ABSTRACTS

R. A. REINERS, Editor. ABSTRACTORS: J. G. Endres, Kazuo Fukuzumi, J. Iavicoli, K. Kitsuta, F. A. Kummerow, C. C. Litchfield, Gladys Macy, Louise R. Morrow, E. G. Perkins, and T. H. Smouse

• Fats and Oils

ESTIMATION OF PHYSICO-CHEMICAL PROPERTIES OF FATTY OIL SOLUTIONS. Kazuhito Kusano (Miyazaki Univ., Japan). *Yukagaku* 13, 185–94 (1964). A general discussion with 23 references.

SOME ASPECTS OF THE RHEOLOGY OF CONDENSED MILK AND ICE CREAM. Fumiyasu Tsuchiya (Meiji Milk Product Co., Kitatamagun, Tokyo). *Yukagaku* 13, 100–8 (1964). A review with 42 references.

THIN LAYER CHROMATOGRAPHY APPLIED TO THE ANALYSIS OF PRESERVATIVES IN FOODSTUFFS, II. M. Covello and O. Schettino (Univ. of Naples, Italy). *Riv. Ital. Sostanze Grasse* 41, 75–8 (1964). In connection with a study on the application of TLC to the analysis of preservatives in foods, a group of substances, prepared by iodizing *p*-oxybenzoic acid esters, have been examined. The study was also extended to other non-iodized esters of the same acid. The experimental technique is described and the R_f values of the various substances are reported.

GRAPHITIZED CARBON BLACK AS A SUPPORT FOR GAS LIQUID CHROMATOGRAPHY. T. F. Brodasky (Research Lab., The Upjohn Co., Kalamazoo, Mich.). *Anal. Chem.* 36, 1604–6 (1964). Overcoming the troublesome adsorption effects of polar compounds in gas liquid chromatography has involved the use of specialized supports or chemical treatment of normal supports to deactivate the surface. The latter may involve lengthy chemical procedures, leading to certain limitations which can be disadvantageous. This paper describes the use of highly graphitized carbon black as a support for gas liquid chromatography. It requires no special treatment and has no temperature or mechanical limitations. Using a nonpolar stationary phase, several support materials representing the classes including silazined supports, perfluorocarbon polymers and untreated diatomaceous earth were compared with graphitized carbon black in their ability to reduce peak asymmetry of homologous series of alcohols, ketones and amines. Graphitized carbon black effected the greatest reduction of peak asymmetry. The column efficiency in terms of HETP and resolution of three alcohols was also evaluated.

SPECTROPHOTOMETRIC STUDY OF SOME ITALIAN OLIVE OILS. R. Casillo. *Olearia* 18, 9–14 (1964). Based on the examination of over 100 samples of olive oil from Southern Italy, a correlation between [illegible] parameters and organoleptic properties of [illegible] as well as limits for the classification of various grades of oil. These limits do not differ greatly from previously reported values and confirm the usefulness of spectrophotometry in assigning grades to olive oil samples. The three grades of oil commonly recognized are: virgin extra fine (FFA $< 1.5\%$, $K_{232} < 2$, $K_{268} < 0.15$), virgin (FFA 4%, $K_{232} < 2.5$, $K_{268} < 0.25$) and blends of virgin and treated oils (FFA $< 4\%$, $K_{232} < 3$, $K_{268} < 0.60$).

TALL OIL. I—II. K. Berger and W. Müller. *Plaste u. Kautschuk* 10, No. 9, 566–8; No. 10, 631–5 (1963). Properties, uses and paper chromatographic analysis of tall oil are considered. Fairly good separation of the component acids is obtained by reversed-phase chromatography, using acetic acid or acetic acid/acetonitrile developing solvents. Clearest separation of the resin and fatty acids is obtained by preliminary hydrogenation of the mixed acids *in situ* on the paper. (Rev. Current Lit. Paint Allied Ind.)

