the determination of peroxides of fats and fatty acids. Acta chemica scandinavia, 3: 954-958.

A method is described for the determination of peroxides in which dichlorodihydroxyphenylenediamine is oxidized by heating with a xylene-acetic acid solution of the peroxidized fat. The dichlorophenolindophenol which is quantitatively formed is measured colorimetrically. The reaction appears to be quantitative and specific.

Haven, F. L. and L. R. Levy

1941. The occurrence and rate of turnover of tumor sphingomyelin. Journal of Biological Chemistry, 141: 417-425.

Sphingomyelin was determined as the reineckate.

Little difference in results was found whether sphingomyelin was reextracted from the residue of Bloor's extract with petropeum ether or chloroform. More sphingomyelin was extracted when an additional extraction with chloroform-methanol was used than by a single alcohol-ether extraction. Radioactive phosphorus was used to track the sphingomyelin turnover.

Hawthorne, J. N. and G. Hübscher

1959. Separation of glycerylphosphoryl inositol and related compounds on ion-exchange columns. Biochemical Journal, 71: 195-200.

The phospholipid is hydrolyzed with NaOH and the hydrolysis products are separated by ion exchange chromatography.

Helrich, K. and W. Rieman, III

1947. Determination of acetyl number of fats and oils. Analytical Chemistry, 19: 691.

A simplification of the method of Roberts and Schuette (Industrial and Engineering Chemistry, Analytical Edition, 4: 257, 1932).

Henley, A. A.

1957. The determination of serum cholesterol. Analyst, 82: 286-287.

A modification of the method of Zlatkis, Zak, and Boyle which avoids protein interference is described. Proteins are precipitated by adding the serum to a stable ferric chloride-acetic acid reagent, and removed by centrifuging. The color is developed on a portion of the protein-free extract by adding H_2SO_4.

Hepburn, J. S. and R. Kotlikoff

1943. Comparative study of certain methods for the determination of serum cholesterol. Review of Gastroenterology, 10: 170-171. Chemical Abstracts, 38: 5864[6](1944).

The methods of Bloor (Journal of Biological

Chemistry, 24: 227, 1916), Myers and Wardell (Journal of Biological Chemistry, 36: 147, 1918), and Reinhold and Shiels (American Journal of Clinical Pathology, 6: 22, 1936) were studied. Results from the Bloor and Reinhold and Shiels methods agreed more closely than the Bloor and Myers and Wardell methods.

Herb, S. F. and R. W. Riemenschneider
1952. Influence of alkali concentration and other factors on the conjugation of natural polyunsaturated acids as determined by ultraviolet absorption measurements. Journal of the American Oil Chemists Society, 29: 456-461. Chemical Abstracts, 47:888e (1953).

Optimum conditions for maximum conjugation of methyl arachidonate were found to be 15 minutes heating at 180°C in 21% KOH-glycerol. Other acids treated accordingly provided greater sensitivity in the spectrophotometric method.

Herb, S. F. and R. W. Riemenschneider
1953. Spectrophotometric micromethod for determining polyunsaturated fatty acids. Analytical Chemistry, 25: 953-955.

A method is described for determination of fatty acids with 2-5 double bonds in a 1-10 mg. sample of fat. The acids are isomerized, and the density of a methanolic solution of the isomerized acids is measured spectrophotometrically. Results by the method agree with macromethods.

Herbain, M.
1959. Estimation of blood cholesterol by colorimetry, with the use of blood coagulant. Process improvements, particularly in the case of high hyperlipemias. Bulletin de la Société de chimie biologique, 41: 821-833. Chemical Abstracts, 53:22188e (1959).

Several modifications of the Sperry and Webb method for cholesterol determination are proposed.

Hess, W. C.
1947. Chromatographic separation of cholesterol and cholesterol esters in blood. Journal of Laboratory and Clinical Medicine, 32: 1163-1168.

Cholesterol esters and cholesterol are separated by adsorption on alumina and elution with 10% ethyl ether in petroleum ether and 10% ethanol in petroleum ether, respectively. The esters are saponified, and cholesterol is determined in both fractions by the Liebermann-Burchard method.

Hill, U. T.
1946. Colorimetric determination of fatty acids and esters. Industrial and Engineering Chemistry, Analytical Edition, 18: 317-319.

A method is described for determination of fatty acids and esters by conversion to their hydroxamic acid derivatives and colorimetric measurement of the color produced with Fe^{+++}. The fatty acids are methylated before treatment. Accuracy of the determinations was within $\pm$ 0.01 mg. of cottonseed oil.

Hill, U. T.
1947. Colorimetric determination of fatty acids and esters. Analytical Chemistry, 19: 932-933.

A modification of Hill's method (Industrial and Engineering Chemistry, Analytical Edition, 18: 317, 1946) which gives a more stable color.

Hirsch, J. and E. H. Ahrens, Jr.
1958. The separation of complex lipide mixtures by the use of silicic acid chromatography. Journal of Biological Chemistry, 233: 311-320.

Complex lipid mixtures are separated into chemical classes by elution from a single silicic acid column. Synthetic mixtures and plasma lipids were used to test the method.

Hodgson-Jones, I. S. and V. R. Wheatley
1952. Studies of sebum. 3. Methods for the collection and estimation of small amounts of sebum. Biochemical Journal, 52: 460-464.

Various methods of collection and estimation of sebum are discussed. Carbon tetrachloride was found to be more suitable than ether, ethanol, or acetone for removal of

sebum from the skin. Gravimetry, nephelometry, and chromate oxidation were found to be suitable methods for estimation of the sebum collected.

Holasek, A. and K. Winsauer

1954. Separation of saturated fatty acids by paper chromatography. Monatshefte, 85: 796-801. Chemical Abstracts, 49: 2757a (1955).

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C_4-C_{18} saturated fatty acids were separated on a potassium alum-impregnated filter paper using CCl_4-MeOH-conc. NH_4OH (81:18:1) as developer. They were located by spraying with Rhodamine B in HCl and viewing under UV light.

Holman, R. T. and G. O. Burr

1948. Alkali conjugation of the unsaturated fatty acids. Archives of Biochemistry, 19: 474-482.

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A study of the effects of KOH concentration and time of reaction on conjugation of polyunsaturated fatty acids.

Holman, R. T. and L. Hagdahl

1950. Displacement analysis of lipides. III. Separation of normal saturated fatty acids from formic to behenic. Journal of Biological Chemistry, 182: 421-427.

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The fatty acids were adsorbed on a Darco G-60 column and separated by displacement analysis using water, alcohol-water mixtures, and chloroform-alcohol as solvents. Yields were not studied.

Holman, R. T.

1951. Displacement analysis of lipids. IV. Carrier displacement separation of saturated fatty acids. Journal of the American Chemical Society, 73: 1261-1263.

★ ●

The saturated fatty acids are separated by carrier displacement on charcoal using their methyl esters as carriers. 5-15 mg. quantities of lauric, myristic, palmitic, and stearic acids are nearly 100% recoverable.

Holman, R. T.

★ 1951. Displacement analysis of lipids. V. Separation of substances analogous to fatty acids. Journal of the American Chemical Society, 73: 3337-3340.

Various aliphatic compounds were studied for use as carriers for displacement separations. A series of C_{18} compounds of varying adsorbability which are suitable for use as displacers is given.

Holman, R. T. and W. T. Williams

1951. Displacement analysis of lipids. VI. Separation of unsaturated acids. Journal of the American Chemical Society, 73: 5285-5289.

★ ●

Unsaturated fatty acids were separated by displacement chromatography on charcoal. Saturated and unsaturated acids of 4-18 C with the same chain length were separated. Effects of conjugation on adsorption characteristics are discussed.

Holman, R. T.

1951. Displacement analysis of lipids. VII. Carrier separation of unsaturated fatty acids. Journal of the American Chemical Society, 73: 5289-5292.

★ ●

A method for separation of unsaturated fatty acids on charcoal using methyl esters as carriers is described. Effects of unsaturation on adsorbability are discussed.

Holman, R. T., H. Hayes, and P. R. Edmondson

1957. Analysis of unsaturated fatty acids. Essential Fatty Acids. Proceedings of the International Conference on Biochemical Problems of Lipids, 4th Oxford, (Pub. 1958) pp. 9-15. Chemical Abstracts, 53:17277f (1959).

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Methods are given for analysis of unsaturated fatty acids by alkali isomerization (Holman, in Methods of Biochemical Analysis, (D. Glick, Ed.) 4: 99, 1957) paper chromatography (Mangold, et al, Journal of the American Chemical Society, 77: 6070, 1956), and near-infra-red spectrophotometry (Holman and Edmondson, Analytical Chemistry, 28: 1533, 1956).

Horecker, B. L., T. S. Ma, and E. Haas

1940. Note on the determination of micro quantities of organic phosphorus.

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Journal of Biological Chemistry, 136: 775-776.

A modified Fiske-Subbarow method (Journal of Biological Chemistry, 66: 375, 1925) which will determine 1 μg. of phosphorus with an accuracy of ± 3% is described. A sulfuric acid concentration of 2 N is used in place of the original 0.5 N, and the color is developed with heat.

Hornstein, I., J. A. Alford, L. E. Elliot, and P. F. Crowe

1960. Determination of free fatty acids in fat. Analytical Chemistry, 32: 540-542.

★ ●

Free fatty acids are separated from fat mixtures by adsorption on Amberlite IRA-400 resin. Fat is removed by washing the resin with petroleum ether. The fatty acids are converted to their methyl esters by treatment, while on the resin, with methanolic HCl and are separated and identified by gas chromatography on a polyvinyl acetate column.

Horowitz, N. H. and G. W. Beadle

1943. A microbiological method for the determination of choline by use of a mutant of neurospora. Journal of Biological Chemistry, 150: 325-333.

A simple, sensitive (0.02 mg. choline/liter), and specific method. Interfering substances are removed from the solution by adsorption on Permutit, and choline is eluted with 5% NaCl. Choline is determined by microbiological assay using a cholineless strain of Neurospora crassa.

Howard, G. A. and A. J. P. Martin

1950. The separation of the C_{12}-C_{18} fatty acids by reversed phase partition chromatography. Biochemical Journal, 46: 532-538.

★ ●

A method is given for the separation of the fatty acids on a dichlorodimethylsilane-impregnated silicic acid column.

Huennekens, F. M., D. J. Hanahan, and M. Uziel

1954. Paper chromatography of lecithins. Journal of Biological Chemistry, 206: 443-447.

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A series of compounds derived from (dipalmitoleyl)-L- α-glycerylphosphorylcholine are separated by paper chromatography, using alcohol-water mixtures as solvent systems.

Hunter, M. O., R. A. Knouff, and J. B. Brown

1945. The microdetermination of tissue lipids. Ohio Journal of Science, 45: 47-54. Chemical Abstracts, 39: 3804[2] (1945).

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Methods are described for the extraction and saponification of tissue lipids and titrimetric estimation of 1-10 mg. of fatty acids to within 2%. Other methods are also discussed.

It was found that liver and adrenal tissue lipids were completely extracted by a 5 hour extraction with 3:1 alcohol-ether at room temperature. The precipitation of cholesterol as the digitonide by the Schoenheimer-Sperry method was found to be incomplete for samples of 0.1 to 0.5 mg.

Ikeda, R. M., A. D. Webb, and R. E. Kepner

1954. Chromatographic separation of p-phenylazophenacyl esters on silicic acid. Analytical Chemistry, 26: 1228-1229.

★ ●

A method is described for the separation of fatty acids by chromatography as their p-phenylazophenacyl esters on silicic acid using benzene and Skellysolve B mixtures as developing solvents.

Inouye, Y. and J. Yukawa

1940. Separation and identification of aliphatic acids. I. Hydroxamic acids derived from saturated aliphatic acids. Journal of the Agricultural Chemical Society of Japan, 16: 504-509. Chemical Abstracts, 35:730[9](1941).

★ ●

Hydroxamic acid derivatives were prepared from ethyl esters of fatty acids by reaction with hydroxylamine hydrochloride and sodium ethylate at room temperature. Data on the melting points of the derivatives are given.

Inouye, Y. and M. Noda

1950. Separation and identification of fatty acids. IX. Paper partition chro-

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matography of hydroxamic acids. Journal of the Agricultural Chemical Society of Japan 18: 294-298. (English summary) Chemical Abstracts, 45: 8449f (1951).

Hydroxamic acid derivatives of C_2-C_{22} saturated aliphatic acids were chromatographed on paper using BuOH, EtOAc, and butrone as solvents, and located with 10% $FeCl_3$ in EtOH. R_f values and colors developed with $FeCl_3$ for the spots of 13 of the hydroxamic acids are given.

Inouye, Y. and M. Noda

1951. Separation and identification of fatty acids. X. Simplified methods of the preparation of hydroxamic acid solutions for paper partition chromatography. Journal of the Agricultural Chemical Society of Japan, 24: 291-295. Chemical Abstracts, 46:6408a (1952).

Four simplified methods for preparing solutions of hydroxamic acids for chromatographic separation are given.

Inouye, Y. and M. Noda

1951. Separation and identification of fatty acids. XI. Paper partition chromatography of aliphatic carboxylic acids by means of hydroxamic acid method. Journal of the Agricultural Chemical Society of Japan, 24: 295-298. Chemical Abstracts, 46:6408c (1952).

A modification of the Inouye and Noda method for saturated fatty acids (Journal of the Agricultural Chemical Society of Japan, 18: 294, 1950) for use with unsaturated, hydroxy, and polybasic acids. R_f values using BuOH solvent, and color with $FeCl_3$ of the hydroxamic acids derived from various acids are given.

Inouye, Y. and M. Noda

1951. Separation and identification of fatty acids. XII. Application of paper chromatography to the analysis of fats. Journal of the Agricultural Chemical Society of Japan, 25: 161-165. Chemical Abstracts, 46:6408c (1952).

The acidic oxidation products of $KMnO_4$ oxidation of a glyceride mixture are esterified, and the esters are converted to their hydroxamic acid derivatives which are separated by paper chromatography.

Inouye, Y., M. Noda, and Y. Hamuro

1952. Separation and identification of fatty acids. XIII. Investigation of the constitution of unsaturated fatty acids by paper chromatography. Journal of the Agricultural Chemical Society of Japan, 25: 491-495. Chemical Abstracts, 46:6408e (1952).

Hydroxamic acid derivatives of unsaturated fatty acids are prepared by oxidizing ozonolysis products of the acids with alkaline AgO, esterifying the acids formed, and converting the esters to their hydroxamic acid derivatives.

Inouye, Y. and M. Noda

1952. Separation and identification of fatty acids. XIV. Paper chromatography of fatty acids using the filter paper impregnated with silicic acid. Journal of the Agricultural Chemical Society of Japan, 25: 496-499. Chemical Abstracts, 46:6408f (1952).

Higher fatty acids were separated on filter paper treated with silicic acid using a butanol-benzene solvent mixture.

Inouye, Y. and M. Noda

1952. Separation and identification of fatty acids. XV. Paper chromatography of saturated higher fatty acids. Journal of the Agricultural Chemical Society of Japan, 26: 634-638. (Pub. 1953) Chemical Abstracts, 47:9635e (1953).

Higher fatty acids were separated from mixtures of fatty acids by chromatography on petroleum hydrocarbon impregnated paper with a polar mobile phase. R_f values of the acids are given.

Inouye, Y. and M. Noda

1953. Separation and identification of fatty acids. XVI. Paper chromatography of unsaturated higher fatty acids. Journal of the Agricultural Chemical Society of Japan, 27: 50-53. Chemical Abstracts, 47:9635g (1953).

The method of Inouye and Noda (Journal of the Agricultural Chemical Society of Japan,

26: 634, 1952) for saturated fatty acids was applied to unsaturated fatty acids. R_f values for 14 unsaturated fatty acids are given.

Inouye, Y. and M. Noda
1955. Separation and identification of fatty acids. XVII. Paper chromatography of saturated fatty acids as their 2, 4-dinitrophenylhydrazides. Bulletin of the Agricultural Chemical Society of Japan, 19: 214-219 (In English). Chemical Abstracts, 50:17478b (1956).

2, 4-dinitrophenylhydrazides were obtained by reaction of fatty acid chlorides with 10% solution of 2,4-dinitrophenylhydrazine in pyridine. The hydrazides were chromatographed on tetralin-impregnated paper using tetralin-AcOH-EtOH or MeOH mixtures as mobile phase. The spots were located by spraying with 0.5 N KOH in EtOH. Data on melting points of the hydrazine derivatives and their R_f values with the various solvents are given.

Inouye, Y., M. Noda, and O. Hirayama
1955. Paper chromatography of unsaturated fatty acid esters as their mercuric acetate addition compounds. Journal of the American Oil Chemists Society, 32: 132-135. Chemical Abstracts, 49:6783f (1955).

Methyl esters of unsaturated fatty acids were chromatographed as their mercuric acetate addition compounds and were detected by treatment with diphenylcarbazone. R_f values of 19 of the addition compounds are given.

Inouye, Y., O. Hirayama, and M. Noda
1956. Separation and identification of fatty acids. XVIII. Paper chromatography of fatty acids as their acetol ester derivatives. Journal of Japan Oil Chemists' Society, 5: 16-18. Chemical Abstracts, 50:17478e (1956).

Acetol esters were obtained by reaction of K soaps of the fatty acids with $BrCH_2COCH_3$ and converted to 2, 4-dinitrophenylhydrazones and thiosemicarbazones. The derivatives were chromatographed on Decalin-impregnated paper with mixtures of MeOH, Decalin, AcOH, and EtOH as solvents. Melting points of acetol esters and R_f values of the derivatives are given.

Inouye, Y., O. Hirayama, and M. Noda
1956. Separation and identification of fatty acids. XX. 2, 4-dinitrophenylhydrazones of p-bromophenacyl esters as derivatives for characterization of unsaturated fatty acids. Bulletin of the Agricultural Chemical Society of Japan, 20: 194-199. Chemical Abstracts, 51: 7297f (1957).

A method is described for the preparation of the p-bromophenacyl ester-2, 4-dinitrophenylhydrazone derivatives of the C_2-C_{20} fatty acids with even-numbered carbon chains for use as identifying derivatives for the unsaturated fatty acids.

Inouye, Y., O. Hirayama, and M. Noda
1956. Separation and identification of fatty acids. XXI. Paper chromatography of fatty acids as their p-bromophenacyl ester derivatives. Bulletin of the Agricultural Chemical Society of Japan, 20: 200-205. Chemical Abstracts, 51:7297g (1957).

A method is described for the separation of fatty acids by paper chromatography as their p-bromophenacyl ester-2, 4-dinitrophenylhydrazone derivatives and their mercuric acetate addition compounds.

Inouye, Y., O. Hirayama, and M. Noda
1957. Separation and identification of fatty acids. XXII. Quantitative analysis of fatty acids by paper chromatography. Journal of the Agricultural Chemical Society of Japan, 31: 568-572. Chemical Abstracts, 53:20841f (1959).

The p-bromophenacyl ester-2, 4-dinitrophenylhydrazones and $Hg(OAc)_2$ derivatives of the fatty acids were separated by paper chromatography, extracted with benzene, and measured spectrophotometrically.

Ireland, J.T.
1941. The colorimetric estimation of total cholesterol in whole blood, serum, plasma, and other biological material. Biochemical Journal, 35: 283-293.

The effects of light, reagent concentra-

tions, and time and temperature of color development in the determination of cholesterol by the Liebermann-Burchard reaction were studied and are discussed.

A method is described for development of color using acetic anhydride-sulfuric acid (20:1), and developing at 18° for 60 minutes in the dark.

Iwayama, Y.

1959. New colorimetric determination of higher fatty acids. Yakugaku Zasshi, 79: 552-554. (English summary).

A method is described for determination of saturated and unsaturated fatty acids from C_{10} to C_{22} by colorimetric measurement of the blue color produced by a chloroform solution of the copper salt of the fatty acid in the presence of triethanolamine. The color follows the Lambert-Beer Law within the concentration range used, and the method has good reproducibility.

Jackson, F. L. and J. E. Callen

1951. Evaluation of the Twitchell isoöleic method: comparison with the infrared trans-isoöleic method. Journal of the American Oil Chemists Society, 28: 61-65. Chemical Abstracts, 45:3619d (1951).

The infrared method was found to be better in general than the Twitchell method.