GAS-CHROMATOGRAPHIC STUDY OF TALL-OIL FATTY ACIDS FRACTIONATED BY COUNTER-CURRENT DISTRIBUTION. Y. Aho, O. Harva and S. Nikkalä. *Tehn. Kem. Aikl.* 19, 390–2 (1962). Countercurrent distribution of a tall-oil fatty acid distillate (between heptane and a 1:1:1 mixture of methanol, acetic acid and formamide) was followed by gas chromatography of the methyl esters of the acids obtained. Seven known acids (including 41% of oleic acid and 38% of linoleic acid) and small amounts of 14 unknown acids of 17–20 C atoms were separated; the latter fraction contained 10% (on total acids) of a trienoic acid shown to be probably *cis*-5,9,12-octadecatrienoic acid (linolenic acid isomer). This gave only diene conjugation after alkali isomerisation. The presence of this acid explains the erroneous results obtained by UV spectrophotometric analysis of tall-oil fatty acids. (Rev. Current Lit. Paint Allied Ind.)

SOME CRYSTAL STRUCTURES OF LONG-CHAIN GLYCERIDES. K. Larsson. *6th Internat. Congress and Symposium, Rome*, 1963; *Acta Cryst.* (suppl.) 16, Pt. 13, A 57 (1963). A terminating methyl group in hydrocarbon chains can often be replaced isotypically by a bromine atom. The crystal structures of the stable forms of simple triglycerides and 2-monoglycerides have been determined using this technique. The racemic 1-mono-glycerides of Br fatty acids have also been studied although they are not isotypic with the unsubstituted compounds much information on the crystal structures of the latter has been obtained. (Rev. Current Lit. Paint Allied Ind.)

THE USE OF SARAN WRAP TO PROTECT CHROMATOPLATES DURING THEIR EXPOSURE TO IODINE VAPOR. T. Negishi, M. E. McKilligan and M. Lepage (Food Res. Inst., Canada Dept. of Ag., Ottawa, Canada). *J. Lipid Res.* 5, 486 (1964). Saran wrap (a commercially available cellophane sheet) is used to prevent loss of polyunsaturated lipids when using iodine vapor for the detection of lipids on thin-layer chromatographic plates.

CHLOROPHYLL CATALYSIS OF FAT PEROXIDATION. J. L. Hall and D. L. Mackintosh (Depts. of Biochemistry and Animal Husbandry, Kansas State Univ., Manhattan, Kans.). *Food Science* 29, 420–1 (1964). Peroxide apparently occurring in freshly prepared sausage was traced to the catalytic effect of chlorophyll in the sage after fat extraction. The extracts were exposed to ordinary laboratory illumination a day before analysis. No peroxide appeared in extracts of the pork fat alone, nor in extracts of the sage alone. But mixtures of fat extract with sage extract developed peroxide. Extracts of green leafy material from other species and purified chlorophyll likewise developed peroxide with fat extracts. This effect is ascribed to the porphyrin structure. Extracts containing fat and chlorophyll should be kept in the dark until peroxide estimation.

SEPARATION OF POLYMERIC PRODUCTS OF AUTOXIDIZED METHYL DOCOSAHEXAENOATE BY PARTITION CHROMATOGRAPHY. Kazuo Fukuzumi and Takero Miyakawa (Nagoya University). *Kogyo Kagaku Zasshi* 66, 1320–4 (1963). Nearly pure methyl docosahexaenoate was autoxidized by aeration at 35C and liquid-partition chromatographic separation of polymerized products from the autoxidized methyl ester gave three well-isolated fractions composed of monomer, dimer and trimer (isolated for the first time). Silica gel and methanol benzene were used in the chromatographic separation. These fractions were characterized by estimating peroxide values, and determining ultraviolet and infrared spectra. All these oxidized fractions contain both —OOH group and conjugated diene. The following mechanism was proposed. By polymerization, α-methylene groups and double bonds decrease, *cis-trans* conjugated dienes almost disappear, and rings containing oxygen are newly produced.

ISOMERIZATION DURING HYDROGENATION OF METHYL DOCOSAHEXAENOATE. Kazuo Fukuzumi and Tomihiro Yatsuo (Nagoya University). *Kogyo Kagoku Zasshi* 66, 1324–8 (1963). Methyl docosahexaenoate of 95% purity (obtained from mackerel pike oil) was hydrogenated at 30 ± 0.1C by using palladium black as a catalyst and ethyl acetate as a solvent. Ultraviolet and infrared spectra, gas-liquid chromatogram and hydrogen value were determined on each sample. As hydrogenation proceeds initial conjugated triene concentration decreases, and conjugated dienes increase to a maximum and then decrease. In case of this study, the fatty acid composition of hydrogenated samples could not be calculated by the usual method of ultraviolet spectroscopy or gas-liquid chromatography. During the course hydrogenation, the concentration of isolated *trans* bonds absorbing at 968 cm^{-1} reaches maximum (at a hydrogen value of about 180) and then decreases.