Jacobi, H. P., C. A. Baumann, and W. J. Meek

1941. The choline content of rats on various choline-free diets. Journal of Biological Chemistry, 138: 571-582.

Choline was determined as the reineckate. The ammonium reineckate was added in a methanol solution rather than water (Beattie, Biochemical Journal, 30: 1554, 1936), as it is more soluble in methanol, and precipitation was found to be just as complete.

Results by the use of the Reineckate method were found to be in agreement with those by the biological assay method of Best, et al (Biochemical Journal, 29: 2278, 1935).

Jaky, M.

1959. Paper chromatography of fats. Fette, Seifen, Anstrichmittel, 61: 6-10. Chemical Abstracts, 53:12709h (1959).

Mono-, di-, and triglycerides were separated by paper chromatography using 80% dimethyl ketone in water as solvent. The separated spots were identified by spraying with Rhodamine B.

James, A. T. and J. P. W. Webb

1957. The behavior of polyunsaturated fatty acids on the gas-liquid chromatogram. Essential fatty acids. Proceedings of the International Conference on Biochemical Problems of Lipids, 4th Oxford, (Pub. 1958) pp. 3-8. Chemical Abstracts, 53:17277d (1959).

Unsaturated fatty acids were separated by chromatography on Apiezon M, and the individual acids were oxidized with permanganate in glacial acetic acid. The resulting acids were extracted, converted to their methyl esters with diazomethane, and separated and identified by rechromatography.

James, A. T.

1958. The separation of the long chain fatty acids by gas-liquid chromatography. American Journal of Clinical Nutrition, 6: 595-600.

A discussion of factors (positional isomers, polarity of phases, etc.) which influence the separation of long-chain fatty acids by gas chromatography. A description of an ionization detector and column heater are given.

Johnson, R. M. and P. H. Dutch

1951. Use of trichloracetic acid in purification of lipids. Proceedings of the Society for Experimental Biology and Medicine, 78: 662-664.

Acid-soluble material was removed from tissue with 10% TCA containing 0.4 M $MgCl_2$, and lipid was then extracted with EtOH-petroleum ether. No measurable hydrolysis of lipids occurred.

Jones, B. J. and F. B. Moreland

1955. p-Toluenesulfonic acid, cholesterol determination, and explosions. Clinical Chemistry, 1: 345.

Explosions which were apparently due to p-toluenesulfonic acid occurred during determination of cholesterol by the method of Pearson, Stern, and McGavack (Analytical

Chemistry, 25: 813, 1953).

Jones, K. K.

1950. A micromethod for fat analysis based on formation of monolayer films. Quarterly Bulletin of Northwestern University Medical School, 24: 253-256. Chemical Abstracts, 45:5754a (1951).

A method is described for estimation of lipids by spreading them in monomolecular films on an acidified water solution and measuring the area of the films.

Jorgensen, K. and H. Dam

1957. An ultramicro method for the determination of total cholesterol in bile based on the Tschugaeff color reaction. Acta chemica scandinavia, 11: 1201-1208.

A method is described for measurement of as little as 2 μg. of cholesterol in a final volume of reaction mixture of 0.5 ml. Hydrolysis, extraction and color development are all carried out in the same tube. The developed color is read spectrophotometrically.

Kabara, J. J.

1954. The light insensitivity of the Liebermann-Burchard reaction during spectrophotometric determination of cholesterol. Journal of Laboratory and Clinical Medicine, 44: 246-249.

It was found that exclusion of light during development of color with the Liebermann-Burchard reaction was unnecessary unless light of 340-540 mμ. was used for measurement. Light had no effect on the L-B reaction from 580-740 mμ. in either polar or nonpolar solvents.

Kapitel, W.

1956. Quantitative column chromatographic separation of mixtures of fatty acids. Fette, Seifen, Anstrichmittel, 58: 91-94. Chemical Abstracts, 50:9761c (1956).

The C_6-C_{23} saturated fatty acids were separated by chromatography on a column of kieselguhr which was impregnated with paraffin and treated with dichlorodimethyl silane. A mixture of various amounts of water and acetone was saturated with paraffin oil and used as eluent.

Kaufmann, H. P. and W. Wolf

1943. Adsorption separation in the field of fats. V. The separation of cis-trans isomers. Fette und Seifen, 50: 519-521. Chemical Abstracts, 39: 205^6 (1945).

A comparative study and discussion of the use of Al_2O_3, SiO_2, and charcoal as adsorbents for the separation of isomeric fatty acids.

Kaufmann, H. P. and J. Budwig

1950. The foam test in paper chromatography. Fette und Seifen, 52: 555-556. Chemical Abstracts, 45:2236d (1951).

As little as 10 μg. of oleic acid is detectable by using an adaptation of the foam test for chromatography. The fatty acid or soap is placed on a copper acetate-impregnated paper and a H_2O_2-NH_4OH reagent is added, causing it to foam.

Kaufmann, H. P.

1950. New methods of fat analysis. Fette und Seifen, 52: 713-721. Chemical Abstracts, 45:8271a (1951).

Various methods suitable for identification of traces of fats and fatty acids on paper chromatograms are described.

Kaufmann, H. P. and J. Budwig

1951. Paper chromatography in the fat field. IV. Radiometry of oleic acid. Fette und Seifen, 53: 69-73. Chemical Abstracts, 45:9893e (1951).

Fatty acid is determined by formation of Co^{60} soaps and radiation counting.

Kaufmann, H. P. and J. Budwig

1951. Paper chromatography in the fat field. V. Radiometric determination of the iodine number. Fette und Seifen, 53: 253-259. Chemical Abstracts, 45:7801i (1951).

An adaptation of the Hanus method for iodine number determination for use in paper chromatography. The iodine number is determined by measurement of the radiation from the I^{139} added by the fat.

Kaufmann, H. P. and J. Budwig

1951. Paper chromatography in the field of fats. VII. Identification and separation of fatty acids. Fette und Seifen, 53: 390-399. Chemical Abstracts, 46:6851b (1952).

Dyes and metallic soaps suitable for use in locating and identifying fatty acids are discussed.

Kaufmann, H. P. and J. Budwig

1952. Biology of fats. V. Paper chromatography of blood lipides, the cancer problem, and fat research. Fette und Seifen, 54: 156-165. Chemical Abstracts 46:8703b (1952).

A method is described for paper chromatographic separation of the lipids in a small sample of blood drawn from the finger or ear.

Kaufmann, H. P. and J. Budwig

1952. Paper chromatography in the field of fats. X. Flourescent dyes as indicators in the paper chromatographic analysis of fat acids and fats. Fette und Seifen, 54: 7-10. Chemical Abstracts, 46:7793d (1952).

Various flourescent indicators suitable for identification of fatty acids on paper chromatograms are given.

Kaufmann, H. P., J. Budwig, and C. W. Schmidt

1952. Paper chromatography in the field of fats. XI. Identification and separation of conjugated unsaturated fat acids. Fette und Seifen, 54: 10-12. Chemical Abstracts, 46:7793g (1952).

Colors obtained with various metallic soaps were used for the identification of conjugated fatty acids after chromatography on paper.

Kaufmann, H. P. and W. H. Nitsch

1954. Paper chromatography in the fat field. XVI. Further experiments on the separation of fatty acids. Fette und Seifen Anstrichmittel, 56: 154-158. Chemical Abstracts, 49:14345d (1955).

Directions are given for separation of fatty acids from capric to stearic by paper chromatography. High-boiling hydrocarbons and acetic acid were used as solvent phases.

Kaufmann, H. P. and H. G. Kohlmeyer

1955. Paper chromatography of waxes. I. Separation of wax alcohols. Fette, Seifen, Anstrichmittel, 57: 231-235. Chemical Abstracts, 49:12859c (1955).

C_{10}-C_{18} even-numbered saturated normal alcohols can be separated from lipid mixtures by paper chromatography using 85% acetic acid as mobile phase.

Kaufmann, H. P. and W. H. Nitsch

1955. Paper chromatography in the fat field. XVII. Separation of unsaturated fatty acids. Fette, Seifen, Anstrichmittel, 57: 473-474. Chemical Abstracts, 50:2190h (1956).

The method of Kaufmann and Nitsch (Fette und Seifen Anstrichmittel, 56: 154, 1954) was used for the separation of unsaturated fatty acids.

Kaufmann, H. P. and W. H. Nitsch

1956. Paper chromatography in the fat field. XVIII. Separation of hydroxylated and brominated fatty acids. Fette, Seifen, Anstrichmittel, 58: 234-238. Chemical Abstracts, 50:13475b (1956).

An application of the method of Kaufmann and Nitsch (Fette, Seifen, Anstrichmittel, 57: 473, 1955) to the separation of hydroxylated and brominated acids.

Kaufmann, H. P.

1956. Paper chromatography in the fat field. XIX. Quantitative paper chromatographic determination of the straight-chain fatty acids and their mixtures. Fette, Seifen, Anstrichmittel, 58: 492-498. Chemical Abstracts, 52:4211h (1958).

Various methods suitable for location of fatty acids are discussed.

Kaufmann, H. P. and E. Mohr

1958. Paper chromatography of fatty acids. XXIV. Fette, Seifen, Anstrichmittel, 60:165-177. Chemical Abstracts, 52:14196h (1958).

A method is described for the separation of fatty acid mixtures on undecane-impregnated paper.

Kaufmann, H. P. and M. M. Deshpande
1958. Paper chromatography of fats. XXVI. The quantitative paper chromatographic-polarographic analysis of fatty acids. Fette, Seifen, Anstrichmittel, 60: 537-541. Chemical Abstracts, 53:1783g (1959).

The paper chromatographic and polarographic methods for separation and determination of fatty acids are compared and discussed.

Kaufmann, H. P. and M. Arens
1958. Paper chromatography of fatty acids. XXVIII. Separation of thiocyanogen derivatives. Fette, Seifen, Anstrichmittel, 60: 803-806. Chemical Abstracts, 53:3737g (1959).

"Critical pairs" of fatty acids may be separated by paper chromatography as their thiocyanogen derivatives using 70% acetic acid as solvent.

Kaufmann, H. P. and H. Schnurbusch
1958. The paper chromatography of fats. XXIX. Analysis of fatty acids by means of copper and mercury method. Fette, Seifen, Anstrichmittel, 60: 1046-1050. Chemical Abstracts, 53:8663h (1959).

A method for identification of fatty acids in a mixture by paper chromatography and conversion to their Cu and Hg soaps is described.

Kaufmann, H. P. and H. Schnurbusch
1959. Paper chromatography in the fat field. XXX. Paper chromatographic analysis of the glycerides. Fette, Seifen, Anstrichmittel, 61: 523-528. Chemical Abstracts, 54:5129a (1960).

Glycerides are separated on paper impregnated with silicone oil using acetone-acetonitrile as solvent. The spots on the developed chromatogram are located by coloring with copper acetate and potassium ferrocyanide after saponification.

Kaufmann, H. P. and Z. Makus
1959. Paper chromatography in the fat field. XXXI. Separation of mixtures of synthetic and natural triglycerides by paper chromatography. Fette, Seifen, Anstrichmittel, 61: 631-636. Chemical Abstracts, 54:5129b (1960).

Mixtures of triglycerides were separated on paper treated with undecane by using acetic acid as solvent.

Kaufmann, H. P. and H. Kirschnek
1959. Paper chromatography in the fat field. XXXIV. Fatty aldehydes. 5. Qualitative and quantitative analysis of fatty aldehydes with the help of paper chromatography. Fette, Seifen, Anstrichmittel, 61: 750-759. Chemical Abstracts, 54: 5129c (1960).

Fatty aldehydes were separated as their 2, 4-dinitrophenylhydrazones by using undecane and nitromethane as solvents for reversed-phase paper chromatography.

Kaye, I. A.
1940. Determination of total and free cholesterol in blood serum. Journal of Laboratory and Clinical Medicine, 25: 996-1001.

A method is described for determination of cholesterol in blood serum by precipitation with digitonin directly on the lipid extract and colorimetric measurement by the Liebermann-Burchard method. Saponification is unnecessary as the concentration of ester cholesterol in serum is constant. Acetic anhydride is used as solvent for the cholesterol standard as it gives a more stable solution than chloroform.

Sobel (Journal of Biological Chemistry, 157: 255, 1945) found the method in agreement with the Schoenheimer-Sperry method.

Kean, E. L. and F. C. Charalampous
1959. New methods for the quantitative estimation of myo-inositol. Biochimica et Biophysica Acta, 36: 1-3.

Methods are described for estimation of inositol by enzymic conversion of the inositol to glucuronic acid and colorimetric measurement by the orcinol reaction, and by spectrophotometric measurement of the

change in absorption caused by enzymic oxidation of triphosphopyridine nucleotide in the presence of glucuronate.

Kennedy, E. P. and H. A. Barker
1951. Paper chromatography of volatile acids. Analytical Chemistry, 23: 1033-1034.

A method is described for chromatography on paper of ammonium salts of the volatile fatty acids using solvents containing free ammonia for development. Bromophenol blue is used to locate the spots after development.

Kenny, A. P.
1952. The determination of cholesterol by the Liebermann-Burchard reaction. Biochemical Journal, 52: 611-619.

A modification of the L-B method for estimation of total cholesterol in serum or plasma. The yellow component is measured at 430 mμ. The method is suitable for routine clinical work. The factors influencing color development in the determination of cholesterol by the L-B reaction were studied and are discussed.

Kepner, R. E., A. D. Webb, R. L. King, and A. D. Bond
1957. p-Phenylazophenacyl esters. Rates of movement relative to p-phenylazophenacyl bromide on silicic acid and identification by paper partition chromatography. Analytical Chemistry, 29: 1162-1164.

An improved method for the preparation of p-phenylazophenacyl bromides and a method for paper chromatography of p-phenylazophenacyl esters are described. Rates of travel of the p-phenylazophenacyl derivatives on silicic acid as compared to phenylazophenacyl bromide are given. Melting points of 30 derivatives are listed.

Kerr, L. M. H. and W. S. Bauld
1953. The chromatographic separation of free and combined plasma cholesterol. Biochemical Journal, 55: 872-875.

Acetone-ethanol extracts of plasma were chromatographed on alumina using petroleum ether and benzene as eluting solvents for the separation of cholesterol and cholesterol esters. Both the cholesterol and the esters were estimated by the Liebermann-Burchard color reaction, using separate standard curves for the cholesterol and the esters. Accuracy of the method is lower than the Schoenheimer-Sperry method.

Ketchum, D.
1946. Semimicrodetermination of saponification equivalent by Rieman's double-indicator method. Industrial and Engineering Chemistry, Analytical Edition, 18: 273-274.

A semimicro modification of Rieman's method (Industrial and Engineering Chemistry, Analytical Edition, 15: 325, 1943).

Kibrick, A. C., T. Roberts, and S. Skupp
1951. Determination of cholesterol in blood plasma or serum by hydrolysis with benzyltrimethylammonium hydroxide. Archives of Biochemistry and Biophysics, 32: 9-13.

The cholesterol esters are hydrolyzed with benzyltrimethylammonium hydroxide during evaporation of the alcohol-ether tissue extract. The color is developed in the residue by the Liebermann-Burchard reaction and read colorimetrically. Recovery of 89-104% of added cholesterol was obtained. The method compares favorably with the Schoenheimer-Sperry method.

Kibrick, A. C., L. B. Safier, and S. J. Skupp
1959. Existence of fatty acid peroxides in normal blood and tissues of man and animals. Proceedings of the Society for Experimental Biology and Medicine, 101: 137-139.

The thiobarbituric reaction (Kohn and Liversedge, Journal of Pharmacology and Experimental Therapeutics, 82: 292, 1944) was modified for determination of fatty acid peroxides in blood as well as in tissues.

Kibrick, A. C. and S. J. Skupp
1959. Chromatographic separation of fatty acids based on chlorophenacyl esters. Analytical Chemistry, 31: 2057-2060.

A method is described for synthesis of 4'-bromo-2-chloroacetophenone and prep-

aration of its fatty acid esters. The fatty acid esters are separated on a polyethylene column using various water-alcohol mixtures as eluents. Melting points, solubilities, and absorptivities of the derivatives are given.

A drawing of a reservoir system for maintaining constant pressure while changing solvents is included.

King, E. J.

1932. The colorimetric determination of phosphorus. Biochemical Journal, 26: 292-297.

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Perchloric acid is used for oxidation of organic matter. 1:2:4-aminonaphtholsulfonic acid is used as a reducing agent to develop molybdate color.

Kingsley, G. R. and R. R. Schaffert

1949. Determination of free and total cholesterol by direct chloroform extraction. Journal of Biological Chemistry, 180: 315-328.

▲

Cholesterol is extracted from 0.2 ml. of serum with chloroform. The extract is dried with anhydrous magnesium sulfate. Acetic anhydride and sulfuric acid are added to a portion of the dried extract and the color is read photometrically.

Kinley, L.

1958. Serum cholesterol determinations as affected by vitamin A. Proceedings of the Society for Experimental Biology and Medicine, 99: 244-245.

▲

Very high vitamin A levels will increase the cholesterol levels as measured by the Zak method due to interference with the ferric chloride reagent. Moderate levels have little effect. Cholesterol values determined by the Schoenheimer-Sperry method were not affected by vitamin A.

Kirchner, J. G., A. N. Prater, and A. J. Haagen-Smit

1946. Separation of acids by chromatographic adsorption of their p-phenylphenacyl esters. Industrial and Engineering Chemistry, Analytical Edition, 18: 31-32.

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A method is described for the separation of fatty acids by chromatography of their p-phenylphenacyl esters on silicic acid. Good separations were obtained.

Kirk, E., I. H. Page, and D. D. Van Slyke

1934. Gasometric microdetermination of lipids in plasma, blood cells, and tissue. Journal of Biological Chemistry, 106: 203-234.

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After alcohol-ether (3:1) extraction, aliquots of the extract are used for determination of lipid constituents. Total lipids are estimated by determination of total non-volatile carbon. Cholesterol is determined by combustion of the digitonide. Phospholipids are estimated as phosphoric acid by precipitation as strychnine phosphomolybdate and determination of carbon in the precipitate.

The maximum temperature of 60° was found to be critical in the evaporation of the alcohol-ether extract, as reextraction with petroleum ether was incomplete if a higher evaporation temperature was used.

Acetone-$MgCl_2$ precipitates all but 2 to 3% of the phospholipids, but all the precipitated phospholipid is not soluble in moist ether.

Kirk, E.

1938. A study on Kimmelsteil's procedure for titrimetric cerebroside determination, with description of an improved technique. Journal of Biological Chemistry, 123: 613-621.

Kimmelsteil's procedure (Biochemische Zeitschrift, 212: 259, 1929) was found to give high values for cerebroside due to the presence of interfering reducing substances. A modification is presented which removes these substances by precipitation from the hydrolyzed sample with zinc hydroxide. The method is suitable for determination of 0.3 to 1.3 mg. of pure cerebroside (±4%) and gives quantitative recovery of added cerebroside.

Kirk, E.

1938. A micro method for approximate estimation of lecithin, cephalin, ether-insoluble phosphoinositide, and cerebrosides in plasma, red blood cells, and

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tissues. Journal of Biological Chemistry, 123, 623-636.

After alcohol-ether extraction of the lipids, the ether-insoluble phosphatide is determined by phosphorus analysis of the residue which is insoluble in moist ether. Lecithin is estimated by choline analysis of the moist ether extract after $Ba(OH)_2$ hydrolysis by the method of Roman (Biochemische Zeitschrift, 219: 218, 1930). Cephalin is calculated as the difference in lecithin and total ether-soluble phosphatide. Cerebrosides are estimated by Kirk's modification (Journal of Biological Chemistry, 123: 613, 1938) of Kimmelsteil's method. The lecithin precipitated with acetone-$MgCl_2$ was found to be wholly soluble in moist ether.

Klee, Leo and G. H. Benham

1950. The determination of the true iodine numbers of oils containing conjugated double bond systems. Journal of the American Oil Chemists Society, 27: 130-133. Chemical Abstracts, 44: 5119e (1950).

The modified Rosenmund-Kuhnhenn procedure is made suitable for determination of iodine numbers of oils with conjugated double bond systems by extending the reaction time to 30-120 minutes.

Klein, P. D. and E. T. Janssen

1959. The fractionation of cholesterol esters by silicic acid chromatography. Journal of Biological Chemistry, 234: 1417-1420.