SEPARATION OF METHYL DOCOSAHEXAENOATE HYDROPEROXIDES BY PARTITION CHROMATOGRAPHY. Kuzo Fukuzumi, Yoshiaki Iwata and Moritaka Takada (Nagoya University). *Kogyo Kagoka Zasshi* 66, 1675–8 (1963). Nearly pure methyl docosahexaenoate was autoxidized at −1 to 2C, and the products were separated by partition chromatography, using silica gel and methanol-

benzene. The properties of these fractions were investigated by determining peroxide values and ultraviolet and infrared spectra. Unoxidized fraction, monohydroperoxide and a third fraction were separated. Hydroperoxide in the third fraction had changed on the chromatographic column.

DIFFERENCE BETWEEN AUTOXIDIZED CONJUGATED AND NONCONJUGATED METHYL DOCOSAHEXAENOATES. Kazuo Fukuzumi and Tadanao Wakita (Nagoya University). *Kogyo Kagaku Zasshi* 66, 1846–9 (1963). Conjugated (A) and nonconjugated methyl docosahexaenoates (B) were autoxidized in the dark at 0 to −2C for 30 days. Peroxide values, refractive indices, molecular weight and both ultraviolet and infrared spectra of the samples taken at intervals during autoxidation were determined. Peroxide values of A were always smaller than those of B. But refractive indices of A were always larger than those of B. Molecular weight of A increased remarkably after 19 days, while that of B did not change appreciably. The amount of conjugated diene of A was almost constant for about 18 days, then increased. The amount of the other polyenes decreased gradually. The amount of conjugated diene of B increased with time for about 18 days. It was ascertained that the absorption band at 3,020 cm^{-1} was due to the α-methylene group.

POLYMERS FORMED DURING THE AUTOXIDATION OF ALKALI-ISOMERIZED METHYL DOCOSAHEXAENOATE. Kazuo Fukuzumi and Kazuma Ishida (Nagoya University). *Kogyo Kagaku Zasshi* 67, 324–7 (1964). Methyl docosahexaenoate of 95% purity (obtained from cuttlefish oil) was conjugated by alkali isomerization and then autoxidized by blowing with dried air at 35C for 90 hr in the intermittent sunlight. The autoxidized methyl ester was extracted by a mixture of *n*-hexane and diethyl ether to yield 10 fractions. The ultraviolet and infrared absorption spectra for these fractions were determined. These autoxidized samples were of lower peroxide number than correspondingly oxidized unconjugated ones. Generally, polyenes higher than trienes were not found. α,β-Unsaturated carbonyl groups were found. Polymerization results in the decrease of the α-methylene groups and double bonds and an increase of *trans-trans* conjugated dienes. Monomers polymerize through the C—O bond.

HYDROGENATION OF CONJUGATED METHYL EICOSAPENTAENOATE. Kazuo Fukuzumi, Yoshiaki Iwata and Mikio Suzuki (Nagoya University). *Kogyo Kagaku Zasshi* 67, 919–21 (1964). Methyl eicosapentaenoate of 91% purity (obtained from cuttlefish oil) was conjugated with alkali and then hydrogenated at 30 ± 0.1C using palladium black as a catalyst and ethyl acetate as a solvent. Ultraviolet and infrared spectra, refractive indices and hydrogen values were determined. The hydrogen values of hydrogenated, conjugated, highly unsaturated acids are not in proportion to refractive indices unlike those of nonconjugated ones. During hydrogenation the rate of disappearance of conjugated diene is the greatest and that of the conjugated pentaene is the smallest. In the sample with a hydrogen value of about 170, conjugated double bonds are quickly replaced by the isolated *trans* double bonds.