A method is described for the separation of cholesterol esters into groups consisting of saturated, oleate, linoleate, and arachidonate esters by chromatography on a silicic acid column.

Klenk, E. and H. Langerbeins

1941. Distribution of neuraminic acid in the brain. (With a micromethod for the estimation of this substance in nerve tissue.) Zeitschrift für physiologische Chemie, 270: 185-193. Chemical Abstracts, 37:899[4] (1943).

The neuraminic acid in about 30 mg. of brain tissue can be detected by the orcinol reaction. Tissue extract is heated with orcinol reagent, centrifuged, and the red to red-violet color obtained is read with a colorimeter. Galactose gives a green color with the reagent, but will interfere only if in large excess.

Knight, H. B., L. P. Witnauer, J. E. Coleman, W. E. Noble, Jr., and D. Swern

1952. Dissociation temperatures of urea complexes of long chain fatty acids, esters, and alcohols. Analytical Chemistry, 24: 1331-1334.

Methods for the preparation of urea adducts and determination of their dissociation temperatures are given. The dissociation temperatures of 42 compounds are given, and the use of the adducts as identifying derivatives is discussed.

Kobrle, V. and R. Zahrodnik

1954. Paper partition chromatography of higher fatty acids. Chemicke Listy, 48: 1189-1196. Chemical Abstracts, 48: 13546h (1954).

Even-numbered C_{10}-C_{22} and C_{26} fatty acids were separated by chromatography on paper impregnated with a 12% toluene solution of vegetable oils. R_f values for various solvent systems and papers are given.

Koehler, A. E. and E. Hill

1949. The molecular microdistillation of cholesterol and cholesterol esters. Journal of Biological Chemistry, 179: 1-10.

A small molecular still for use in analysis of small amounts (up to 5 mg.) of lipids is described. It was used for the quantitative separation of cholesterol and cholesterol esters by distillation.

Koenig, R. A. and C. R. Johnson

1942. Colorimetric determination of phosphorus in biological materials. Industrial and Engineering Chemistry, Analytical Edition, 14: 155-156.

Phosphorus is precipitated as phosphovanadiomolybdate and measured spectrophotometrically. Effects of variables are discussed.

Kolb, D. K. and J. B. Brown

1955. Low temperature solubilities of fatty acids in selected organic solvents. ★ ● Journal of the American Oil Chemists Society, 32: 357-361. Chemical Abstracts; 49:10639d (1955).

Data on the low-temperature solubilities (10° to -70°) of several purified fatty acids in various organic solvents are given. The application of the data to separation of fatty acids by low-temperature crystallization is discussed.

Korpaczy, I.

1959. Semimicro colorimetric determination of the phosphorus content of lipoids. ▲ Fette, Seifen, Anstrichmittel, 61: 748-750. Chemical Abstracts, 54: 4730i (1960).

A modification of the method of Thaler (Fette und Seifen 54: 763, 1952). The sample is ashed with MgO and the color produced with molybdate is measured colorimetrically.

Krainick, H. G. and F. Muller

1941. Photometric microdetermination of fat acids. Mikrochemie (vereinigt mit ● Mikrochemica Acta) 30: 7-14. Chemical Abstracts, 37:4416[8] (1943).

Blood or plasma is saponified and the fatty acids are freed and extracted. An aliquot of the extract is heated with rosanaline reagent, the color developed is measured, and the fatty acid concentration is estimated from a standard curve. Suitable for determination of 0.05 - 0.5 mg. of fatty acid in biological materials such as blood.

Krewson, C. F.

1951. Refractive indices for the methyl esters of the C_{12}-C_{28} saturated n-aliphatic acids. ● Journal of the American Chemical Society, 73:1365.

Methyl esters of the saturated fatty acids were prepared and refractive indices of the esters were determined. The refractive indices for the even-numbered C_{12}-C_{28} methyl esters are given.

Kruty, M., J. B. Segur, and C. S. Miner, Jr.

1954. The determination of monoglycerides and glycerol in mixtures. ● Journal of the American Oil Chemists Society, 31: 466-469. Chemical Abstracts, 49: 1346i (1955).

A modification of the periodic acid method of Pohle and Mehlenbacher (Journal of the American Oil Chemists Society, 27: 54, 1950).

Kuemmel, D. F.

1958. Direct determination of saturated fatty acids in fats, oils, and methyl esters. ● Journal of the American Oil Chemists Society, 35: 41-45. Chemical Abstracts, 52:3365g (1958).

Saturated fatty acids with a chain length of C_{16} or greater are determined by methanolysis of the triglycerides, oxidation of the unsaturated methyl esters with $KMnO_4$, removal of the oxidation products by alkaline washing, and direct weighing of the isolated methyl esters of the saturated fatty acids.

Kummerow, F. A. and B. F. Daubert

1950. Limitations of the periodate oxidation method for the determination of monoglycerides in fats and oils. ● Journal of the American Oil Chemists Society, 27: 100-102. Chemical Abstracts, 44:4268c (1950).

It was shown that the method of Handschumaker and Linteris (Journal of the American Oil Chemists Society, 24: 143, 1947) is not specific for monoglycerides in natural fats and oils.

Kushner, D. J.

1956. A spectrophotometric microdetermination of choline. ▲ Biochimica et Biophysica Acta, 20: 554-555.

A modification of the method of Appleton (Journal of Biological Chemistry, 205: 803, 1953).

LaBarrere, J. A., J. R. Chipault, and W. O. Lundberg

1958. Cholesteryl esters of long-chain ★ fatty acids. Infrared spectra and sep- ● aration by paper chromatography. Ana- ■ lytical Chemistry, 30: 1466-1470.

Methods are described for the paper chromatographic separation of the individual cholesteryl esters from mixtures containing both saturated and unsaturated esters.

10 to 20 μg. of each compound are required. Data on melting points, specific rotation, and infrared and near-infrared spectra are included.

Lambert, M. and A. C. Neish
1950. Rapid method for estimation of glycerol in fermentation solutions. Canadian Journal of Research, 28B: 83-89.

Glycerol is oxidized to formaldehyde with periodic acid, the iodate and periodate formed are reduced to iodide with sodium arsenite, and formaldehyde is determined on the oxidation mixture by the chromotropic acid color reaction.

Lea, C. H. and D. N. Rhodes
1953. Phospholipins. 1. Partition chromatography of egg-yolk phospholipins on cellulose. Biochemical Journal, 54: 467-469.

The authors were unable to confirm the report of Bevan, et al (Journal of the Chemical Society, p. 841, 1951) that ethanolamine and choline-containing phospholipids were separable by partition chromatography on paper or cellulose columns. However, amino acid contaminants of the phospholipids in ether or chloroform solution were removed by this method.

Lea, C. H. and D. N. Rhodes
1954. Phospholipids. 2. Estimation of amino nitrogen in intact phospholipids. Biochemical Journal, 56: 613-618.

A modification of the ninhydrin method of Moore and Stern (Journal of Biological Chemistry, 176: 367, 1948) for the estimation of phosphatidylethanolamine in the presence of phosphatidylcholine. Preliminary hydrolysis of the phospholipid is unnecessary.

Lea, C. H. and D. N. Rhodes
1954. Determination of the iodine value of phospholipids. Analyst, 79: 304-305.

The Rosenmund-Kuhnhenn method was found to give results 2 to 10% low on samples of pure methyl esters, and the Yasuda method gave results 4 to 14% low.

Lea, C. H., D. N. Rhodes, and R. D. Stoll
1955. Phospholipids. 3. On the chromatographic separation of glycerophospholipids. Biochemical Journal, 60: 353-363.

Phosphatidyl ethanolamine and phosphatidyl choline were prepared from egg yolk phospholipids by silicic acid chromatography using $CHCl_3$-MeOH as eluting solvent. Their lyso-equivalents were prepared from venom-treated egg yolk phospholipids in a similar manner. A method and discussion of chromatography of egg yolk phospholipids on silicic acid-impregnated paper is also given.

Egg lecithin prepared by chromatography on alumina separated into lecithin and lysolecithin when rechromatographed on silicic acid.

Lea, C. H.
1955. Some observations on the preparation and properties of phosphatidyl ethanolamine. Proceedings of the International Conference on Biochemical Problems of Lipids, 2nd Ghent. (Pub. 1956) pp. 81-90.

The changes which occur during autooxidation of phosphatidyl-ethanolamine are discussed.

Deoxygenation of the SiO_2 column before chromatography of phospholipids led to decreased formation of oxidation products.

Lees, M.
1956. Simple procedure for the isolation of brain sulfatides. Federation Proceedings, 15: 298.

Brain white matter is extracted with $CHCl_3$-MeOH and the extract is washed according to Folch, et al (Federation Proceedings, 13: 209, 1954). The extract is purified by reextraction and the sulfatide is precipitated at -10°.

Leffler, H. H.
1959. Estimation of cholesterol in serum. American Journal of Clinical Pathology, 31: 310-313.

Isopropyl alcohol is used for the simultaneous precipitation of serum proteins and the extraction of total cholesterol. Free cholesterol is precipitated as the digitonide

from an aliquot. $FeCl_3$ in 87% H_3PO_4 is used as color reagent.

Leupold, F. and H. Buttner

1953. The hydrolysis of acetals of higher fatty aldehydes. Zeitschrift für physiologische Chemie, 292: 7-13. Chemical Abstracts, 48:11306b (1954).

A study was made of the hydrolysis of dimethyl acetals of palmitaldehyde and stearaldehyde in AcOH and AcOH-2 N HCl mixtures. The hydrolysis of the aldehydes was found to be incomplete in 99-100% AcOH, but proceeded readily in 90% AcOH. The aldehydes were determined photometrically at 293 mμ. after extracting with cyclohexane and drying the extract with Na_2SO_4.

Leupold, F. and D. Eberhagen

1959. A simple method for the separation of unsaturated acids from small amounts ★ of lipides. Fette, Seifen, Anstrichmit- ● tel, 60: 809-811. Chemical Abstracts, 53:3738d (1959).

Polyunsaturated fatty acids were separated by low-temperature urea fractionation.

Levene, P. A. and I. P. Rolf

1927. The preparation and purification ★ of lecithin. Journal of Biological Chem- ▲ istry, 72: 587-590.

Methods are given for the extraction and purification of lecithin from egg yolk, brain, and liver tissues. The lecithin is precipitated from ethanol with $CdCl_2$ and purified by precipitation from chloroform solution with methanol-gaseous ammonia. Analyses of purified products are given.

Levine, C. and E. Chargaff

1951. Procedures for the microestimation of nitrogenous phosphatide constit- ▲ uents. Journal of Biological Chemistry, 192: 465-479.

Ethanolamine, serine, and choline, in amounts of 5-75 μg., were determined as follows: The phosphatides were hydrolyzed, the fatty acids were removed, and the bases were separated by paper chromatography. Ethanolamine and serine were located on the developed chromatogram with ninhydrin and estimated by colorimetry of the eluted spots with ninhydrin. Choline spots were converted to choline phosphomolybdate, reduced to molybdenum blue, and the spots were measured by planimetry.

Levine, C. and E. Chargaff

1951. Chromatographic behavior of analogues of the nitrogenous lipide constituents. Journal of Biological Chemistry, 192: 481-483.

Analogues of ethanolamine, serine, and choline were separated by paper chromatography using several solvent systems, and estimated by the ninhydrin reaction.

Liberti, A., G. P. Cartoni, and U. Pallotta

1958. Vapor-phase chromatography of methyl esters of fatty acids and their ★ quantitative determination by automatic ● coulometry. Annali di Chimica (Rome) 48: 40-49. Chemical Abstracts: 52: 9869e (1958).

Methyl esters were separated on a silicone grease column using glass powder as a support and burned to CO_2 at the exit. The CO_2 was passed into an ethanolic solution of $BaCl_2$, $Ba(OH)_2$, and H_2O_2. The current used to maintain constant pH of the solution, which is a quantitative measurement of the CO_2 formed, is recorded graphically. Sensitivity of the method is 0.2 microequivalents, and accuracy is 2-5%.

Lieboff, S. L.

1928. A colorimetric method for the determination of lipoidal phosphorus in ▲ blood. Journal of Biological Chemistry, 80: 211-214.

Lipids are extracted with alcohol-ether, the solvent is evaporated, and organic matter is oxidized with H_2SO_4 and H_2O_2. Phosphorus is precipitated as uranium phosphate, dissolved in trichloracetic acid, converted to uranium ferrocyanide, and the ferrocyanide is measured colorimetrically.

Lipmann, F. and L. C. Tuttle

1945. A specific micromethod for the determination of acyl phosphates. Jour- ▲ nal of Biological Chemistry, 159: 21-28.

Acyl phosphates are determined by conversion to hydroxamic acid derivatives and

measurement of the color produced by the ferric hydroxamate complex.

Lips, H. J.
1953. Observations on the determination of iodine value with pyridine sulfate dibromide. Journal of the American Oil Chemists Society, 30: 399-403. Chemical Abstracts, 47:12843b (1953).

A study of the effects of variables (reaction time, reagent concentration, catalyst, solvent, peroxides, and sulfur) on iodine values determined by the pyridine sulfate dibromide method of Benham and Klee (Journal of the American Oil Chemists Society, 27: 127-130, 1950).

Lipsky, S. R. and R. A. Landowne
1959. Evaluation of a stationary phase for fatty acid analysis by gas-liquid chromatography. Annals of the New York Academy of Sciences, 72 (article 13): 666-674.

The saturated and unsaturated fatty acids of chain length C_{12}-C_{26} were quickly separated on a diethylene glycol-succinate polyester column by gas chromatography.

Lipsky, S.R., R. A. Landowne, and J. E. Lovelock
1959. Separation of lipides by gas-liquid chromatography. Analytical Chemistry, 31: 852-856.

A method is described for separation of methyl esters of fatty acids to C_{22} by gas-liquid chromatography on a capillary column coated with Apiezon L, using an argon detector. The minimum amount of organic vapor detectable by the method is approximately 10^{-15} mole.

Long, C. and D. A. Staples
1959. Determination of neuraminic acid in crude brain lipids. Biochemical Journal, 73: 385-389.

Brain tissue lipids were extracted with $CHCl_3$-MeOH (2:1) and partitioned according to Folch, et al (Journal of Biological Chemistry, 226: 497, 1957). The neuraminic acid content of the aqueous methanol phase was determined by measuring the extinction value of the orcinol reaction products before and after treatment of the sample with acid. A discussion of the method is given.

Lovern, J. A.
1950. Estimation of choline as the reineckate. Chemistry and Industry, p. 707. Chemical Abstracts, 45:3448i (1951).

The method of Marenzi and Cardini (Journal of Biological Chemistry, 147: 363, 1943) for the oxidative determination of choline reineckate will give more stable results if the acetone is evaporated from the reineckate precipitate prior to oxidation.

Lovern, J. A.
1952. The application of counter-current distribution to the separation of phospholipins. Biochemical Journal, 51: 464-470.

In an aqueous ethanol-petroleum ether system, the separation of phosphatidylcholine from ethanolamine and serine phospholipids was fair, but separation of phosphatidyl ethanolamine from phosphatidyl serine was poor.

Lovern, J. A. and J. Olley
1953. The lipids of fish. 2. The acetone-soluble lipids in the flesh of the haddock. Biochemical Journal, 54: 128-137.

The acetone-soluble lipids are fractionated by precipitation with acetone at 0° from an ether or chloroform solution. The soluble portions were fractionated further by counter-current distribution between petroleum ether and 85% ethanol.

Lovern, J. A. and J. Olley
1953. The lipids of fish. 4. The lipids extracted by an ethanol:ether mixture from haddock flesh previously extracted with acetone. Biochemical Journal, 55: 686-696.

An ethanol-ether (3:1) extract of haddock flesh which had been extracted previously with acetone was reextracted with petroleum ether. The petroleum ether extract was washed and the component lipids were fractionated by counter-current distribution between petroleum ether and 85% ethanol.

Lovern, J. A.

1956. The lipids of fish. 8. The triglycerides and cholesterol esters of haddock flesh. Biochemical Journal, 63: 373-380.

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The acetone extract of haddock flesh, containing the cholesterol esters and triglycerides, was chromatographed on silicic acid with petroleum ether, petroleum ether-benzene, and benzene-ether as eluting solvents. Fair separation of the hydrocarbons, cholesterol esters, and triglycerides was achieved.

Luecke, R. W. and P. B. Pearson

1944. The determination of free choline in animal tissues. Journal of Biological Chemistry, 155: 507-512.

Lecithin is removed by precipitation with acetone. Free choline is then separated by adsorption on Decalso and elution with 5% NaCl, and assayed with a cholineless strain Neurospora crassa.

The method gave results in agreement with reineckate precipitation method for the total choline in liver, but chemical methods were not sensitive enough for determination of free choline.

Lynn, W. S., Jr., L. A. Steele, and E. Staple

1956. Separation of 2, 4-dinitrophenylhydrazones of aldehydes and ketones by paper chromatography. Analytical Chemistry, 28: 132-133.

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The 2, 4-dinitrophenylhydrazones were separated by chromatography on phenoxyethanol-impregnated paper using heptane as mobile phase.

MacGee, J.

1959. Enzymatic determination of polyunsaturated fatty acids. Analytical Chemistry, 31: 298-302.

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A method is described for the determination of total polyunsaturated fatty acids by oxidation of the potassium salts of the fatty acids with atmospheric oxygen in the presence of lipidoxidase, and spectrophotometric measurement of the conjugated product.

(Essentially the same as MacGee, et al, Essential Fatty Acids. Proceedings of the International Conference on Biochemical Problems of Lipids, 4th Oxford, 1957, (Pub. 1958) pp. 21-29.)

McKibbin, J. M. and W. E. Taylor

1949. The nitrogenous constituents of the tissue lipids. I. The extraction, purification, and hydrolysis of tissue lipids. Journal of Biological Chemistry, 178: 17-27.

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Tissue was extracted with ethanol-ether (3:1), followed by a six-hour continuous extraction with chloroform. Non-lipid impurities were removed by emulsification of a chloroform extract with a 0.25 M $MgCl_2$, freezing to break the emulsion, and removal of the $MgCl_2$ solution. A five-hour $Ba(OH)_2$ hydrolysis was used to free combined choline.

McKibbin, J. M. and W. E. Taylor

1949. The nitrogenous constituents of the tissue lipids. II. The determination of sphingosine in tissue lipid extracts. Journal of Biological Chemistry, 178: 29-35.

The lipid extract was hydrolyzed with saturated $Ba(OH)_2$ and refluxed with HCl, and the liberated sphingosine was extracted with $CHCl_3$. The sphingosine was then digested for N according to Koch and McMeekin (Journal of the American Chemical Society, 46: 2066, 1924) and determined colorimetrically.

The extraction method is claimed to be quantitative and specific in the presence of other lipid bases. The method was used routinely for a range of 6 to 20 μmoles of sphingosine.

McKibbin, J. M.

1959. Determination of inositol, ethanolamine, and serine in lipides. Methods of Biochemical Analysis, D. Glick, editor, New York, Interscience Pub. Inc., Vol. 7, pp. 111-143.

▲

Methods are described for determination of lipid inositol by bioassay with Saccharomyces carlsbergensis, and colorimetric determination of ethanolamine and serine with sodium 1, 2-naphthoquinone-4-sulfonate after sealed-tube hydrolysis with 4 N HCl and separation on Permutit.

Various other methods for determination of inositol, ethanolamine, and serine are discussed.

MacLachlan, P. L.

1944. Determination of the iodine number of whole phospholipid. Journal of Biological Chemistry, 152: 97-102.

▲

When chloroform was used to dissolve phospholipid which had been precipitated with acetone and $MgCl_2$ (Yasuda, Journal of Biological Chemistry, 94: 410, 1931-32) the iodine numbers of the phospholipid were erratic. Reliable iodine numbers were obtained when the chloroform solution of phospholipid was evaporated to dryness and the phospholipid redissolved in chloroform. Reliable iodine numbers were also obtained when chloroform-ether (1:1) or moist ether were used as solvents, but neither was as good a solvent for the phospholipid as chloroform. It is suggested that the erratic values with the original chloroform solution are due to the formation of a phospholipid-$MgCl_2$ complex which is partially soluble in chloroform.

MacLean, H.

1914. A simple method for the preparation of lecithin. Journal of Pathology and Bacteriology, 18: 490-494.

★ ▲

Lecithin was prepared by a series of precipitations with acetone from an alcohol-water emulsion.

Ma, T. S. and J. D. McKinley

1953. Determination of phosphorus in organic compounds: A new microprocedure. Mikrochimica Acta, 1: 4-13 (In English). Chemical Abstracts, 47: 9858a (1953).

▲

A method is described for determination of organic phosphorus using the yellow phosphovanadomolybdate color in place of molybdenum blue.

Machebouef, M. A. and J. L. Delsal

1942. Colorimetric determination of small quantities of free or esterified cholesterol. Bulletin de la Société de chimie biologique, 24: 296-309. Chemical Abstracts, 40: 103^9 (1946).

■

A discussion of factors affecting the development of maximum color in the Liebermann-Burchard reaction.

Mai, S. H.

1951. Ultramicromethod for separation and determination of fatty acids on nylon thread. Federation Proceedings, 10: 388.

★ ●

A method is described for the separation of straight-chain, even numbered, saturated fatty acids from C_8 to C_{18} by chromatography on nylon thread. The acids are separated into groups, and the individual members of the groups are separated by use of various solvent mixtures for development. The separated acids are estimated by the monolayer film method. Suitable for determination of 2-6 μg. of each acid in a mixture of the acids.

Makita, T.

1958. Stability for oxidation of unsaturated fatty acids in their urea adduct crystals. Review of Physical Chemistry of Japan, 28: 31-35. (In English). Chemical Abstracts, 53: 14546b (1959).

●

The urea adduct of unsaturated fatty acids becomes unstable with oxidation in air at 80°, but stabilizes at higher pressures and protects the acid from oxidation by air.

Mallov, S., J. M. McKibbin, and J. S. Robb

1953. The distribution of some of the essential lipides in beef heart muscle and conducting tissue. Journal of Biological Chemistry, 201: 825-837.

Modifications of the anthrone colorimetric carbohydrate procedure for use in lipid sugar determination are given. The method was found to be more satisfactory than other reduction and colorimetric procedures. Attempts to apply the selective hydrolysis and reineckate precipitation methods for sphingomyelin to beef heart lipid extracts were unsuccessful. The Brand-Sperry (Journal of Biological Chemistry, 141: 545, 1941) sugar reduction method and the Brückner (Zeitschrift für physiologische Chemie, 286: 163, 1941) orcinol sugar color method were found to be unsatisfactory for beef heart lipid sugar determination.

Man, E. B. and E. F. Gildea

1932. A modification of the Stoddard and Drury titrimetric method for the determination of the fatty acids in blood serum. Journal of Biological Chemistry, 99: 43-60.

The alcohol-ether extract of serum is saponified, and the fatty acids are freed with HCl and titrated with NaOH.

Bloor's extraction procedure gave incomplete extraction.

Man, E. B. and J. P. Peters

1933. Gravimetric determination of serum cholesterol adapted to the Man and Gildea fatty acid method, with a note on the estimation of lipoid phosphorus. Journal of Biological Chemistry, 101: 685-695.

A micro modification of the digitonin precipitation method which utilizes the solution left from the Man and Gildea (Journal of Biological Chemistry, 99: 43, 1932) fatty acid determination. Phospholipid is determined by the Fiske-Subbarow method on an aliquot of the same solution.

Man, E. B.

1937. A note on the stability and quantitative determination of phosphatides. Journal of Biological Chemistry, 117: 183-187.

Phosphatide decomposition occurs during the evaporation in air of solvents from solutions of phospholipids. Evaporation under N_2 at low pressure and temperature avoids the decomposition.

Man, E. B. and E. F. Gildea

1937. Notes on extraction and saponification of lipids from blood and blood serum. Journal of Biological Chemistry, 122: 77-88.

No significant difference was found in the use of NaOH or KOH for saponification, contrary to earlier findings.

Refluxing of blood with alcohol-ether for 1 hour gave greater yield of lipid phosphorus than heating for 1 to 5 minutes, and higher yields were also obtained when the refluxing was carried out in an inert atmosphere.

Mangold, H. K., B. G. Lamp, and H. Schlenk

1955. Indicators for the paper chromatography of lipids. Journal of the American Chemical Society, 77: 6070-6072.

Indicators are described for the detection of various lipids on paper chromatograms.

Marcali, K. and W. Rieman, III

1946. Microdetermination of the saponification number of fats and oils. Industrial and Engineering Chemistry, Analytical Edition, 18: 144-145.

A description of methods for determination of the saponification number of fats and oils using samples of about 500, 50 and 15 mg.

Marenzi, A. D. and C. E. Cardini

1943. Colorimetric micromethod for determining total and unsaturated fat acids of blood. Revista de la Sociedad Argentina de Biologia, 19: 118-130. Chemical Abstracts, 38: 563^6 (1944).

Blood lipids are extracted and the fatty acids are converted to Pb soaps. The Pb is then precipitated from the soaps as $PbCrO_4$ and the Cr is determined.

Marenzi, A. D. and C. E. Cardini

1943. The colorimetric determination of choline. Journal of Biological Chemistry, 147: 363-370.

Beattie's method (Biochemical Journal, 30: 1554, 1936) was modified as follows: 1) 60% acetone in water was used to dissolve the reineckate, eliminating the problems of evaporation caused by use of acetone alone. Lower concentrations of acetone did not dissolve the salt completely, or else gave rise to a precipitate on standing. 2) The choline reineckate was precipitated by a 20 minute immersion in ice water, as precipitation was found to be complete under those conditions.

A new method for the determination of choline was proposed, based on the precipitation of choline as the reineckate and colorimetric determination of the chromium in the precipitate, which is sensitive to 15 μg. of choline. See also: Lovern, Chemistry and Industry, 707, 1950.

Marenzi, A. D. and C. E. Cardini

1943. On the determination of the phospholipids in blood. Journal of Biological Chemistry, 147: 371-378.

▲

Total lipid phosphorus was determined by the Fiske-Subbarow method, sphingomyelin by determination of the phosphorus content of its reineckate, and choline by determination of the chromium content of its reineckate. Satisfactory results were not obtained by colorimetric measurement of sphingomyelin reineckate.

Marinetti, G. V. and E. Stotz

1955. Paper chromatographic separation of phospholipids. Journal of the American Chemical Society, 77: 6668-6670.

★ ▲

Phosphatidylethanolamine and acetal phospholipid were acylated with acetic anhydride, benzoyl chloride, or 2, 4-dinitroflourobenzene and separated by chromatography on paper with solvent mixtures containing lutidine, acetic acid, and methanol or octanol.

Marinetti, G. V. and E. Stotz

1956. Chromatography of phosphatides on silicic acid impregnated paper. Biochimica et Biophysica Acta, 21: 168-170.

★ ▲

Phosphatides were separated on silicic acid-impregnated paper using diisobutyl ketone-acetic acid-water (40:30:7) or n-butyl ether-acetic acid-chloroform-water (40:35:6:5) as solvents.

Marinetti, G. V., J. Erbland, and J. Kochen

1957. Quantitative chromatography of phosphatides. Federation Proceedings, 16: 837-844.

★ ▲

Methods are described for the separation of phosphatides on silicic acid-impregnated paper and silicic acid columns. Diisobutyl ketone-acetic acid-water and diisobutyl ketone-n-butyl ether-acetic acid-water, in various ratios, are used as developing solvents.

Washing the silicic acid with methanol-chloroform before use gave more reproducible results than activation by heating. (cf. Kay and Trueblood, Analytical Chemistry, 26: 1566, 1954).

Marinetti, G. V., J. Erbland, and E. Stotz

1959. The quantitative analysis of plasmalogens by paper chromatography. Biochimica et Biophysica Acta, 31: 251-252.

★ ▲

Lecithin and phosphatidylethanolamine are isolated by silicic acid chromatography and hydrolyzed with acetic acid. The resulting lysophosphatides are separated by paper chromatography. The plasmalogen content of the original lipid is calculated from the amount of lipid phosphorus in the lysolecithin or lysophosphatidylethanolamine.

Marinetti, G. V., M. Albrecht, T. Ford, and E. Stotz

1959. Analysis of human plasma phosphatides by paper chromatography. Biochimica et Biophysica Acta, 36: 4-13.

★ ▲

Plasma lipids were extracted and chromatographed on silicic acid-impregnated paper using diisobutyl ketone-acetic acid-water (40:25:5) as solvent. The phosphatide spots were extracted with methanolic HCl and digested with perchloric acid and phosphorus was determined by spectrophotometric measurement of the phosphomolybdate.

Marquardt, P. and G. Vogg

1952. A sensitive assay of choline and acetylcholine by means of sodium tetraphenylboron. Zeitschrift für physiologische Chemie, 291: 143-147. Chemical Abstracts, 48: 12855e (1954).

▲

Choline or acetylcholine is precipitated quantitatively by tetraphenylboron in acid solution. Data on infrared absorption spectra and solubilities of the complexes are given.

Mata, M.

1948. Simplified determination of blood lipides. Revista Farmaceutica de Cuba, 26: 29-31. Chemical Abstracts, 43: 3871i (1949).

■

The sample of blood is dried on filter paper. The lipids are extracted with Et_2O-EtOH (5:1), dried, and weighed. Cholesterol is extracted from the residue with

$CHCl_3$ and determined by the Liebermann-Burchard reaction.

Matthews, F. W., G. G. Warren, and J. H. Michell
1950. Derivatives of fatty acids. Analytical Chemistry, 22: 514-519.
●
X-ray diffraction powder patterns of silver salts, amides, and anilides of fatty acids were studied and found to be suitable for fatty acid identification. Methods for preparation of the derivatives and data on their X-ray diffraction are given.

Mead, J. F.
1957. The metabolism of the essential fatty acids. VI. Distribution of unsaturated fatty acids in rats on fat-free supplemented diets. Journal of Biological Chemistry, 227: 1025-1034.
★
Unsaturated fatty acids were chromatographed on siliconized Celite, hydrogenated, and rechromatographed. Acids with differing degrees of unsaturation were well separated.

Meredith, P. and H. G. Sammons
1952. Horizontal paper chromatography. Analyst, 77: 416-418.
★
A method for horizontal paper chromatography is described. The method was used to separate the water-soluble constituents of a methanolic-HCl hydrolysate of nerve lipids.

Meredith, P. and H. G. Sammons
1959. A reaction between meso-inositol and uranyl acetate. Analyst, 83: 686.
Inositol can be detected on paper chromatograms by its flourescence when sprayed with uranyl acetate.

Michaels, G. D., G. Fukayama, H. P. Chin, and P. Wheeler
1958. Technics for separation of plasma cholesterol esters for determination of iodine value, and of cholesterol. Proceedings of the Society for Experimental Biology and Medicine, 98: 826-829.
★
■
Cholesterol esters are separated from blood lipids by chromatography on silicic acid. Iodine number is determined by a colorimetric modification of Yasuda's method. Cholesterol is determined by a modification of the Schoenheimer-Sperry method using an orcinol-ferric chloride color reagent.

Michaels, G. D.
1958. A note on increasing the accuracy of the micro alkaline isomerization technic for determination of unsaturated fatty acids. American Journal of Clinical Nutrition, 6: 593-594.
●
Absorption due to reagent blank in the method of Herb and Riemenschneider (Analytical Chemistry, 25: 953, 1953) is reduced by acidification of the isomerized fatty acids, extraction of the fatty acids with petroleum ether, evaporation of the extract to dryness, and dissolving the fatty acid residue in methanol for spectrophotometric reading.

Michaels, G. D., P. Wheeler, G. Fukayama, and H. P. Chin
1958. Column chromatographic fractionation of plasma lipids. American Journal of Clinical Nutrition, 6: 604-605.
★
●
■
A method is described for separation of cholesterol esters, free cholesterol, and mono-, di-, and triglycerides from blood plasma lipids by chromatography on silicic acid using petroleum ether and diethyl ether in varying concentrations as eluents. Recovery of added glycerides is quantitative.

Michaels, G. D., P. Wheeler, G. Fukayama, and L. W. Kinsell
1959. Plasma cholesterol fatty acids in human subjects as determined by alkaline isomerization and by gas chromatography. Annals of the New York Academy of Sciences, 72 (art. 13): 633-640.
●
Cholesterol was determined by a modification of the Schoenheimer and Sperry method using orcinol for color development.
A procedure is described for determination of unsaturated fatty acids in amounts of approximately 0.3 mg. by a modification (Michaels, American Journal of Clinical Nutrition, 6: 593, 1958) of the spectrophotometric method of Herb and Riemenschneider (Analytical Chemistry, 25: 953, 1953).

The microisomerization method of Holman (in Methods of Biochemical Analysis, D. Glick, Editor, 1957, vol. 4, p. 99) was found to give errors as high as 50%, presumably due to the high blank.

Michalec, C.

1957. The nature of cholesteryl esters in the higher fatty acids in human blood serum. Essential fatty acids. Proceedings of the International Conference on Biochemical Problems of Lipids, 4th Oxford, (Pub. 1958) pp. 105-110. Chemical Abstracts, 53: 17278h (1959).

★ ● ■

Cholesterol and cholesteryl esters were separated from blood lipid extracts by chromatography on paraffin oil-impregnated paper using an AcOH-$CHCl_3$-paraffin oil (65:25:10) mobile phase. Higher fatty acids were separated by using 93-95% AcOH as mobile phase. "Critical pairs" were separated by $KMnO_4$ oxidation of the unsaturated fatty acids prior to chromatography.

Michalec, C.

1958. Two-dimensional paper chromatography of higher fatty acids. Biochimica et Biophysica Acta, 28: 212-213.

★ ●

Fatty acids were separated by two-dimensional chromatography on paraffin oil-impregnated paper. The first chromatogram was run at 20° using 93% acetic acid, and the second chromatogram was run at -8° using formic acid-acetic acid-water as solvent.

Molines, J. and P. Desnuelle

1948. Colorimetric determination of phosphorus in oils and lecithins. Bulletin mensuel d'information ITERG, No. 2: pp 1-3. Chemical Abstracts, 42:4493d (1948).

▲

The sample is digested with HNO_3 and H_2SO_4 and phosphorus is determined colorimetrically, using metol as reducing agent for molybdic acid. As little as 0.01 mg. of phosphorus can be determined by this method.

Monasterio, G. and G. Gigli

1947. A new method for the study of the lipides of feces. Rassegna de fisiopatologica clinica e terapeutica (Pisa), 19: No. 11/12, 33 pp. Chemical Abstracts, 43:2658e (1949).

Lipids are extracted from feces with acetone and ether. Directions are given for the determination of the individual lipids.

Moretti, J. and J. Polonovski

1954. Chromatographic separation of bromine derivatives of stearic acid. Bulletin. Société chimique de France, pp. 935-936. Chemical Abstracts, 48: 13544a (1954).

★ ●

Bromo derivatives of stearic acid are separated by chromatography on paper or a cellulose or Celite column, and the compounds are eluted according to their solubilities in petroleum ether, ethyl ether, dichloroethylene and trichloroethylene.

Morgan, D. M. and K. J. Kingsbury

1959. A modified hydroxamic acid method for determining total esterified fatty acids in plasma. Analyst, 84: 409-414.

●

The esterified fatty acids in plasma were converted to their hydroxamates and the colored complex formed with ferric chloride was measured at 515 mμ. Variation ± 0.85%. Recoveries from glyceryl trioleate = 99.7%.

Moyle, V., E. Baldwin, and R. Scarisbrick

1949. Separation and estimation of saturated C_2-C_8 fatty acids by buffered partition columns. Biochemical Journal, 43: 308-317.

★ ●

The fatty acids are separated on a heavily buffered silica gel column with butanol-water mixtures as eluting solvents. The separated acids are estimated by titration. Effects of variables and applications of the method are discussed.

Mukerjee, H.

1959. Paper chromatography of volatile fatty acids. Analytical Chemistry, 31: 1284.

★ ●

MeOH-NH_3 and MeOH-acetone-NH_3 solvent mixtures are used to separate the ammonium salts of formic, acetic, propionic, and butyric acids by paper chromatography.

Munier, R.

1951. Partition microchromatography on paper of alkaloids and various biological nitrogenous bases. V. Separation of the nitrogenous constituents of the phosphoaminolipides; choline, ethanolamine and serine. Bulletin de la Société de chimie biologique, 33: 862-867. Chemical Abstracts, 46: 4172a (1952).

★ ▲

Good separation on paper was obtained by using BuOH-ethylene chlorohydrin concd. NH_4OH-H_2O (50:10:5:16), or BuOH saturated with H_2O and acidified with AcOH, as the mobile phase.

Nakamura, G. R.

1952. Microdetermination of phosphorus. Analytical Chemistry, 24: 1372.

▲

A micro modification of King's method (Biochemical Journal, 26: 292, 1932) for determination of 1-10 μg. of phosphorus.

Narayan, K. A. and B. S. Kulkarni

1954. Urea complexes of fatty acids. II. Effect of solvent dilution on complex formation. Journal of Scientific and Industrial Research (India) (B) 13: 9-15. Chemical Abstracts, 49: 5861h (1955).

●

The effects of H_2O dilution, urea concentration, and other variables on the formation of urea complexes of fatty acids are discussed.

Nelson, G. J. and N. K. Freeman

1959. Serum phospholipide analysis by chromatography and infrared spectrophotometry. Journal of Biological Chemistry, 234: 1375-1380.

★ ▲

A semimicro method is described in which the extracted lipids are separated by elution from a silicic acid-Celite column with methylene chloride, acetone, 35% methanol in methylene chloride, and methanol. The amounts of cephalin, lecithin, and sphingomyelin are determined by infrared spectrophotometry. Error of the method is approximately 10%, depending on the sample.

Ng, H., A. D. Webb, and R. E. Kepner

1956. Use of 2, 4-dinitrophenylhydrazones of p-phenylphenacyl esters as second derivatives of organic acids. Analytical Chemistry, 28: 1975-1977.

★ ●

A method is described for the separation and identification of the fatty acids from acetic to octadecanoic by chromatography as their p-phenylphenacyl-2, 4-dinitrophenylhydrazone derivatives on a silicic acid-nitromethane column. Nitromethane-saturated Skellysolve B is used as eluting solvent.

Nicolet, B. H. and L. A. Shinn

1941. The determination of serine by the use of periodate. Journal of Biological Chemistry, 139: 687-692.

▲

The formaldehyde formed from serine by the action of periodate as determined as the dimedon derivative. Accuracy of 2-3% was achieved with amino acid samples containing about 20 mg. of serine. The method is not suitable for use in the presence of carbohydrates or hydroxylysine, as they react with periodic acid to yield formaldehyde.

Niermierko, W.

1947. Micromethods for the determination of iodine and thiocyanogen numbers of fatty acids. Acta biologiae experimentalis (Warsaw) 14: 199-205. Chemical Abstracts, 42:9204i (1948).

●

A modification of the method of Rosenmund and Kuhnhenn (Zeitschrift für Untersuchung der Nahrungs- und Genussmittel, 46: 154, 1923) was used for the microdetermination of iodine number.

Niermierko, W.

1947. A micromethod for the determination of saturated acids. Acta biologiae experimentalis (Warsaw) 14: 207-209. Chemical Abstracts, 42:9205b (1948).

●

A modification of Bertram's method (Zeitschrift für Untersuchung der Lebensmittel, 55: 179, 1928) for use in microdetermination of saturated acids.

Nobil, E., M. G. Hagney, E. J. Wilder, and F. N. Briggs

1954. Simplified method for determination of total adrenal cholesterol. Proceedings of the Society for Experimental Biology and Medicine, 87: 48-50.

■ An adaptation of the Zlatkis, Zak, and Boyle (Journal of Laboratory and Clinical Medicine, 41: 486, 1953) procedure for use in determination of adrenal cholesterol. It is faster and simpler than the Liebermann-Burchard method.

Noda, M., O. Hirayama, and Y. Inouye

1956. Separation and identification of fatty acids. XIX. Paper-chromatographic analysis of component fatty acids of natural fats. Journal of the Agricultural Chemical Society of Japan, 30: 106-111. Chemical Abstracts, 52: 21166h (1958).

★ ● Unsaturated fatty acids were determined by a modified mercuric acetate addition method (Inouye, et al, Journal of the American Oil Chemists Society, 32: 132, 1955), higher saturated fatty acids by 2, 4-dinitrophenylhydrazide formation (Inouye and Noda, Bulletin of the Agricultural Chemical Society of Japan, 19: 214, 1955) and chromatography of their hydroxamic acids, and volatile fatty acids by chromatography of their hydroxamic acid derivatives.