A STUDY OF THE INFLUENCE OF THE INACTIVATION OF THE FERMENTATION SYSTEM ON THE QUALITY OF OIL IN THE TREATMENT OF LINSEED. E. A. Siminov and N. I. Gribova. *Trudy Vniiz* 23, 58 69 (1963). When linseed during the normal course of processing is heated at 95–100C for 40 45 minutes, a quantity of nonhydratable phosphatides passes into the oil. In addition there appears in the cooked cake a quantity of HCN equal to the quantity of cake that is lost. This phenomenon is due to the fermentation of complex phospholipoproteins and glucosides. The same phenomenon does not occur when linseed cake is cooked for a shorter period of time. (Rev. Franc. Corps Gras)

EXTRACTION OF SOYBEANS WITH ETHYL ALCOHOL. F. A. Visnepol'skaja. *Trudy Vniiz* 23, 131–44 (1963). Ethyl alcohol at a ratio of 4:1 and at a temperature close to its boiling point extracts the oil from soybeans. The solvent can be reused. Products obtained are an oil that does not need refining, a light colored cake free from nonnutritive substances, light colored and sweet tasting phosphatides and a new product, concentrated phosphoglucoside. (Rev. Franc. Corps Gras)

PROCESS FOR PREPARING POWDERED FOOD-STUFF CONTAINING FINELY DISPERSED FAT. J. J. Scheidegger (Koopmans Meelfabrieken N.V.). *U.S. 3,142,569.* A process is described for the preparation of a powdered food-stuff in which fat is dispersed. A mixture containing from 5 to 25% fat, to which 2 to 15% of water is added, is subjected to a grinding action so that the fat is dispersed in the form of fine droplets. The mixture is then dried in such a way that the moisture content is reduced to less than 5%.

ELIMINATION OF THE GOSSYPOL GLANDS STARTING WITH COTTONSEED CAKE WITH SIMULTANEOUS EXTRACTION OF THE OIL. V. P. Rzehin *et al. Trudy Vniiz* 23, 70–7 (1963). Elimination of gossypol glands from cottonseed cake by a method of fractionation using a mixture of liquid and gaseous carbon tetrachloride is worthy of attention. The method permits: a) the extraction of an oil which is no different from that obtained by ordinary extraction, b) the separation of relatively pure gossypol glands at a yield of 2.2% of the weight of the cake, c) the preparation of an oil cake containing a maximum of soluble protein and of free sugar, and a minimum of free and bound gossypol. The gossypol glands obtained by this separation contain after purification about 24 to 30% free gossypol, about 1.39 to 1.54% gossypurpurine, about 2.93 to 3.48% bound gossypol, about 2% nitrogen, 0.14% phosphorus, 4 to 6% moisture and volatile substances. (Rev. Franc. Corps Gras)

APPLICATION OF THIN-LAYER CHROMATOGRAPHY TO THE STUDY OF FATS AND LIPIDS. M. M. Loury (Lab. of the Inst. of Fats and Oils). *Rev. Franc. Corps Gras* 11, 259–272 (1964). A review is presented regarding the use of TLC in the separation of fatty acids, triglycerides, derivatives, identification of minor components, unsaponifiables, vitamins, carotenoids and the separation of antioxidants.

AUTOXIDATION: PART XXVIII. HYDROPEROXIDE ISOMERS IN THE AUTOXIDATION OF METHYL OLEATE. N. A. Kahn (Food Tech. East. Reg. Lab. Pakistan Council of Sci. and Ind. Res. Dacca). *Oleagineux* 19, 397 (1964). The controversy between free-radical initiation and the transition state theory was discussed. The different experimental observations have supported the theory involving activated pi-electron-complexes. The free-radical theory has been found untenable in the initial autoxidation processes from all points of view. The characterization of isolated hydroperoxides obtained from autoxidized methyl oleate by oxygen has proven beyond doubt the formation of only hydroperoxide isomers at carbons 9 and 10 as required by the pi-electron-complex theory.

PROCESS FOR REMOVING THE HALPHEN TEST RESPONSE FROM COTTONSEED OIL. E. T. Rayner, Dorothy C. Heinzelman and H. P. Dupuy. *U.S. 3,135,775.* Cottonseed oil is treated in an inert atmosphere at a temperature of 180–225C for a period of 0.5–2 hours with from ¼% to 2% by weight (on weight of oil) of a reagent selected from the group consisting of mellitic acid, trimellitic acid anhydride, pyromellitic dianhydride, and ortho-phosphoric acid. The treated oil is eluted in petroleum ether (boiling range 30–60C) through a column of alumina and the solvent is removed.