Noda, M.

1959. Paper chromatography of unsaturated glycerides. Scientific Reports, Kyoto Prefectural Univ. Agr. No. 11, pp. 169-175.

★ ● A method is described for the separation and identification of unsaturated glycerides by paper chromatography of their mercuric acetate addition compounds.

Nojima, S. and N. Utsugi

1957. Quantitative determination of lipide-ethanolamine and lipide-serine and their distribution in rat and pig tissues. Journal of Biochemistry (Tokyo) 44: 565-573. Chemical Abstracts, 52: 1333b (1958).

▲ A specific and sensitive method is described for the determination of ethanolamine and serine in lipids. The lipids are extracted with ethanol-ether (3:1), hydrolyzed with 6 N NaOH, and determined by spectrophotometric measurement of their dinitrophenyl derivatives at 420mμ. Choline and inositol do not interfere, but amino acids will.

Norris, F. A. and R. J. Buswell

1944. Stability of Wijs solution for iodine number determinations. Industrial and Engineering Chemistry, Analytical Edition, 16: 417.

No significant change occurred during storage of Wijs solution in brown bottles for 505 days at room temperature.

O'Brien, R. G., et al

1938. Iodine value of oils and fats. Australian Chemical Institute Journal and Proceedings, 5: 329-334. Chemical Abstracts, 33: 420^{8} (1939).

The Wijs (Analyst, 54: 12, 1929), Rosenmund and Kuhnhenn (Zeitschrift für Untersuchung der Nahrungs- und Genussmittel, 46: 154, 1923), and Toms bromine vapor (Analyst 53: 69, 1928) methods were studied and compared. The R & K method gave low values and required frequent restandardization. The Toms method is recommended for its speed.

Okey, R.

1930. A micro method for the estimation of cholesterol by oxidation of digitonide. Journal of Biological Chemistry, 88: 367-379.

■ An adaptation of Bloor's oxidative procedure for lipids (Journal of Biological Chemistry, 77: 63, 1928) for use in the determination of cholesterol as the digitonide. See also: Yasuda, Journal of Biological Chemistry, 92: 303, 1931.

Olley, J.

1953. Effect of solute concentrations on countercurrent distribution of phospholipins. Biochimica et Biophysica Acta, 10: 493-498.

★ ▲ Variation in the concentration of the original solution of phospholipids causes a change in the distribution patterns.

Olley, J.

1955. Quantitative paper chromatography of lipid constituents. Proceedings of the International Conference on Biochemical Problems of Lipids, 2nd Ghent. (Pub. 1956) pp. 49-55.

★

A discussion of methods for estimation of the constituents of lipid hydrolysates by paper chromatography.

Ono, F. and Y. Toyama

1943. The highly unsaturated acids in fats and oils. I. Re-examination of the sodium salt-acetone and lithium salt-acetone methods for the separation of highly unsaturated acids of fish oils. Journal of the Chemical Society of Japan, 64: 1327-1331. Chemical Abstracts, 41:3755a (1947).

★
●

The lithium salt-acetone method was found to be better than the sodium salt-acetone method for separation of the highly unsaturated acids of fish oils.

Ory, R. L., W. G. Bickford, and J. W. Dieckert

1959. Glass paper chromatography of the long-chain fatty acids, brominated derivatives, and methyl esters. Analytical Chemistry, 31: 1447-1448.

★
●

A method is described for separation of long-chain fatty acids, both saturated and unsaturated. The methyl esters of the acids are brominated and chromatographed on glass paper impregnated with silicic acid. Isooctane is used as the developing solvent.

Alumina paper did not give as good results.

Page, E. and L. Michaud

1951. The titrimetric determination of plasma fatty acids. Canadian Journal of Medical Science, 29: 239-244.

●

Plasma lipids are extracted and saponified, and the fatty acids are freed with HCl and extracted. The fatty acids are then dissolved in an alcoholic solution of thymol blue and titrated with tetramethylammonium hydroxide.

Pangborn, M. C.

1941. A note on the purification of lecithin. Journal of Biological Chemistry, 137: 545-548.

★
▲

Lecithin is precipitated with $CdCl_2$ from an alcoholic solution, and purified by repeated extraction from an 80% alcohol-petroleum ether solution. Sphingomyelin and plasmalogen are not removed from the lecithin.

See Pangborn, Journal of Biological Chemistry, 188: 471, 1951, for modifications of the method.

Pangborn, M. C.

1942. Isolation and purification of a serologically active phospholipid from beef heart. Journal of Biological Chemistry, 143: 247-256.

★
▲

"Cardiolipin" was extracted from beef heart with methanol and purified by recrystallization of the $CdCl_2$ salt from alcohol and ether. It was identified as essential material for reactivity of beef heart antigens in the serological test for syphilis.

Pangborn, M. C.

1944. Acid cardiolipin and an improved method for the preparation of cardiolipin from beef heart. Journal of Biological Chemistry, 153: 343-348.

★
▲

Cardiolipin is extracted from the tissue with methanol (Pangborn, Journal of Biological Chemistry, 143: 247, 1942), precipitated with $BaCl_2$, and purified by reprecipitation from solvent solutions with $BaCl_2$ and $CdCl_2$. Yield of 0.6 grams was obtained from a kilogram of moist tissue.

Pangborn, M. C.

1945. A note on the preparation of cardiolipin. Journal of Biological Chemistry, 157: 691-692.

▲

Na_2SO_4 is recommended for decomposition of the crude barium salts of cardiolipin in place of the NaCl previously used (Pangborn, Journal of Biological Chemistry, 153: 343, 1944). The NaCl method does not give complete recovery of cardiolipin in a large scale preparation.

Pangborn, M. C.

1945. A simplified preparation of cardiolipin, with a note on purification of lecithin for serologic use. Journal of Biological Chemistry, 161: 71-82.

▲

A simplification of Pangborn (Journal of Biological Chemistry, 153: 343, 1944).

Pangborn, M. C.

1951. A simplified purification of lecithin. Journal of Biological Chemistry, 188: 471-476.

★ ▲

Lecithin is purified by repeated precipitation from alcohol with $CdCl_2$. Lecithin with an N:P ratio of 1.01:1 is obtainable. Yield: 10.5 gm. from 12 eggs.

Paquot, C.

1947. The determination of the saponification number of fats. Journal des recherches du Centre national de la recherche scientifique (Paris) pp. 131-135. Chemical Abstracts, 42:5689a (1948).

●

The double bonds of unsaturated aliphatic acids may react during saponification of triglycerides with alcoholic KOH, and lead to erroneous results.

Park, J. T. and M. J. Johnson

1949. A submicro determination of glucose. Journal of Biological Chemistry, 181: 149-151.

A very sensitive method. Glucose is determined by reduction of ferricyanide and colorimetric measurement. Range of 1-9 μg. of glucose in 1-3 ml. sample. The reaction is not specific for glucose.

Pearson, S., S. Stern, and T. H. McGavack

1953. A rapid, accurate method for the determination of total cholesterol in serum. Analytical Chemistry, 25: 813-814.

■

Glacial acetic acid, p-toluenesulfonic acid and acetic anhydride are added to 0.1 ml. of serum and allowed to cool without mixing. Con. H_2SO_4 is added and the solution is mixed. After 20 minutes, the optical density is measured at 550 mμ. Recovery of added cholesterol and cholesterol acetate = 99.2 $\pm$ 3.6%. Average variations in values for analyses of the same serum over a 24 hour period ranged from 0.4 to 2.4%. Average deviation of duplicates was less than 5%. Average deviation from values by the Schoenheimer-Sperry method $\pm$ 3.5%. Equimolar quantities of cholesterol and cholesteryl acetate give equal color densities by this method.

Perilä, O.

1956. Separation of saturated straight chain fatty acids. III. Quantitative paper chromatography. Acta chemica scandinavia, 10: 143-144.

★ ●

Solvent systems are described for separation of the fatty acids from formic to cerotic by paper chromatography.

Peterson, M. H. and M. J. Johnson

1948. The estimation of fatty acids of intermediate chain length by partition chromatography. Journal of Biological Chemistry, 174: 775-789.

★ ●

The fatty acids from formic to capric were separated on a Celite column using sulfuric acid as the stationary phase with Skellysolve B-benzene and butanol-chloroform as mobile phases. Higher fatty acids do not interfere.

Phillips, G. B.

1958. The isolation and quantitation of the principal phospholipid components of human serum using chromatography on silicic acid. Biochimica et Biophysica Acta, 29: 594-602.

★ ▲

A modification of the Lea, Rhodes, and Stoll chromatographic method (Biochemical Journal, 155: 19, 1944). Lecithin, sphingomyelin, lysolecithin, and phosphatidyl ethanolamine were isolated from human serum by chromatography on silicic acid using methanol and chloroform as eluents.

Extraction of serum with chloroform-methanol (1:1) was found to be as effective as Bloor's alcohol-ether (3:1) extraction.

Phillips, G. B. and N. S. Roome

1959. Phospholipides of human red blood cells. Proceedings of the Society for Experimental Biology and Medicine, 100: 489-492.

★ ▲

Red blood cell phospholipids were separated into ethanolamine- and serine-containing phospholipids, lecithin, sphingomyelin and lysolecithin by chromatography of silicic acid.

Pierfitte, M. and J. Barrier

1959. Microdetermination of serum cholesterol by a reaction with ferric chloride. Bulletin de la Société de pharmacie de Nancy, No. 40, pp. 31-39. Chemical Abstracts, 54:1648e (1960).

■

A modification of the method of Zlatkis, et al (Journal of Laboratory and Clinical Medicine, 41: 486, 1953). The unmodified Zlatkis method gave results approximately 7% too high.

Pikaar, N. A. and J. Nijhof

1958. Microdetermination of the fatty acids in blood serum. Biochemical Journal, 70: 52-57.

●

The lipids in 2 ml. of serum are extracted with ethanol-ether (3:1) and saponified. The fatty acids are isomerized with KOH in ethylene glycol, and the amounts of each of the polyethenoid fatty acids are calculated from the difference in optical densities at various wave lengths before and after the isomerization.

Saturated fatty acids are determined by a modification of the method of van de Kamer (Biochemical Journal, 61: 180, 1955).

The method compares well with the method of Herb and Riemenschneider (Analytical Chemistry, 25: 953, 1953).

Pippen, E. L., E. J. Eyring, and M. Nonaka

1957. Chromatographic separation of 2, 4-dinitrophenylhydrazones. Analytical Chemistry, 29: 1305-1307.

★

Methods for the preparation and chromatography on silicic acid-celite of the 2, 4-dinitrophenylhydrazone derivatives of aliphatic aldehydes and ketones are described. Data on separation of various combinations of the derivatives are given.

Platt, B. S. and G. E. Glock

1943. The estimation of inositol in animal tissues. Biochemical Journal, 37: 709-712.

The ether-soluble-70% acetone-insoluble fraction is separated from an aqueous extract of tissue. Glucose is removed from the extract by yeast fermentation and acidic and basic substances are removed by ion-exchange resins. The free inositol is oxidized with HIO_4 and the excess acid is estimated iodometrically.

Pohle, W. D. and V. C. Mehlenbacher

1950. A modification of the periodic method for the determination of monoglycerides and free glycerol in fats and oils. Journal of the American Oil Chemists Society, 27: 54-56. Chemical Abstracts, 44:3272a (1950).

●

Free glycerol and monoglycerides are separated by extraction of the glycerol with water from a chloroform solution of the sample. Glycerol is determined on the water solution, and monoglycerides are determined in the chloroform solution by periodic acid oxidation.

Pollak, O. J. and B. Wadler

1952. Rapid turbidimetric assay of cholesterols. Journal of Laboratory and Clinical Medicine, 39: 791-794.

■

Free or total cholesterol is assayed by nephelometric measurement of the turbidity produced by cholesterol digitonide.

Polonovski, J., J. Etienne, M. Paysant, and M. Petit

1959. Chromatographic fractionation of plasma phospholipides. Annales de biologie clinique (Paris) 17: 186-192. Chemical Abstracts, 53:22181e (1959).

★
▲

A method is described for separation of plasma phospholipids by paper chromatography.

Popják, G.

1943. Colorimetric determination of total, free, and ester cholesterol in tissue extracts. Biochemical Journal, 37: 468-470.

■

A modified combination of the Kelsey (Journal of Biological Chemistry, 127: 15, 1939) and Schoenheimer-Sperry (Journal of Biological Chemistry, 106: 745, 1934) methods. Cholesterol is precipitated as the

digitonide and separated. The digitonide is then decomposed and the cholesterol is determined by colorimetric measurement of the color produced with Liebermann-Burchard reagent.

Privett, O. S., E. Breault, J. B. Covell, L. N. Norcia, and W. O. Lundberg
1958. Solubilities of fatty acids and derivatives in acetone. Journal of the American Oil Chemists Society, 35: 366-370. Chemical Abstracts, 52: 15095h (1958).

Solubilities of fatty acids, alcohols, triglycerides, and methyl esters in acetone were studied using purified materials. Data on solubility of the materials at different temperatures are given.

Quaife, M. L., R. P. Geyer, and H. R. Bolliger
1959. Rapid paper chromatographic microassay of free and ester cholesterol of blood. Analytical Chemistry, 31: 950-955.

A method is described for separation of free and ester cholesterol from blood serum by chromatography of an acetone-ethyl ether (1:1) extract on $ZnCO_3$-impregnated filter paper. The cholesterol-containing areas are eluted with $CHCl_3$ and cholesterol is determined on the eluate by a modified Zlatkis procedure. Effects of variables and possible interferences are discussed. Coefficient of variation of duplicates done on different days was 3.5%.

Radin, N. S., F. B. Lavin, and J. R. Brown
1955. Determination of cerebrosides. Journal of Biological Chemistry, 217: 789-796.

Interfering ions are removed by ion-exchange chromatography. Cerebrosides are determined by a modified anthrone procedure (Black, Analytical Chemistry, 23: 1792, 1951), after solution in phosphoric acid. Average recovery is 99.7% and standard deviation is 1.9%.

Radin, N. S. and J. R. Brown
1955. Preparative isolation of cerebrosides. Federation Proceedings, 14: 266.

Lipids from beef spinal cord are mixed with ether and Celite and filtered. The filter cake is air dried and extracted with hot alcohol. The filtrate is cooled and the resulting precipitate is filtered off, dissolved, and passed through a "Flourisil" column to remove the phospholipids. The eluate is dried, dissolved in chloroform-alcohol-water (8:10:1), and passed through an ion exchange resin mixture. The resulting cerebroside material assays 87% pure.

Radin, N. S.
1957. Glycolipide chromatography. Federation Proceedings, 16: 825-826.

In the method described, a mixture of brain lipids is passed through a "Flourisil" (activated magnesium silicate) column to remove phosphatides and gangliosides, after which the sulfatides are adsorbed on a mixture of ion exchange resins. The cerebrosides and other non-ionic materials are then chromatographed on an unsulfated Dowex 50 column. The sulfatides may be eluted from the ion exchange column with a chloroform-alcohol-water solution of lithium acetate and precipitated with $BaCl_2$.

Ramsay, W. N. M. and C. P. Stewart
1941. The analysis of blood phospholipins. Biochemical Journal, 35: 39-47.

The amounts of lecithin, sephalin, and sphingomyelin in blood are calculated from the analysis for phosphorus, choline, glycerol, and ethanolamine. Choline is determined as the reineckate, and glycerol as formaldehyde.

Ramsey, L.L. and W. I. Patterson
1945. Separation and identification of the volatile saturated fatty acids (C_1 to C_4). Journal of the Association of Official Agricultural Chemists, 28: 644-656.

A method is described for separation of the C_1-C_4 saturated fatty acids by chromatography on silicic acid. The fatty acids are eluted with butanol-chloroform, using water as the stationary phase, and identified by microscopic examination of their crystalline derivatives. n-Butyric and

isobutyric acids are not separated from each other. The method is capable of determining 1 mg. of an individual acid in a 5-10 mg. mixture of the acids.

Ramsey, L. L. and W. 1. Patterson
1948. Separation and determination of the straight-chain saturated fatty acids C_5 to C_{10} by partition chromatography. Journal of the Association of Official Agricultural Chemists, 31: 139-150.

C_5 to C_{10} fatty acids are separated by chromatography on silicic acid, using methanol and 2,2,4-trimethylpentane as solvent phases. Isomers of the same chain length are eluted together.

Ramsey, L. L. and W. I. Patterson
1948. Separation of the saturated straight-chain fatty acids C_{11} to C_{19}. Journal of the Association of Official Agricultural Chemists, 31: 441-452.

The fatty acids are separated by chromatography on silicic acid, using furfuryl alcohol-2 aminopyridine and n-hexane as solvent phases, and identified by titration with NaOH.

Rapport, M. M. and N. Alonzo
1955. Photometric determination of fatty acid ester groups in phospholipides. Journal of Biological Chemistry, 217: 193-198.

Phospholipid esters are converted to hydroxamic acids, and the color produced with acid ferric perchlorate is read spectrophotometrically at 530 mμ. The sensitivity can be increased by decreasing the amount of ferric perchlorate added. As little as 20 μg. of phospholipid can be measured by the method.

Carlson and Wadstrom (Scandinavian Journal of Clinical and Laboratory Investigation, 10: 407, 1958) found this method to be the simplest, with good accuracy and ready reproducibility. Hirsch (Journal of Biological Chemistry, 233: 213, 1958) recommends substitution of n-butanol as solvent for cholesterol esters, as it would circumvent the insolubility of reaction products in the ethanolic ferric perchlorate color reagent.

Rapport, M. M. and N. Alonzo
1955. Identification of phosphatidal choline as the major constituent of beef heart lecithin. Journal of Biological Chemistry, 217: 199-204.

Modifications of the Schiff test and Korey and Wittenberg's p-phenylhydrazone formation test (Federation Proceedings, 13: 244, 1954) are used for the identification of aldehydes. The methods are both quantitative, and are in correspondence with each other for phosphatidal choline analysis.

Rapport, M. M. and B. Lerner
1958. A simplified preparation of sphingomyelin. Journal of Biological Chemistry, 232: 63-65.

A method is given for the preparation of pure sphingomyelin from commercial beef heart lecithin by hydrolysis and solvent recrystallization.

Reid, R. L. and M. Lederer
1951. Separation and estimation of saturated C_2-C_7 fatty acids by paper partition chromatography. Biochemical Journal, 50: 60-67.

The acids are separated by chromatography of their ammonium salts with a n-butanol-aqueous ammonia solvent system. The spots are located by spraying with a bromocresol purple-formaldehyde solution and the acids are estimated by measurement of spot area. Accuracy ± 2 to 5%.

Rhodes, D. N. and C. H. Lea
1955. Chromatographic separation of glycerophospholipids. Proceedings of the International Conference on Biochemical Problems of Lipids, 2nd Ghent (Pub. 1956) pp. 73-79.

The glycerophospholipids were separated on silicic acid columns or silicic acid-impregnated paper using chloroform-methanol mixtures as eluting solvents.

Rhodes, D. N.
1958. Interference by polyunsaturated fatty acids in the determination of cholesterol. Biochemical Journal, 71: 26P

Highly purified fatty acid esters were found to give spurious colors in determination of

cholesterol by the ferric chloride method of Zlatkis, Zak, and Boyle (Journal of Laboratory and Clinical Medicine, 41: 486, 1953). Color yields, expressed as percentages of cholesterol value, varied from 0.047 to 6.5%.

Rice, E. W. and D. B. Lukasiewicz
1957. Influence of bromine in the Zak cholesterol method. Clinical Chemistry, 3: 160-162.
■
Traces of Br will give high values in the $FeCl_3$-H_2SO_4 color reaction for cholesterol. Shaking the serum with $AgIO_3$ before extraction with ethanol-dimethyl ketone will eliminate the Br interference.

Rice, F. A. H. and A. G. Osler
1951. Chromatographic purification and serologic studies of a beef heart lecithin. Journal of Biological Chemistry, 189: 115-121.
★ ▲
A method is described for the purification of lecithin for use in sero-diagnosis of syphilis. The lecithin is adsorbed on a "Magnesol" (hydrated magnesium acid silicate)-Celite column and eluted with 2% tertiary-butyl alcohol in benzene.