ANTI-SPATTERING DRY SHORTENING COMPOSITION. J. C. Wootton (Procter & Gamble Co.). *U.S. 3,138,463.* A dry shortening composition having improved antispattering properties when used in frying consists of a fatty triglyceride and a partial ester of a compound selected from the group consisting of straight chain pentitols and hexitols with fatty acids having a chain length of 12 to 22 carbon atoms. The ester has at least 4 unesterified and uncombined hydroxyl groups present in the molecule and is present in an amount of from 0.3% to 0.9% by weight of the composition but insufficient to cause substantial foaming.

DIRECT GAS CHROMATOGRAPHIC FRACTIONATION OF MIXED NEUTRAL LIPIDS OF NATURAL ORIGIN. A. Kuksis (Dept. of Biochem., Queen's Univ., Kingston, Ont.). *Can. J. Biochem.* 42, 419–30 (1964). Experimental conditions which the author developed for the gas chromatographic separation of natural triglyceride mixtures have been found to be satisfactory for a direct gas chromatographic fractionation of mixed neutral lipids. The type of analysis used has been shown to be readily applicable to the mixtures of free sterol, steryl ester and triglyceride found in lymph, blood plasma and certain molecular distillates of corn oil. The separations are based primarily on the carbon numbers or molecular weight of the material and are effective because of the virtual absence of short chain triglycerides from these samples. The results are essentially quantitative and can be obtained within an hour on 10 to 50 micrograms of total lipid. For these analyses a solvent-refined lipid extract is usually satisfactory.

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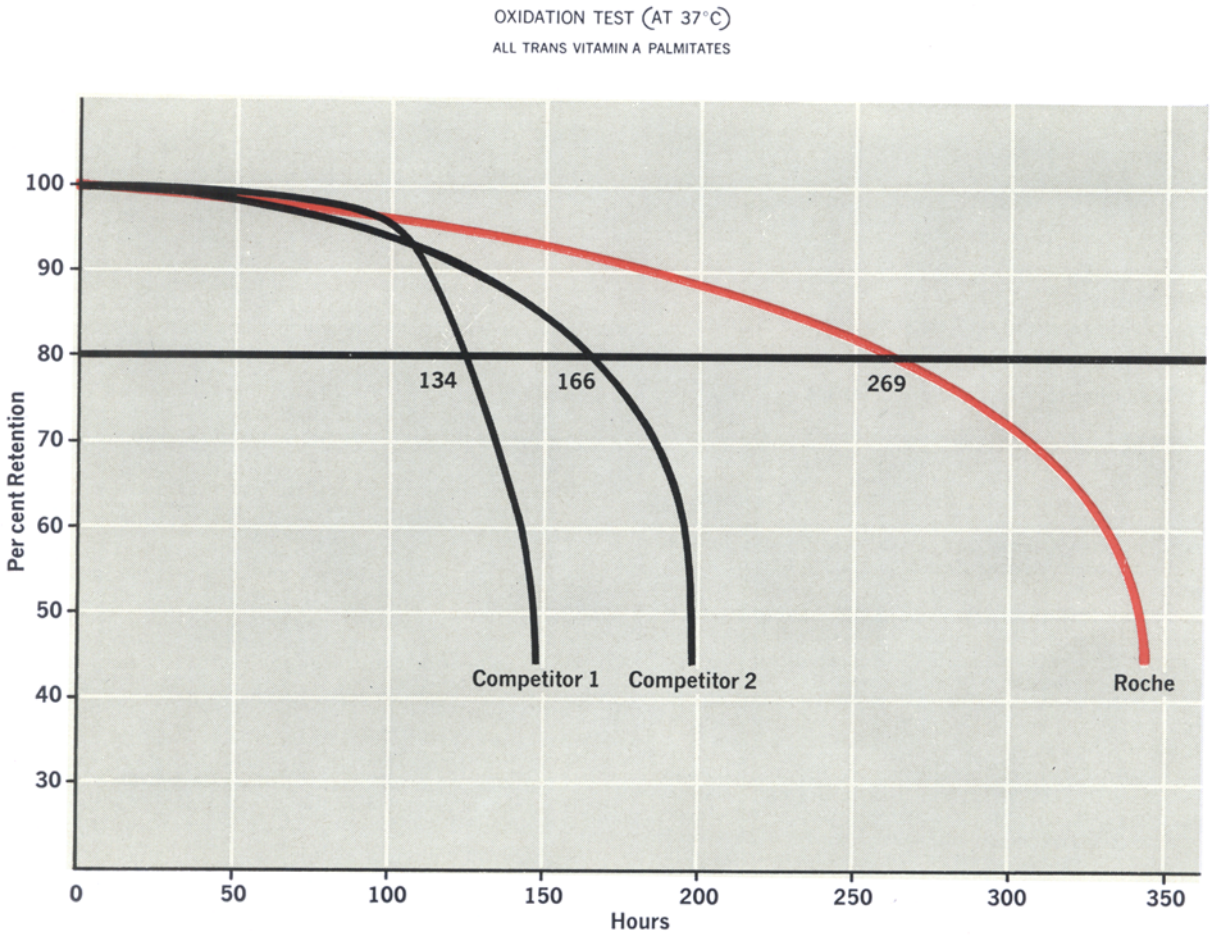