Riemenschneider, R. W., S. F. Herb, and P. L. Nichols, Jr.
1949. Isolation of pure natural linoleic acid and linolenic acids as their methyl esters by adsorption fractionation on silicic acid. Journal of the American Oil Chemists Society, 26: 371-374. Chemical Abstracts, 43:6839i (1949).
★ ●
The methyl esters were prepared from linoleic and linolenic acids from tobacco oil and linseed oil, respectively, and chromatographed on silicic acid-Celite with petroleum ether and petroleum ether-ethyl ether.

Riemenschneider, R. W., F. E. Luddy, and S. G. Morris
1958. Determination of fatty acids in small amounts of plasma and in lipid components of tissues by ultraviolet spectroscopy. American Journal of Clinical Nutrition, 6: 587-591.
●
An adaptation of the spectrophotometric method of Herb and Riemenschneider (Analytical Chemistry, 25: 953, 1953) for determination of the fatty acid composition of 0.5 to 2 ml. of plasma or of the fractions from a chromatographed (SiO_2) sample of tissue lipids.

Rigamonti, R. and V. Ricco
1954. Separation of fatty acids of different molecular weight by means of addition compounds with urea. Annali di Chimica (Rome) 44: 288-298.
●
Fatty acids of 90% purity may be separated from fatty acid mixtures by fractional precipitation of their urea complexes.

Rikimaru, M., Y. Tanaka, and M. Hoshino
1955. Paper chromatography of phospholipides. Fukushima Journal of Medical Science, 2: 131-133. Chemical Abstracts, 50: 13156d (1956).
★ ▲
A study of solvents suitable for the paper chromatography of lecithins and lysolecithin. Best results were obtained with a butanol-glycerol-water mixture.

Rikimaru, M.
1955. Chromatographic separation of lecithin and lysolecithin on silica-treated filter paper disk. Fukushima Journal of Medical Science, 2: 175-177. Chemical Abstracts, 50: 13147g (1956).
★ ▲
Lecithin and lysolecithin were separated on silica-impregnated filter paper using $CHCl_3$-MeOH or $CHCl_3$-EtOH as developing solvents. The bands were located with phosphomolybdic acid. R_f values are given.

Roberts, H. R. and W. Bucek
1957. Rapid procedure for separation of C_2 to C_6 volatile fatty acids by horizontal paper chromatography at elevated temperature. Analytical Chemistry, 29: 1447-1449.
★ ●
The C_2 to C_6 fatty acids were separated by horizontal paper chromatography of their ethylamine salts using butanol saturated with water as solvent and a temperature of 50° for development.

Roberts, W. L. and H. A. Schuette
1932. Determination of hydroxyl number

of oils, fats and waxes. Industrial and Engineering Chemistry, Analytical Edition, 4: 257-259.

A method is described in which the hydroxyl number is determined by reaction of the sample with acetic anhydride in a sealed tube, after which the excess anhydride is hydrolyzed and titrated.

Robins, E., O. H. Lowry, K. M. Eydt, and R. E. McCaman

1956. Microdetermination of phospholipides and sphingolipides in brain. ▲ Journal of Biological Chemistry, 220: 661-675.

Cephalins, lecithins, and sphingolipids were determined in 10 μg. of brain tissue by a modification of the method of Schmidt, et al (Journal of Biological Chemistry, 166: 505, 1946).

Roboz, E., W. C. Hess, R. R. DiNella, and W. Cevallos

1958. Determination of total lipids, cholesterol, and phospholipids in cerebro- ● ▲ spinal fluid. Journal of ■ Clinical Medicine, 52: 158-162.

Methods suitable for use in the determination of total lipids, free and esterified cholesterol, and phospholipids in spinal fluid are described.

Roman, W. III

1930. A chemical method for the determination of choline and some physicochemical data on choline and its salts. ▲ Biochemische Zeitschrift, 219: 218-231. Chemical Abstracts, 24:3029 (1930).

Choline is precipitated as choline periodide with I_2, and the excess I_2 is titrated with $Na_2S_2O_3$. Determines 5 μg. to 5 mg. of choline with accuracy of $\pm$ 5%.

Rose, A. R., F. Schattner, and W. C. Exton

1941. A method for determining blood cholesterol. ■ American Journal of Clinical Pathology, Supplement 5: 19-23.

Cholesterol is extracted with alcohol-acetone (1:1) and determined colorimetrically with the $ZnCl_2$-acetyl chloride reaction (Tschugaeff, Chemiker-Zeitung, 24: 542, 1900).

Rosen, H.

1957. A modified ninhydrin colorimetric analysis for amino acids. Archives of Biochemistry and Biophysics, 67: 10-15.

A modification of the method of Yemm and Cocking (Analyst, 80: 209, 1955) which avoids the use of unstable solutions of reduced ninhydrin.

Rosenmund, K. W. and W. Kuhnhenn

1923. A new method for the determination of the iodine number in fats and oils by the use of pyridine sulfate dibromide. Zeitschrift für Untersuchung der Nahrungs- und Genussmittel, 46: 154-159. Chemical Abstracts, 18:477 (1924).

The sample is brominated by using pyridine sulfate dibromide reagent, and the excess reagent is titrated iodometrically.

Rosenthal, H. L., M. L. Pfluke, and S. Buscaglia

1957. A stable iron reagent for the determination of cholesterol. Journal of ■ Laboratory and Clinical Medicine, 50: 318-322.

The use of phosphoric acid in place of glacial acetic acid as solvent for ferric chloride gave a more stable reagent for cholesterol determination. The reagent may be stored at room temperature instead of frozen, as is required by the acetic acid solution. The results found by using the new reagent agreed well with other methods.

Rosenthal, H. L., M. L. Pfluke, and J. Callerami

1959. The colorimetric estimation of serum fatty esters. Clinica Chimica Acta, ● 4: 329-333.

A modification of the Stern and Shapiro (Journal of Clinical Pathology, 6: 158, 1953) hydroxamic acid procedure which uses cholesterol acetate as a standard in place of olive oil.

Rossi, L., A. D. Marenzi, and R. Lobo

1942. Photometric microdetermination of the chromate ion. Anales de farmacia

y bioquimica (Buenos Aires) 13: 1-8. Chemical Abstracts, 36:6442[9] (1942).

A method for use with Reinecke precipitates. NaOH and H_2O_2 are added directly to a solution of the salt, the solution is acidified with H_2SO_4, diphenylcarbazide is added, and the solution is compared with a standard in a colorimeter.

Rouser, G., G. V. Marinetti, R. F. Witter, J. F. Berry, and E. Stotz

1956. Paper chromatography of phospholipides. Journal of Biological Chemistry, 223: 485-497.

★ ▲

Individual phospholipids were separated from each other and other lipids by paper chromatography using mixtures of lutidine and acetic acid with alcohols or chloroform as solvents. Factors influencing the mobility of phospholipids on paper were studied and are discussed. An explanation of the chromatographic behavior of phospholipids is presented.

Rudloff, E. von

1956. Periodate-permanganate oxidations. IV. Determination of the position of double bonds in unsaturated fatty acids and esters. Journal of the American Oil Chemists Society, 33: 126-128. Chemical Abstracts, 50:6815b (1956).

●

Unsaturated fatty acids were quantitatively oxidized with periodate-permanganate reagent. The oxidation products were separated by chromatography on silicic acid and identified.

Rusoff, I. I., R. T. Holman, and G. O. Burr

1945. Table of spectroscopic data on fats, fatty acids, and their esters. Oil and Soap, 22: 290-294. Chemical Abstracts, 40:224[7] (1946).

●

A table of spectroscopic data with 81 references.

Saifer, A. and O. F. Kammerer

1946. Photometric determination of total cholesterol in plasma or serum by a modified Liebermann-Burchard reaction. Journal of Biological Chemistry, 164: 657-677.

■

Acetic anhydride-dioxane (3:2) is used for simultaneous extraction of cholesterol and precipitation of protein in serum or plasma. Color is developed by addition of H_2SO_4 to the extract. Accuracy is ± 5%. The effects of the variables are given.

Saifer, A.

1951. Photometric determination of total and free cholesterol and the cholesterol ester ratio of serum by a modified Liebermann-Burchard reaction. American Journal of Clinical Pathology, 21: 24-32.

■

A modification of the total cholesterol method of Saifer and Kammerer (Journal of Biological Chemistry, 164: 657, 1946) for use in the determination of free cholesterol in serum.

Sakagami, T.

1958. A new colorimetric method for determination of sphingosine base in lipids. Journal of Biochemistry, (Tokyo) 45: 313-317. Chemical Abstracts, 52: 18598g (1958).

Sphingosine base is oxidized with lead acetate and the fatty aldehyde formed is determined by measurement of the color produced by the plasmal reaction.

Sakuri, H.

1954. Complexes of urea with fatty acids and their derivatives. I. Separation of fatty acids and their methyl esters by urea complexes, and consideration of the reaction mechanism. Journal of the Chemical Society of Japan, Industrial Chemistry Section, 57: 50-51. Chemical Abstracts, 49:2098b (1955).

★ ●

Stearic and oleic acid are more readily separated if urea complex methods are used on their methyl esters rather than the free acids.

Samuelson, G.

1953. The photometric determination of choline and choline derivatives. Journal of Pharmacy and Pharmacology, 5: 239-244.

Choline is determined by spectrophotometric measurement at 415 m . of an acetone solution of its 2,4,6-hexanitrodiphenylamine derivative. The color is stable and the method is accurate to within 2%.

Sato, Y., G. T. Barry, and L. C. Craig
1947. Identification of small amounts of organic compounds by distribution studies. VII. Separation and estimation of normal fatty acids. Journal of Biological Chemistry, 170: 501-507.

Counter-current distribution is used for the separation and quantitative estimation of the C_2-C_5 normal fatty acids to within 2-3%.

Schaffer, F. L., J. Fong, and P. I. Kirk
1953. Microgram and submicrogram determination of phosphate. Analytical Chemistry, 25: 343-346.

A method is described for determination of phosphorus in quantities from 5-7 μg. to 2 mμg. The sample is digested with H_2SO_4 in a sealed tube, and the phosphorus is converted to phosphomolybdic acid. The phosphomolybdic acid is extracted with octyl alcohol, reduced with stannous chloride, and measured spectrophotometrically.

Octyl alcohol extracts phosphomolybdic acid, but not appreciable amounts of molybdic acid; an improvement over the isobutyl alcohol used by Berenblum and Chain.

Schlenk, H. and R. T. Holman
1950. Separation and stabilization of fatty acids by urea complexes. Journal of the American Chemical Society, 72: 5001-5004.

The use of urea complexes for separation of fatty acids is described, and factors influencing the separation are discussed.

Urea complexes of unsaturated fatty acids are not subject to autooxidation.

Schlenk, H., J. L. Gellerman, J. A. Tillotson, and H. K. Mangold
1957. Paper chromatography of lipides. Journal of the American Oil Chemists Society, 34: 377-386. Chemical Abstracts, 51:15689c (1957).

Detailed procedures are given for the qualitative and quantitative paper chromatography of lipids, and applications of the methods. R_f values for 16 fatty acids are given.

Schmidt, G., J. Benotti, B. Herschman, and S. J. Thannhauser
1946. A micromethod for the quantitative partition of phospholipid mixtures into monoaminophosphatides and sphingomyelin. Journal of Biological Chemistry, 166: 505-511.

The monoaminophosphatides are removed by selective saponification with KOH, and the value of sphingomyelin phosphorus is calculated as the difference between phosphorus values of total phospholipid and saponified phospholipid.

Schmidt, G., L. Hecht, P. Fallot, L. Greenbaum, and S. J. Thannhauser
1952. The amounts of glycerylphosphorylcholine in some mammalian tissues. Journal of Biological Chemistry, 197: 601-609.

Glycerylphosphorylcholine was determined as the difference in the amount of choline reineckate obtained before and after 20 minute hydrolysis of aqueous tissue extracts with 10 N HCl. Recovery of added glycerylphosphorylcholine was quantitative within limits of error of the method ($\pm$ 5%).

Schmidt, G., B. Ottenstein, W. A. Spencer, C. Hackethal, and S. J. Thannhauser
1957. Quantitative partition of acetal phospholipides and of free lipide aldehydes. Federation Proceedings, 16: 832-835.

The lipid aldehydes are converted to Schiff-negative compounds by incubation for 16 hours in an aqueous emulsion at pH 8.3 and room temperature. The acetal phospholipids are then determined colorimetrically using Schiff's reagent.

Schmidt, G.
1958. Quantitative estimation of fatty acids on filter paper. Naturwissenschaften, 45: 41. Chemical Abstracts, 52:8851d (1958).

Mercuric acetate esters of the fatty acids are chromatographed on filter paper and located by conversion of the acetate to sulfide.

Schmidt, L. H.

1935. The nature of the difference in phospholipid content of oxalated and heparinized plasma. Journal of Biological Chemistry, 109: 449-453.

▲

The higher phospholipid content of heparinized plasma than of oxalated plasma is due to shrinkage of red blood cells and consequent increase in plasma volume of the oxalated blood.

Schmidt-Neilson, K.

1942. Microtitration of fats in amounts of 10^{-5} gram. Compte rendu des travaux du laboratoire Carlsberg, Serie chimique, 24: 233-246. Chemical Abstracts, 38:2225[7] (1944).

●

The sample is saponified, and the fatty acids are freed, extracted with toluene, and titrated with tetramethylammonium hydroxide.

Schmidt-Neilson, K.

1944. Microdetermination of the iodine number of fat in quantities of 10^{-5} gram. Compte rendu des travaux du laboratoire Carlsberg, Serie chimique, 25: 87-96. Chemical Abstracts, 40:2011[3] (1946).

●

The I number is determined by saturating the double bonds with Br and titrating excess Br iodometrically.

Schmidt-Neilson, K.

1944. Extraction and fractionation of the lipides in one milligram of tissue. Compte rendu des travaux du laboratoire Carlsberg, Serie chimique, 25: 97-105. Chemical Abstracts, 40:2173[7] (1946).

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●

Frozen tissue is sliced and the tissue is destroyed with KOH. Unsaponifiable lipids are extracted with toluene, the fatty acids are freed from their soaps by adding HCl, and the freed fatty acids are extracted with toluene.

Schon, H. and F. Gey

1956. Elution chromatographic separation of free and esterified cholesterols in organ and serum fats and direct colorimetric determination. Zeitschrift für physiologische Chemie, 303: 81-90. Chemical Abstracts, 50:16934f (1956).

★
■

Free and ester cholesterol were separated from fat by elution from an Al_2O_3 column with petroleum ether-ethanol mixtures. The cholesterol was estimated by the Tschugaeff reaction.

Schoenheimer, R. and W. M. Sperry

1934. A micromethod for the determination of free and combined cholesterol (cholesterol in blood). Journal of Biological Chemistry, 106: 745-760.

■

Free cholesterol is precipitated as the digitonide, and total cholesterol is precipitated as the digitonide after hydrolysis of the sample. The color is developed with acetic acid, acetic anhydride, and sulfuric acid, and read at 610-620 mμ. The method requires 0.2 cc. of serum or blood. Standard deviation for free cholesterol is 1.48%; for total cholesterol is 1.16%.

See Sobel and Meyer (Journal of Biological Chemistry, 157: 255, 1955) for a modification for use with free cholesterol.

Schuette, H. A. and S. Dal Nogare

1951. An oxidation-adsorption method for analysis of methyl ester fractions. Journal of the American Oil Chemists Society, 28: 229-231. Chemical Abstracts, 45:6402g (1951).

★
●

A method is described for the separation of saturated from unsaturated methyl esters of the fatty acids. The unsaturated components are oxidized with permanganate, and the oxidation products are adsorbed on a column of alumina which has been treated with bromothymol blue.

Schwarz, H. P.,, L. Dreisbach, R. Childs, and S. V. Mastrangelo

1957. Infrared studies on tissue lipides. Annals of the New York Academy of Sciences, 69: 116-130.

★

Tissue lipids were chromatographed on silicic acid with MeOH and $CHCl_3$-MeOH as eluting solvents, and the eluted fractions were analyzed by infrared spectrophotometry.

Seher, A.

1956. Determination of paper-chromatographically separated long-chain carboxylic acids by photometric means. ★ ● Fette, Seifen, Anstrichmittel, 58: 498-504. Chemical Abstracts, 52:4211h (1958).

Fatty acids are separated by paper chromatography of their copper soaps, and the separated soaps are eluted from the paper and determined by photometry and polarography.

Seki, T.

1958. Chromatographic separation of lower fatty acids. ★ ● Journal of Biochemistry (Tokyo) 45: 855-860. Chemical Abstracts, 53:6326d (1959).

A method is described for the separation of the lower fatty acids by chromatography on Amberlite IRC-50 resin.

Sheftel, A. G.

1944. Determination of total and free cholesterol. ■ Journal of Laboratory and Clinical Medicine, 29: 875-878.

Glacial acetic acid is used to produce a more stable color in the Liebermann-Burchard reaction.

Showell, J. S.

1959. A uniform basis for reporting analytical data on fatty materials. Journal of the American Oil Chemists Society, 36: 343-345. Chemical Abstracts, 53:18513b (1959).

A recommendation is presented for reporting of iodine number, saponification number, acid number, hydroxyl number, acetyl number, peroxide value, carbonyl oxygen, and oxirane oxygen in basic units in order to simplify the correlation of data from different sources. Equations for calculation of the numbers are given.

Siegel, L.

1945. The microbiological determination of choline. Science, 101: 674-675.

Constant weight is more easily attained if fritted glass filters are used in place of paper for filtration of the mold growth.

Silk, M. H. and H. H. Hahn

1954. The resolution of mixtures of C_{16}-C_{24} normal-chain fatty acids by reversed-phase partition chromatography. ★ ● Biochemical Journal, 56: 406-410.

An extension of the method of Howard and Martin (Biochemical Journal, 46: 421, 1950). C_{16}-C_{24} normal fatty acids are chromatographed on a paraffin-coated kieselguhr column with aqueous acetone as developing solvent.

Silk, M. H., H. H. Sephton, and H. H. Hahn

1954. South African pilchard oil. 2. Concentrates of highly unsaturated fatty acids and alcohols derived from South African pilchard oil. ● Biochemical Journal, 57: 574-577.

Urea adduct formation and lithium soap-acetone fractionation are used to separate the highly unsaturated fatty acids from pilchard oil. A discussion of the merits of each system and comparison of their effectiveness is given.

Silk, M. H. and H. H. Hahn

1954. South African pilchard oil. 3. The fatty acid composition of South ★ ● African pilchard oil. Biochemical Journal, 57: 577-582.

The techniques of lithium soap-acetone separation, distillation, chromatography, and urea adduct formation are used to subdivide the fatty acid fractions of pilchard oil. The composition of the oil can then be calculated without isolation of the individual acids.

Simmons, R. O. and F. W. Quackenbush

1953. Chromatographic separation of unsaturated fatty acids as their 2,4-dinitro- ★ benzenesulfenyl chloride derivatives. ● Journal of the American Oil Chemists Society, 30: 614-616.

The 2,4-dinitrobenzenesulfenyl chloride derivatives of unsaturated fatty acids were separated by chromatography on $MgSO_4$. Saturated acids did not form derivatives.

Sinclair, R. G. and M. Dolan

▲ 1942. The so-called ether-insoluble

phospholipids in blood and tissues. Journal of Biological Chemistry, 142: 659-670.

Acetone alone precipitated 40-70% of the phospholipid from plasma and tissues when 0.3 to 2 mg. of phospholipid in 1 cc. of petroleum ether and 7 cc. of acetone were used. Complete precipitation was achieved on addition of 1 drop of saturated or 0.1 cc. of 30% $MgCl_2 \cdot 6H_2O$ in 95% ethanol. Addition of more $MgCl_2$ caused incomplete precipitation. When $MgCl_2$ was not used for precipitation, all of the tissue phospholipid and most of the plasma phospholipid precipitated were soluble in moist ether. The percentage of ether-insoluble phospholipid precipitate increased in proportion to the amount of $MgCl_2$ used, up to about 20% for the tissue phospholipid, and 90% for the plasma phospholipid. The ether-insoluble portion was found to be a portion of the total phospholipid mixture, and not simply the sphingomyelins.

Sloot, W. J. T. A. K.

1939-40. Determination of total fat content in small amounts of tissue. Acta Neerlandica morphologiae normalis et pathologicae, 3: 406. Chemical Abstracts, 37:401[4] (1943).