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OCCURRENCE OF THE ALLYLIC COMPOUNDS OF MONOSATURATED FATTY CHAINS. I. THE HYDROLYSIS OF FATTY ALLYL BROMIDES. M. Naudet, E. Ucciani and A. Peretz (Lab. of Fatty Acid Chem. Marseille, Fr.). *Rev. Franc. Corps Gras* 11, 247–251 (1964). Hydrolysis of fatty allyl bromides is a good method for preparation of unsaturated hydroxy fatty acids. Unsaturated fatty acids which have been brominated in the allylic position by N-bromosuccinimide yield 93% of the unsaturated acid by titrating with 2 M sodium bicarbonate at 100C. The hydroxy acids can be easily separated by crystallization at low temperatures. The hydrolysis by sodium bicarbonate is also applicable to the terminal functional compounds when the reaction is run under pressure in tetrahydrofurane.

COMPOSITION CONSISTING OF MALEIC ANHYDRIDE AND A FATTY ACID. M. Gans and J. L. Russell (Halcon International, Inc.). *U.S. 3,140,300.* The described composition consists of at least 99.5% by weight maleic anhydride and at least 0.07% of an aliphatic monocarboxylic acid having from 10 to 25 carbon atoms.

ACYL LACTYLIC ACID COMPOSITIONS AND METHODS OF PREPARATION THEREOF. B. D. Buddemeyer and J. R. Moneymaker (The Paniplus Co.). *U.S. 3,141,030.* Described are lactylic acid compositions corresponding to the formula $RCO(OCHCH_3CO)_nOZ$ in which RCO is a member selected from the group consisting of acyl radicals of fatty acids containing 16–24 carbon atoms, and mixtures thereof, Z is a cation, and n is the number of lactyl groups present per molecule of the composition, the value of n being on the average less than 1 but greater than 0. They are prepared by reacting a concentrated lactic acid composition having an average equivalent weight of between 95 and 130, based on free titratable acidity and containing less than 5 weight % polyacyllactic acid, with a fatty acid halide containing 16 to 24 carbon atoms at a temperature of between 50 and 95C.

REACTION PRODUCT OF MALEIC ANHYDRIDE UNSATURATED FATTY ACID ADDUCT AND POLYETHYLENE GLYCOL. S. B. Crecelius, T. E. Brunelle and L. M. Rue (Economics Laboratory, Inc.). *U.S. 3,141,897.* A composition of matter consists of a monoester of substantially equimolar proportions of a maleic anhydride unsaturated organic at least 10 carbon atom-containing fatty acid adduct and a polyethylene glycol having a molecular weight from about 300 to 800. The monoester is formed by opening of the anhydride functional group of the adduct and reaction of the polyethylene glycol therewith to form simultaneously the monoester and a free carboxyl group.

BREAD EMULSIFIER COMPOSITION AND PROCESS OF MAKING THE SAME. G. Dalby and H. C. Fisher (H. C. Fisher Co.). *U.S. 3,144,339.* An emulsion for making bread dough consists of an aqueous emulsion containing 15–25 parts by weight of glycerol monostearate, 11–20 parts of glycerine and 0.75–5 parts of lecithin and water to make a total of 100 parts.