Lipids are extracted from dried tissue with absolute EtOH and the solvent is evaporated. The dry lipid is oxidized with $K_2Cr_2O_7$ and Ag reagent, KI is added, and excess dichromate is titrated with $Na_2S_2O_3$.

Smits, G.

1957. Modification of the periodide method for the determination of choline. Biochimica et Biophysica Acta, 26: 424-427.

The choline periodide precipitate is dissolved in ethylene dichloride which contains a little iodine, and water is added. The extinction of the ethylene dichloride solution of choline periodide is measured at 365 mμ. The interfering I^{-3} ions remain in the water phase.

Snyder, F. and N. Stephens

1959. Simplified spectrophotometric determination of ester groups in lipids. Biochimica et Biophysica Acta, 34: 244-245.

A simplification of the hydroxamic acid-ferric perchlorate spectrophotometric method which is adapted for use with multiple samples. The color can be developed in 20 to 30 samples in 15 to 20 minutes. Range to 4.00 micro equivalents of ester.

Sobel, A. E., I. J. Drekter, and S. Natelson

1936. Estimation of small amounts of cholesterol as the pyridine cholesteryl sulfate. Journal of Biological Chemistry, 115: 381-390.

A method is described for determination of free, esterified, and total cholesterol in the same sample of blood serum. The free cholesterol is isolated as pyridine cholesteryl sulfate, and measured colorimetrically by the Liebermann-Burchard reaction. Esterified cholesterol is determined in the same manner after hydrolysis of the cholesterol esters. Recovery of 0.025 mg. of added cholesterol was quantitative ($\pm$ 4%), with better recovery when large amounts were used.

See also: Drekter, Journal of Biological Chemistry, 115: 391, 1936.

Sobel, A. E. and A. M. Mayer

1945. Improvements in Schoenheimer-Sperry method for determination of free cholesterol. Journal of Biological Chemistry, 157: 255-264.

Extraction of the serum at room temperature was found to be as good as the hot extraction of S & S. Fifty percent alcoholic digitonin gave a more workable precipitate than the aqueous digitonin of S & S. Precipitation of cholesterol was found to be complete in 3 hours at 37°.

Sobel, A. E., J. Goodman, and M. Blau

1951. Cholesterol in blood serum. Studies of microestimation as the pyridinium cholesteryl sulfate. Analytical Chemistry, 23: 516-519.

A method for determination of cholesterol as the pyridinium cholesteryl sulfate is described. Cholesterol is dissolved in CCl_4, and pyridine and chlorosulfonic acid are added. The resulting precipitate

is dissolved in acetic acid and estimated colorimetrically by the Liebermann-Burchard reaction. Values by this method are similar to those obtained with digitonin.

Sorrel, M. F. and R. Reiser
1957. Identification of some marine oil constituents by chromatography. Journal of the American Oil Chemists Society, 34: 131-134.
★
Marine oils were separated into fractions on a silicic acid column and the fractions were further separated by chromatography on silicic acid-impregnated paper. R_f values of several components are given.

Soyenkoff, B. C.
1952. An improved micromethod of phosphate determination. Journal of Biological Chemistry, 198: 221-227.
▲
A modification of the method of Soyenkoff (Journal of Biological Chemistry, 168: 447, 1947) which eliminates many interferences. 2-p-dimethylaminostyrylquinoline ethosulfate and ammonium molybdate are added to the digested samples and the resulting color is measured colorimetrically. Sensitivity of the method is about 15 times that of the Fiske-Subbarow method. Accuracy with serum filtrates is 2%.

Sperry, W. M. and R. Schoenheimer
1935. A comparison of serum, heparinized plasma, and oxalated plasma in regard to cholesterol content. Journal of Biological Chemistry, 110: 655-658.
■
The content of total and free cholesterol in oxalated blood plasma was found to be lower than in serum or heparinized plasma from the same blood sample. Cholesterol content of serum and heparinized plasma was the same.

Sperry, W. M.
1942. Electrophotometric microdetermination of phosphorus in lipide extracts. Industrial and Engineering Chemistry, Analytical Edition, 14: 88-90.
▲
A method is described for the determination of 2 to 25 μg. of lipid phosphorus using reduced concentrations of the Fiske-Subbarow reagents and spectrophotometric measurement at about 400 mμ. Reagent concentrations and temperature are not critical. Recovery of added phosphorus is quantitative.

Sperry, W. M. and F. C. Brand
1943. The colorimetric determination of cholesterol. Journal of Biological Chemistry, 150: 315-324.
■
A method for the determination of total cholesterol using the Liebermann-Burchard color reaction. Time of development and temperature are critical. Saponification with KOH is necessary, as esterified cholesterol develops a stronger color faster than free cholesterol.

Sperry, W. M. and M. Webb
1950. A revision of the Schoenheimer-Sperry method for cholesterol determination. Journal of Biological Chemistry, 187: 97-110.
■
Dilute (50%) alcoholic solution of digitonin was found to be preferable to aqueous solutions of digitonin, as the aqueous solutions may lose the ability to quantitatively precipitate cholesterol as they age. Precipitation of the digitonide is not complete in 3 hours at 37°. The precipitation appears to be independent of the amount of cholesterol present in normal sera. Recovery of 99-101% was obtained.

Sperry, W. M.
1954. A method for the determination of total lipids and water in brain tissue. Journal of Biological Chemistry, 209: 377-386.
●
Tissue is homogenized and an aliquot of the homogenate is weighed. Water is extracted with acetone and evaporated, and the tissue is dried to constant weight in a desiccator. Difference in tissue weight = water removed. Lipids are extracted and washed according to Folch, et al (Journal of Biological Chemistry, 191: 833, 1951) and weighed. This method for determination of water avoids errors from high temperature drying.

Sperry, W. M. and F. C. Brand

1955. The determination of total lipides in blood serum. Journal of Biological Chemistry, 213: 69-76.

Lipids are determined gravimetrically after $CHCl_3$-MeOH extraction and purification. The use of N_2 atmosphere helps to overcome partial insolubility of the lipid residue. Sperry's (Journal of Biological Chemistry, 209: 377, 1954) and Folch's (Federation Proceedings, 13: 209, 1954) purification procedures remove all urea and presumably all other water-soluble contaminants.

Sperry, W. M.

1955. Lipide analysis. In Methods of Biochemical Analysis (D. Glick, Editor). New York, Interscience Publ. Inc., Vol. II, pp. 83-111.

A description and discussion of methods of lipid analysis suitable for use with blood tissues.

Spiteri, J. and G. Nunez

1952. Partition chromatography of fat-soluble substances. Compte rendu, 234: 2603-2604. Chemical Abstracts, 47:941f (1953).

A method is described for the separation of higher fatty acids, alcohols, sterols, vitamins, etc. from a mixture by chromatography on triglyceride-impregnated filter paper.

Spiteri, J.

1954. Partition chromatography of fat acids. Bulletin de la Société de chimie biologique, 33: 1355-1362. Chemical Abstracts, 49:1325f (1955).

Good separations of higher fatty acids were obtained by chromatography on paraffin-impregnated filter paper.

Stahli, H.

1955. Determination of the iodine number. Mitteilungen aus dem Gebeit der Lebensmitteluntersuchung und Hygiene, 46: 121-162. Chemical Abstracts, 49: 14345b (1955).

A comparison of the Hanus, Kaufmann, and Wijs methods. A modification of the Wijs solution is presented.

Stern, I. and B. Shapiro

1953. A rapid and simple method for the determination of esterified fatty acids and for total fatty acids in blood. Journal of Clinical Pathology, 6: 158-160.

A method is described for determination of fatty acids by hydroxamic acid formation and colorimetric measurement of the color produced with $FeCl_3$. Standard deviation $\pm$ 4%.

Stewart, C. P. and E. B. Hendry

1935. The phospholipins of blood. Biochemical Journal, 29: 1683-1689.

The Fiske-Subbarow method for determination of phosphorus is discussed. Conditions for accurate use of the F-S method are described.

Stoffel, W., F. Chu, and E. H. Ahrens, Jr.

1959. Analysis of long-chain fatty acids by gas-liquid chromatography. Micromethod for preparation of methyl esters. Analytical Chemistry, 31: 307-308.

A method is described for preparation of methyl esters of long-chain fatty acids obtained from biological material. 1 to 10 mg. of the lipid material is dissolved in a methanol-5% HCl-benzene mixture and interesterified by refluxing at 80-100° for 2 hours. The methyl esters are isolated from the reaction mixture by microsublimation and are then ready for separation by gas-liquid chromatography. Recovery of esters is over 95% of theory.

Sturges, S. and A. Knudson

1938. Application of the Schoenheimer-Sperry method to the determination of cholesterol and cholesterol esters in tissues. Journal of Biological Chemistry, 126: 543-550.

A microcolorimetric method for determination of tissue cholesterol which is based on the Schoenheimer-Sperry method (Journal of Biological Chemistry, 106: 745, 1934) is described.

Surrey, B. D.

1954. Modified iodometric determination

of organic peroxides. Analyst, 79: 86-90.

The method is similar to that of Lea (Journal of the Society of Chemical Industry, 64: 106, 1945). The reagents are all mixed in a boiling solution of AcOH and $CHCl_3$, which makes deaeration of the reagents and use of an inert atmosphere unnecessary.

Svennerholm, L.

1954. Partition chromatography of brain gangliosides on cellulose. Acta chemica scandinavia, 8: 1108.

★

Gangliosides were separated from the lipid extract of gray matter of human brain by chromatography on cellulose.

Swahn, B.

1952. A method for the localization and determination of serum lipids after electrophoretic separation on paper. Scandinavian Journal of Clinical and Laboratory Investigation, 4: 98-103.

★ ●

Lipid components of serum are separated by paper electrophoresis and treated with a 50% alcoholic solution of Sudan Black. The color absorbed by the lipids may be measured by direct colorimetry on the paper, or by elution of the spots and reading in a colorimeter.

Swahn, B.

1953. Studies on blood lipids: I. A micromethod for determination of total lipids in serum. II. A micromethod for determination of serum lipids after electrophoretical separation on filter paper. III. Electrophoretical mobility of "chylomicrons". Scandinavian Journal of Clinical and Laboratory Investigation, 5 (Suppl. 9): 114 pp.

●

A method for determination of total lipids in 0.02 ml. of serum is described. The serum is placed on a filter paper and dried, and the paper is placed in an ethanolic solution of Sudan Black B. After washing and drying, the dissolved dye is extracted and its concentration is measured colorimetrically. Effects of variables are discussed. Error is 1.89%.

See also: Bernes and McDonald (Archives of Biochemistry and Biophysics, 70: 49, 1957), who separated Sudan Black into 10 components by chromatography. All fractions were found to stain lipids, but to varying degrees.

Swern, D., H. B. Knight, O. D. Shreve, and M. R. Heether

1950. Comparison of infrared spectrophotometric and lead salt alcohol methods for determination of trans-octadecenoic acids and esters. Journal of the American Oil Chemists Society, 27: 17-21. Chemical Abstracts, 44:2260f (1950).

●

The infrared method was found to be more accurate, specific, and rapid than the lead salt method.

Szent-Gyorgi, A.

1957. Detection of chromatographic spots on paper. Science, 126: 751.

The developed chromatogram is cooled in liquid nitrogen and viewed under ultra violet light. Spots may be detected by their phosphorescence.

Taurog, A., C. Entenman, B. A. Fries, and I. L. Chaikoff

1944. An adsorption procedure for the separation of choline-containing from non-choline-containing phospholipids of liver. Journal of Biological Chemistry, 155: 19-25.

★ ▲

Total phospholipids are adsorbed on MgO and the choline-containing phospholipids are eluted with MeOH.

Taylor, W. E. and J. M. McKibbin

1953. The determination of lipide inositol in animal tissues. Journal of Biological Chemistry, 201: 609-613.

A method is described for determination of inositol in tissues by microbiological assay with Saccharomyces carlsbergensis on the acid hydrolysates of purified lipid extracts and turbidimetric measurement of the yeast growth.

It was inconvenient to use Saccharomyces cerviseae as the test organism as its medium turns very dark after autoclaving, making direct turbidimetric readings unsuitable.

Teeri, A. E.

1944. On the determination of esterified cholesterol. Journal of Biological Chemistry, 156: 279-281.

■

Esterified cholesterol was found to produce approximately 25% more color with Liebermann-Burchard reagents than free cholesterol.

Teorell, T.

1931. Spectrophotometric determination of phosphorus. Biochemische Zeitschrift, 230: 1-9. Chemical Abstracts, 25: 1756 (1931).

▲

A modification of the Fiske-Subbarow method for use in the determination of 0.01 to 0.05 mg. P by spectrophotometry. Accuracy of the method is ±2%.

Thaler, H. and E. Just

1944. Determination of the phosphatide content of fats. Fette und Seifen, 51: 55-59. Chemical Abstracts, 42:1069d (1948).

▲

A method for determination of phosphatides by burning off the organic matter in the presence of MgO and precipitation of P as ammonium phosphomolybdate is described.

Thaler, H.

1952. Determination of phosphorus in fats of low phosphatide content. Fette und Seifen, 54: 763-765. Chemical Abstracts, 47:6675b (1953).

▲

Details are given of a method for determination of phosphorus in fats containing little phosphatide by an adaptation of the method of Beveridge and Johnson (Canadian Journal of Research, 27E: 159, 1949).

Thannhauser, S. J. and P. Setz

1936. Studies on animal lipids. XI. The reineckate of the polydiamino phosphatide from spleen. Journal of Biological Chemistry, 116: 527-531.

▲

Diaminophosphatide is prepared by continuous extraction with MeOH-$CHCl_3$ (1:1) from freeze-dried tissue. Monoaminophosphatide was separated from diaminophosphatide by precipitation of the diamino as the reineckate.

Thannhauser, S. J., J. Benotti, and H. Reinstein

1939. Studies on animal lipids. XIV. The determination of lecithin, cephalin, and sphingomyelin in body fluids and tissues; with analyses of normal human sera. Journal of Biological Chemistry, 129: 709-716.

▲

Sphingomyelin is determined as the reineckate according to Thannhauser and Setz (Journal of Biological Chemistry, 116: 533, 1936) except that the precipitate is washed with acetone to remove reineckate other than sphingomyelin, and the sphingomyelin calculation is based on phosphorus content of the reineckate rather than total weight of the precipitate.

Choline is liberated by hydrolysis with gaseous HCl in methanol and determined as the reineckate.

The value of lecithin is calculated as the difference in sphingomyelin and choline values, and cephalin is calculated as the difference in total and choline-containing phospholipids.

Thannhauser, S. J., J. Benotti, and N. F. Boncoddo

1946. The preparation of pure sphingomyelin from beef lung and the identification of its component fatty acids. Journal of Biological Chemistry, 166: 677-681.

★ ● ▲

Pure sphingomyelin was prepared by alkaline saponification or by exhaustive extraction with 97% acetone in water of crude sphingomyelin extracted from lung. The fatty acids were identified by vacuum distillation and recrystallization, followed by direct titration of the free acids.

Thannhauser, S. J., N. F. Boncoddo, and G. Schmidt

1951. Studies of acetal phospholipides of brain. I. Procedure of isolation of crystallized acetal phospholipide from brain. Journal of Biological Chemistry, 188: 417-421.

★ ▲

A method is described for the isolation and purification of acetal phospholipid from beef brain by a series of solvent extractions and saponification.

Theile, O. W.

1958. A method for the determination of total lipides in organs. Zeitschrift für physiologische Chemie, 311: 136-139. Chemical Abstracts, 52:15633i (1958)

The tissue is lypholized, and the lipid is extracted with $CHCl_3$-MeOH (1:3) under reflux. The extract is evaporated, the residue is dissolved in $CHCl_3$-MeOH (2:1), overlaid with H_2O, and left standing overnight. The water is removed, and the lipid material is dried and weighed.

Thompson, A. R.

1950. A colorimetric method for the determination of esters. Australian Journal of Scientific Research, (A) 3: 128-135. Chemical Abstracts, 45:499f (1951).

An extension of the colorimetric method of Hill (Industrial and Engineering Chemistry, Analytical Edition, 18: 317, 1946; Analytical Chemistry, 19: 932, 1947) for determination of esters as their hydroxamic acid derivatives. The method is for use in determination of volatile esters and includes fixed reaction times and lower reaction temperature.

Thompson, A. R.

1951. Separation of saturated monohydroxamic acids by partition chromatography on paper. Australian Journal of Scientific Research, (B) 4: 180-186. Chemical Abstracts, 46:427f (1952).

The lower hydroxamic acids were separated by paper chromatography using various solvent systems, and located on the chromatograms by spraying with $FeCl_3$ solution.

Thornton, M. H. and F. K. Broome

1942. Determination of choline. A photometric modification of Beattie's method. Analytical Chemistry, 14: 39-41.

Choline reineckate in acetone solution is measured colorimetrically. For use with 0.6 to 16.0 mg. of choline. Maximum error is 2%.

Toms, H.

1928. Oil bromide films and their use in determining the halogen absorption of oils. Analyst, 53: 69-97.

The unsaturation of an oil is determined by absorption of bromine vapor and gravimetric determination of the change in weight of the sample caused by the bromine absorbed. Results by this method are in accord with Wijs determination.

Tomsett, S. L. and W. S. Tennant

1956. A method for determining esterified fatty acid with zone electrophoresis of serum proteins. American Journal of Clinical Pathology, 26: 1226-1230.

After electrophoresis of serum, the fatty acids are extracted from the paper and determined spectrophotometrically as their hydroxamic acid derivatives.

Tomsett, S. L.

1958. The determination of formaldehyde and acetaldehyde liberated in the periodate and ninhydrin reactions. Analytica Chimica Acta, 19: 360-363. (In English). Chemical Abstracts, 54:2092i (1960).

Formaldehyde and acetaldehyde are separated by aeration and are determined colorimetrically by means of chromotropic acid and p-hydroxybiphenyl, respectively. The method is suitable for determination of acetaldehyde and formaldehyde liberated by reaction of sugars, serine and threonine with periodate, and glycine and alanine with ninhydrin.

Tourtellotte, W. W., A. J. Vander, B. A. Skrentny, and R. N. DeJong

1958. A study of lipids in the cerebrospinal fluid. II. The determination of total lipids. Journal of Laboratory and Clinical Medicine, 52: 481-490.

A method is described for the determination of 2 μg. of total lipids by oxidation with dichromate-sulfuric acid reagent and colorimetric measurement of the reduced chromium ion.

Tourtellotte, W. W., F. M. Parker, and R. N. DeJong

1958. A study of lipids in the cerebrospinal fluid. III. The determination of total phospholipids. Journal of Laboratory and Clinical Medicine, 52: 491-495.

▲

The method is based on co-precipitation of proteins and lipids, determination of phosphorus on an aliquot using $HClO_4$-H_2SO_4 for ashing, and colorimetric measurement of molybdate color developed with ascorbic acid as reducing agent. The method is capable of determining 0.7 mμmoles (0.02μg.) of phospholipid with coefficient of variation of about 5%.

Tourtellotte, W. W., B. A. Skrentny, and R. N. DeJong

1959. Lipides in the cerebrospinal fluid. IV. The determination of free and total cholesterol. Journal of Laboratory and Clinical Medicine, 54: 197-206.

■

A method is described for determination of cholesterol in 0.25 ml. of cerebrospinal fluid. The proteins and lipids are co-precipitated with trichloracetic acid and the lipids are extracted from the precipitate. Total cholesterol is determined fluorometrically by the procedure of Albers and Lowry (Analytical Chemistry, 27: 1829, 1955) and free cholesterol by a modification of the Sperry and Webb method (Journal of Biological Chemistry, 187: 97, 1950). The method is capable of determining 1μg. of free cholesterol and 0.5μg. of total cholesterol with variations of 3 and 7%, respectively.

Trappe, W.

1938. Modification of the Kaufmann method for determining the iodine number for very small amounts of fat. Biochemische Zeitschrift, 296: 180-185. Chemical Abstracts, 33: 420^6 (1939).

The fat (0.2 to 2.0 mg.) is dissolved in $CHCl_3$, brominated with Br_2 in MeOH, and titrated iodometrically.

Trappe, W.

1940. Separation of biological fats from natural mixtures by means of adsorption columns. I. The eluotropic series of solvents. Biochemische Zeitschrift, 305: 150-161. Chemical Abstracts, 35: 477^8 (1941).