LACTATE COMPOUND EMULSIFIERS AND SHORTENING CONTAINING THE SAME. S. W. Thompson (Lever Brothers Co.). *U.S. 3,144,341.* The described shortening agent comprises an edible triglyceride fat and an emulsifier containing a mono-diglyceride and a compound selected from the group consisting of stearyl lactoyl lactate, cetyl lactoyl lactate and mixtures thereof.

• Biology and Nutrition

COMPARISON OF UPTAKE OF PALMITIC, STEARIC, OLEIC AND LINOLEIC ACID BY POLYMORPHONUCLEAR LEUKOCYTES. P. Elsbach (Dept. of Med., New York Univ. School of Med., N.Y.). *Biochim. Biophys. Acta* 84, 8–17 (1964). A comparison was made of the incorporation *in vitro* of albumin bound 1-C^{14}- palmitic, stearic, oleic and linoleic acid by polymorphonuclear leukocytes obtained from rabbit peritoneal exudates. The distribution of radioactivity between phospholipid and non-phospholipid was different for each acid and remained constant during incubation for 1 hr. The amount of each acid incorporated is dependent upon its concentration in the medium. At equimolar concentrations in an artificial incubation mixture equal amounts of the four fatty acids were taken up. The possibility is discussed that fatty acid metabolism by the leukocyte meets an important part of the energy requirements of the cell, particularly during phagocytosis.

α-GLYCEROPHOSPHATE AS REGULATORY FACTOR IN FATTY ACID ESTERIFICATION. R. Tzur, E. Tal and B. Shapiro (Dept. of Biochem., The Hebrew Univ.-Hadassah Medical School, Jerusa-

lem, Israel). *Biochim. Biophys. Acta* 84, 18–23 (1964). L-α-Glycerophosphate has been shown to be the dominating fatty acid acceptor in the esterifying systems catalyzed by rat liver microsomes and mitochondria. The amounts of α-glycerophosphate present in the liver is near to the optimum for esterifying activity by the mitochondria but much below that by the microsomes. Almost all the α-glycerophosphate was found in the cell sap. Starvation, ethanol drinking and epinephrine injection reduce the glycerophosphate levels. Realimentation after starvation causes rapid regeneration of glycerophosphate levels.

LIPOLYTIC ACTIVITY IN ADIPOSE TISSUE HOMOGENATE TOWARD TRI-, DI- AND MONOGLYCERIDE SUBSTRATES. E. Gorin and E. Shafrir (Hadassah Univ. Hosp., Jerusalem). *Biochim. Biophys. Acta* 84, 24–34 (1964). In homogenates of adipose tissue, the rate of lipolysis of mono- and diglyceride substrates was 3–5 times higher than that of triglycerides. This relation between the rates of lipolysis was found in adipose tissues from various anatomic sites of rat, guinea pig and man; magnitude of activity toward all substrates varied with the species. Most of the lipolytic activity was found in the "fat-poor" portion of the homogenate. The activity toward all the three substrates resided mainly in the soluble supernatant fraction, about one-third to one-fourth in the microsomes and little remained in the mitochondria. The effect of incubation time, substrate and enzyme concentration of the lipolytic activity was studied. The optimal pH for the cleavage of triglycerides was found in the range of 6.8–7.0 and shifted to more alkaline ranges with partial glycerides. The susceptibility of lipolysis to temperature decreased from tri- to monoglyceride.

THE ISOLATION OF PHOSPHATIDYL GLYCEROL FROM RAT-LIVER MITOCHONDRIA. G. M. Gray (Dept. of Physiol. Chem., Johns Hopkins School of Med., Baltimore, Md.). *Biochim. Biophys. Acta* 84, 35–40 (1964). Phosphatidyl glycerol was isolated from a lipid extract of rat-liver mitochondria by a combination of column chromatography and thin-layer chromatography on silicic acid. It accounted for about 0.4% of the total phospholipids present. The fatty acid composition consisted mainly of palmitic (12%), stearic (14%), oleic (21%), linoleic (20%) and C_{20} polyenoic (20%) acids, and was very different from that of the cardiolipin in the mitochondria. The 0-amino acid esters of phosphatidyl glycerol which have been found in some bacteria were not detected in the mitochondria.