★

The "eluotropic series" (solvents arranged according to their eluting power) is given.

Trappe, W.

1940. Separation of biological lipides from their natural mixtures by adsorption columns. II. Isolation of phosphorus- and nitrogen-free lipide fractions. Biochemische Zeitschrift, 306: 316-336. Chemical Abstracts, 35: 2169^7 (1941).

★

The relative adsorbabilities of lipid fractions from biological materials are listed.

Trappe, W.

1941. Separation of biological lipides from their natural mixtures by means of adsorption columns. III. Separation of phosphorus- and nitrogen-free lipide fractions. Biochemische Zeitschrift, 307: 97-106. Chemical Abstracts, 35: 4053^3 (1941).

★

Free fatty acids with phosphatides were separated from total lipid extract on an Al_2O_3 column. An activated Frankonite KL column was used to separate hydrocarbons and cholesterol esters.

Trappe, W.

1942. Determination of free and esterified cholesterol in 0.1 cc. blood from the finger tip. Klinische Wochenschrift, 21: 651-652. Chemical Abstracts, 38: 2058^5 (1944).

■

Blood lipids are adsorbed on SiO_2. Esterified cholesterol is eluted with benzene, free cholesterol is eluted with Et_2O-$CHCl_3$, the solvents are evaporated, and cholesterol is estimated in each fraction with AcOH-$ZnCl_2$ and AcCl.

Trappe, W.

1943. Simple method for the separate quantitative estimation of free and esterified cholesterol in blood serum without digitonin precipitation and saponification. Zeitschrift für physiologische Chemie, 273: 177-190. Chemical Ab-

■

stracts, 37: 3119[9] (1943).

The cholesterol is extracted from plasma with EtOH-petroleum ether (2:3), and the extract is dried and evaporated. The residue is dissolved in CCl_4 and chromatographed on an Al_2O_3 column. Esterified cholesterol is eluted with CCl_4 and free cholesterol with $CHCl_3$, and cholesterol content of each fraction is estimated colorimetrically using $ZnCl_2$-AcOH and AcCl.

Trinder, P.

1952. Determination of cholesterol in serum. Analyst, 77: 321-325.

■

Cholesterol esters are hydrolyzed by heating the serum with alcoholic KOH, and the cholesterol is extracted with petroleum ether. Color is developed with acetyl chloride-sulfuric acid.

Trusov, V. I.

1950. Determination of choline in biological materials. Biokhimiya, 15: 495-498. Chemical Abstracts, 45:3448f (1951).

Choline is precipitated as the reineckate, the Cr is oxidized by $KBrO_3$ or H_2O_2, and the resulting chromate is titrated with $Na_2S_2O_3$.

Turner, M. E.

1931. A simplification of the Okey method for the determination of cholesterol by the oxidation of the digitonide. Journal of Biological Chemistry, 92: 495-498.

■

A modification of the Okey method (Journal of Biological Chemistry, 88: 367, 1930) for oxidation of cholesterol digitonide.

Twitchell, E.

1921. The precipitation of solid fatty acids with lead acetate in alcoholic solution. Journal of Industrial and Engineering Chemistry, 13: 806-807.

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●

A method is described for separation of the liquid and solid fatty acids in which the solid acids are precipitated from a hot alcoholic solution by addition of lead acetate and cooling. The liquid acids remain in solution and are filtered off.

Uzman, L. L.

1953. A general method for the preparation of cerebrosides. Archives of Biochemistry and Biophysics, 45: 149-155.

Tissue is homogenized and extracted with $CHCl_3$-MeOH (2:1) and trichloracetic acid is added. The mixture is centrifuged, the upper and lower layers of the three-phase system are drawn off, and the interphase layer is dialyzed against distilled water. The resulting material is recrystallized from alcohol-chloroform (1:1). 65-75% of the total tissue cerebrosides are obtained from spleen and brain.

Van Beers, G. J., H. De Iongh, and J. Boldingh

1957. Isolation of phospholipides by dialysis through a rubber membrane. Essential Fatty Acids. Proceedings of the International Conference on Biochemical Problems of Lipids, 4th Oxford, (Pub. 1958) pp. 43-47. Chemical Abstracts, 53:17278a (1959).

★
▲

A method is described for separation of phospholipids from lipid mixtures by dialysis through a rubber membrane. The phospholipids do not pass through when a nonpolar solvent is used.

Van de Kamer, J. H., N. A. Pikaar, A. Bolsaens-Frankena, C. Couvee-Ploeg, and L. van Ginkel

1955. Quantitative determination of the different higher saturated fatty acids in fat from blood, chyle, and faeces, by means of partition chromatography on rubber. Biochemical Journal, 61: 180-186.

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●

The fats are extracted and saponified, and the fatty acids are extracted from the hydrolysate. The unsaturated fatty acids are oxidized with $KMnO_4$, and the saturated fatty acids are separated on a rubber chromatographic column with acetone-water solvent mixtures.

Vandenheuvel, F. A. and D. R. Vatcher

1956. Partition chromatography of aliphatic acids. Quantitative resolution on normal chain even acids from C_{12} to C_{24}. Analytical Chemistry, 28: 838-845.

★
●

C_{12}-C_{24} fatty acids were chromatographed on a dichlorodimethylsilane-treated column of silicic acid by using 2,2,4-trimethylpentane as stationary phase and eluting with aqueous methanol. The eluted acids are measured by titration. Error is less than 2.5%.

An apparatus for automatically and continuously changing the composition of the eluting solvent and a semiautomatic, motor driven microburette are described.

Van Handel, E.

1959. Separation and chemical assay of lipide classes. Journal of the American Oil Chemists Society, 36: 294-297. Chemical Abstracts, 53:17226h (1959).

A review and discussion of methods for microdetermination of cholesterol, triglycerides, and phospholipids.

Van Handel, E. and D. B. Zilversmit

1953. Micromethod for the direct determination of serum triglycerides. Journal of Laboratory and Clinical Medicine, 50: 152-157.

Triglycerides are determined as glycerol by the chromotropic acid method of Lambert and Neish (Canadian Journal of Research, 28: 83, 1950) after quantitative removal of phosphatides and saponification.

Wachsmuth, H. and L. van Koeckhoven

1959. The determination of choline in blood serum. Clinica Chimica Acta, 4: 206-212 (In French). Chemical Abstracts, 53:12381g (1959).

Precipitation of choline by silico-tungstic acid and reduction of the silico-tungstate with HSO_3^- gives a color reaction which is 600 times more intense than the choline reineckate reaction.

Weigensberg, B. I. and G. McMillan

1959. Ultraviolet spectrophotometric method for the determination of cholesterol. American Journal of Clinical Pathology, 31: 16-25.

Serum or tissue cholesterol or the cholesterol digitonide is determined by spectrophotometric measurement of the absorption of UV light at 204-206 mμ. The method is simple and reproducible, and is 3 to 4 times more sensitive than the Schoenheimer-Sperry method.

Weil, L. and M. A. Russell

1942. Studies on plasma phosphatase activity and on blood phospholipids in rats with obstructive jaundice. Journal of Biological Chemistry, 114: 307-314.

A method is described for the determination of the phospholipids in an alcohol-ether extract of 8 mm^3 of plasma or 4 mm^3 of blood by a micro modification of the method of Boyd (Journal of Biological Chemistry, 144: 223, 1936). The phospholipids are digested with $HClO_4$ and color is developed with ammonium molybdate, using aminonaphtholsulfonic acid as reducing agent.

Weiss, B.

1957. The separation of sphingolipides by adsorption chromatography. Journal of Biological Chemistry, 223: 523-534.

A method is described for chromatographic separation of sphingolipides of the central nervous system by gradient elution with chloroform and methanol from a silicic acid column.

Attempts to use the method of Taurog, et al, (Journal of Biological Chemistry, 155: 19, 1944) for separation of choline-containing from non-choline-containing phospholipids on magnesium oxide were unsuccessful.

Diminished resolving power was observed when Hyflo Super-Cel was added to the adsorbent, or when the silicic acid was added to the column as a chloroform suspension rather than a chloroform-methanol suspension.

Wheeldon, L. W. and F. D. Collins

1957. Studies on phospholipids. 1. The determination of amino nitrogen in unhydrolyzed phospholipids. Biochemical Journal, 66: 435-441.

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1-flouro-2:4-dinitrobenzene is reacted directly with the NH_2 groups of the unhydrolyzed phospholipid in the presence of triethylamine, and the colored compounds formed are estimated colorimetrically.

Wheeldon, L. W. and F. D. Collins

1958. Studies on phospholipids. 3. Determination of choline. Biochemical Journal, 70: 43-45.

▲

The lipid sample is hydrolyzed at 100° in ethanol-HCl. Choline is precipitated as choline phosphomolybdate, washed with isobutanol, and dissolved in acetone. H_2SO_4 and $SnCl_2$ are added and the optical density of the blue solution is measured at 630 mμ. The method agrees closely with the reineckate method of Glick. Added choline was quantitatively recovered.

White, L. K.

1946. Spectrophotometric determination of glycerol. Oil and Soap, 23: 323-326. Chemical Abstracts, 41:56d(1947).

A procedure for determination of glycerol by spectrophotometric measurement of the blue color of the copper-glycerol complex is described.

White, M. F. and J. B. Brown

1949. A study of the tetrabromide method of estimating linoleic acid in fatty acid mixtures. Journal of the American Oil Chemists Society, 26: 385-388.

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The yield of insoluble tetrabromides from bromination of linoleic acid is affected by the composition of the petroleum ether used as solvent, and by the amounts of linoleic acid and other fatty acids in the sample used.

Whitehorn, J. C.

1924-1925. A method for the determination of lipoid phosphorus in blood and plasma. Journal of Biological Chemistry, 62: 133-138.

▲

The extracted lipids are digested with H_2SO_4-HNO_3. Molybdate color is developed with sodium sulfite and hydroquinone.

Whittaker, V. P. and S. Wijesundera

1952. The separation of esters of choline by filter-paper chromatography. Biochemical Journal, 51: 348-351.

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Choline esters were separated by paper chromatography using n-propanol- or n-butanol-water as solvents. The esters were detected on the chromatogram by using hydroxylamine-ferric chloride, iodine, or phosphotungstic acid-stannous chloride.

Wiese, H. F. and A. E. Hansen

1953. Semimicromethod for unsaturated fatty acids of blood serum. Journal of Biological Chemistry, 202: 417-423.

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The unsaturated fatty acids from 3 ml. of blood serum are isomerized by treatment with 1% KOH in ethylene glycol at 180° for 25 minutes, and read spectrophotometrically.

Williams, H. H., B. N. Erickson, I. Avrin, S. S. Bernstein, and I. G. Macy

1938. Determination of cephalin in phospholipids by the estimation of choline. Journal of Biological Chemistry, 123: 111-118.

▲

The total phospholipid is determined on an aliquot of tissue extract by Bloor's oxidative titrimetric method (Journal of Biological Chemistry, 82: 273, 1929). Choline is determined as the reineckate on a second aliquot after $Ba(OH)_2$ hydrolysis, and cephalin is calculated as the difference in total and choline-containing phospholipid.

Willis, G. C., Jr.

1950. Preparation of fatty acid methyl esters. Chemist-Analyst, 39: 62.

●

Methyl esters of the fatty acids may be prepared by treating a methanolic solution of the acid with concentrated H_2SO_4 and heating to remove excess MeOH. The reaction is essentially complete.

Winzler, R. J. and E. R. Meserve

1945. Spectrometric determination of small amounts of choline. Journal of Biological Chemistry, 159: 395-397.

Choline reineckate in acetone is

determined spectrophotometrically at 327 mμ. Accuracy is ± 5% with 50-400 μg. of choline hydrochloride.

Wittenberg, J. B.

1955. The separation of sphingosine and related compounds by reversed phase partition chromatography. Journal of Biological Chemistry, 216: 379-390.

★

After conversion to their N-succinyl derivatives with succinic anhydride, sphingosine and related compounds are separated on a dichlorodimethylsilane-treated column of diatomaceous earth.

A method for assay of sphingosine compounds is proposed which entails conversion of sphingosine and related compounds to their N-succinyl derivatives and titration with alkali of the carboxylic acid group which has been introduced. Three to 100 μmoles were determined with an accuracy of ±5%.

Wittenberg, J. B., S. R. Korey, and F. H. Svenson

1956. The determination of higher fatty aldehydes in tissues. Journal of Biological Chemistry, 219: 39-47.

A method is described for the estimation of higher fatty aldehydes in tissue lipids by colorimetric measurement of their p-nitrophenylhydrazone derivatives. The method is specific for higher aldehydes, and lower aldehydes and keto acids do not interfere. Determines 0.1 to 7 μmoles within 5%.

Wittenberg, J. B.

1957. The separation of the C_6-C_{12} fatty acids by reversed-phase partition chromatography. Biochemical Journal, 65: 42-45.

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Chloroform-Skellysolve S-water-methanol solvent systems were used to separate the C_6-C_{12} fatty acids on a dichlorodimethylsilane-treated column of Hyflo Super-Cel.

Witter, R. F., G. V. Marinetti, A. Morrison, and L. Heicklin

1957. Paper chromatography of phospholipides with solvent mixtures of ketones and acetic acid. Archives of Biochemistry and Biophysics, 68: 15-20.

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Lysolecithin, sphingomyelin, phosphatidyl ethanolamine, lecithin, and phosphatidic acid in quantities of 10 to 25 μg. in 20 μl. were separated by paper chromatography using mixtures of various ketones and acetic acid as solvent systems.

Wolman, M.

1950. Staining of lipids by the periodic-acid-Schiff reaction. Proceedings of the Society for Experimental Biology and Medicine, 75: 583-585.

Schiff's reagent will stain unsaturated lipids after they have been oxidized with periodate. Sphingolipids will stain even if they do not contain a carbohydrate.

Woolley, D. W.

1941. A method for the estimation of inositol. Journal of Biological Chemistry, 140: 453-459.

A basal medium was developed which supported practically no growth under the experimental conditions. Inositol content of the sample material added to the basal medium was determined by colorimetric measurement of the turbidity produced by the yeast Saccaromyces cereviseae.

It was noted that inositol does not produce marked stimulation unless the basal medium was otherwise complete for optimal growth.

Attempts to use Eastcott's basal medium (Journal of Physical Chemistry, 32: 1096, 1928) were not entirely successful.

Woolley, D. W.

1943. Isolation and partial determination of structure of soy bean lipositol, a new inositol-containing phospholipid. Journal of Biological Chemistry, 147: 581-591.

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▲

Lipositol was prepared by extraction with $CHCl_3$ and recrystallization from $CHCl_3$ with methanol and ethanol.

Wren, J. J. and H. K. Mitchell

1958. Silicic acid chromatography of lipids of whole human blood. Proceedings of the Society for Experimental Biology and Medicine, 99: 431-435.

★
●

Blood lipids were separated into 20 components by chromatography on silicic acid.

Wycoff, H. D. and J. Parsons
1957. Chromatographic microassay of cholesterol and cholesterol esters. Science, 125: 347-348.

Free and esterified cholesterol are separated from about 0.02 ml. of plasma by chromatography on SiO_2, and estimated by measurement of the color produced with $FeCl_3$ reagent.

Yasuda, M.
1931. Contributions to the micro determination of cholesterol. Journal of Biological Chemistry, 92: 303-312.

Modifications of Okey's procedure (Journal of Biological Chemistry, 88: 367, 1930). Acetone is used to separate the digitonide from excess digitonin and lipid material. The digitonide is freed of impurities by solution in hot absolute alcohol and filtration.

Yasuda, M.
1931-2. The determination of the iodine number of lipids. Journal of Biological Chemistry, 94: 401-409.

The lipids are halogenated with pyridine sulfate dibromide, and the excess halogen is titrated with thiosulfate. The method is combined with determination of total lipid by Bloor's oxidative procedure. Chloroform dissolves the phospholipid precipitated from acetone-$MgCl_2$ as well as moist ether, and dissolves a smaller amount of the $MgCl_2$.

Young, L.
1934. The determination of inositol in animal tissues. Biochemical Journal, 28: 1435-1443.

Inositol is extracted from tissue with aqueous acetone, purified by precipitation, and estimated by oxidation with potassium iodomercurate.

Youngs, C. G. and B. M. Craig
1951. A note on the preparation of methyl esters of fatty acids. Journal of the American Oil Chemists Society, 28: 521-522. Chemical Abstracts, 46: 1271f (1952).

A method is described for direct conversion of saponified fatty acids to their methyl esters with dimethyl sulfate. Yield is over 99%.

Youngs, C. G., A. Epp, B. M. Craig, and H. R. Sallans
1957. Preparation of long-chain fatty acid chlorides. Journal of the American Oil Chemists Society, 34: 107-108. Chemical Abstracts, 51:7039d (1957).

A method is described for preparation of fatty acid chlorides by treating the free acid with PCl_5 or PCl_3 in an inert organic solvent. Excess chlorinating agent is removed by washing the solvent phase with water. Yield is quantitative, with less than 1.5% of free acid unreacted.

Zak, B., R. C. Dickenham, E. G. White, H. Burnett, and P. J. Cherney
1954. Rapid estimation of free and total cholesterol. American Journal of Clinical Pathology, 24: 1307-1315.

Cholesterol is extracted from serum with 50:50 alcohol-acetone. Total cholesterol is determined directly on the dried residue of an aliquot of the extract, and free cholesterol is precipitated with digitonin in another aliquot. Color is developed in both portions with the $FeCl_3$-acetic acid reagent of Zlatkis, et al (Journal of Laboratory and Clinical Medicine, 41: 486, 1953).

Zak, B., N. Moss, A. J. Boyle, and A. Zlatkis
1954. Reactions of certain unsaturated sterols with acid iron reagent. Analytical Chemistry, 26: 776-777.

Colors produced by reaction of various sterols with ferric chloride-acetic acid-sulfuric acid reagent are given.

Zak, B.
1957. Simple rapid microtechnic for serum total cholesterol. American Journal of Clinical Pathology, 27: 583-588.

Serum proteins are precipitated with a ferric chloride-acetic acid solution. A portion of the supernatant is diluted with H_2SO_4 and the resulting color is measured in a spectrophotometer at 560 mμ.

Zbinovsky, V.
1955. New solvent for separating monocarboxylic acids (C_2 to C_{16}) and dicarboxylic acids (C_2 to C_{22}). Analytical

Chemistry, 27: 764-768.

All of the individual saturated fatty acids C_2-C_{14}, the C_{13}-C_{16} fatty acids differing by two C in chain length, and the dicarboxylic acids C_2 to C_{22} were separated on a silicic acid column. Methyl cellosolve-water was used as a stationary phase, and Skellysolve B, n-butyl ether, or their mixtures were used as eluting solvents.

Zilch, K. T. and H. J. Dutton

1951. Analysis of fat oxidation product
★ by countercurrent distribution methods. Analytical Chemistry, 23: 775-778.

Effects of various functional groups on the distribution of compounds were studied. Partition coefficients are given for a number of model compounds.

Zilversmit, D. B. and A. K. Davis

1950. Micro-determination of plasma phospholipids by trichloracetic acid
▲ precipitation. Journal of Laboratory and Clinical Medicine, 35: 155-160.

A method is described for determination of phospholipids in 0.2 ml. of plasma. The phospholipids are precipitated with trichloracetic acid and phosphorus is determined colorimetrically after perchloric acid digestion of the precipitate.

Zlatkis, A., B. Zak, and J. Boyle

1953. A new method for the direct determination of serum cholesterol. Journ-
■ nal of Laboratory and Clinical Medicine, 41: 486-492.

A sulfuric acid, acetic acid, and ferric chloride reagent is added directly to a 0.1 ml. sample of serum and the color is read spectrophotometrically at 400-700 mμ. Cholesterol and cholesterol esters yield the same molar color, thus eliminating the need for saponification. The method compares well with other methods, and is rapid and reproducible.

Zuckerman, J. L. and S. Natelson

1948. A convenient and rapid procedure for total cholesterol estimation using
■ an acid chloroform extraction. Journal of Laboratory and Clinical Medicine, 33: 1322-1325.

Sulfuric acid and chloroform are added to the serum sample and mixed. The sample is centrifuged and the acid and precipitated protein are removed. Acetic anhydride and sulfuric acid are added to an aliquot of the chloroform solution and the color is read at 625 mμ. The method requires about 45 minutes. Results are comparable to those obtained by Bloor's extraction method.

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