HYDROLYSIS OF CARDIOLIPIN BY SNAKE VENOM PHOSPHOLIPASE A. G. V. Marinetti (Biochemistry Dept., Univ. of Rochester School of Med., Rochester, N.Y.). *Biochim. Biophys. Acta* 84, 55–59 (1964). Phospholipase A (phosphatide acylhydrolase, EC 3.1.1.4) from snake venom has been shown to hydrolyze cardiolipin in wet ether and yield lyso-cardiolipin. The enzyme reaction is slow but is greatly stimulated by the addition of beef heart lecithin. The lysocardiolipin was characterized by chemical and chromatographic analysis and by its hydrolysis products. Lyso-cardiolipin has an ester: P ratio of 0.96 and yields monoglycerides upon acetic acid hydrolysis. Cardiolipin, on the other hand, has an ester: P ratio of 1.9 and yields diglycerides upon acetic acid hydrolysis. The fatty acids on the cardiolipin and lyso-cardiolipin are the same and thus it appears that these fatty acids have a fairly random distribution.

HEMOLYSIS AND SPLITTING OF HUMAN ERYTHROCYTE PHOSPHOLIPIDS BY SNAKE VENOMS. E. Condrea and A. DeVries (Labor Sick Fund-Beilinson Hosp., Petah Tikva, Israel). *Biochim. Biophys. Acta* 84, 60–73 (1964). Lysis of washed human erythrocytes induced by Cobra snake venoms (*Naja naja, Hemachatus haemachates*) was accompanied by splitting of the red-cell phospholipids to lysophosphatides. Phospholipase A fractions (phosphatide acyl-hydrolase, EC 3.1.1.4) isolated from the Cobra venoms were devoid of hemolytic activity and caused no significant breakdown of phospholipids in the intact erythrocytes. Another protein component derived from the same venoms was weakly hemolytic but showed no phospholipase activity. The two fractions, when combined, produced strong hemolysis and concomitant hydrolysis of erythrocyte phospholipids to lysophosphatides. Viper venoms (*Vipera palestinae* and *Vipera russellii*) containing phospholipase A but no direct lytic factor induced neither lysis nor substantial phospholipid splitting of washed erythrocytes unless fortified with the direct lytic factor fraction derived from Cobra venom.

EFFECT OF PROTEIN ON GLYCEROL COLOR DEVELOPMENT DURING THE ASSAY OF LIPOPROTEIN LIPASE. R. S. Levy and E. D. McGee (Dept. of Biochemistry, Univ. of Louisville School of Medicine, Louisville, Ky.). *J. Lipid Res.* 5, 265–7 (1964). The inhibition by proteins of color development in the estimation of glycerol during assay of lipoprotein lipase is shown to be due to the binding of formaldehyde to protein in a form preventing subsequent color formation.

IMPROVED DETERMINATION OF GLYCEROL AND FATTY ACIDS IN GLYCERIDES AND ETHANOLAMINE PHOSPHATIDES BY GAS-LIQUID CHROMATOGRAPHY. K. S. Holla, L. A. Horrocks and D. G. Cornwell (Lab. of Neurochemistry, Cleveland Psychiatric Inst., Cleveland, Ohio). *J. Lipid Res.* 5, 263–5 (1964). The fatty acid and glycerol content of glycerides were estimated by a simplified hydrogenolysis-acetylation procedure and GLC. When fatty acids interfered in the analysis, glycerol alone was estimated by saponification-acetylation and GLC. Preliminary acetolysis was necessary for the estimation of glycerol in ethanolamine phosphatides by these methods.

INFLUENCE OF VITAMIN D ON VARIOUS ASPECTS OF THE REPRODUCTIVE PROCESS IN MATURE HENS. J. L. Turk and J. McGinnis (Dept. of Animal Sciences, Washington State Univ., Pullman, Wash.). *Poultry Sci.* 43, 539–46 (1964). Experiments with laying hens revealed that while a deficiency of vitamin D caused a marked decrease in egg production, it had no effect on the secondary sex characteristics of the birds. Oviduct size was slightly reduced in the deficient hens, but they were still functional. There was no observable effect on the ovaries of the deficient hens; however, ova size was somewhat reduced. These observations suggest that vitamin D is involved in regulating ovulation and/or follicular growth in the laying hen. In addition, a deficiency of vitamin D resulted in a marked decrease in egg weight and shell thickness and an increase in the incidence of shell-less and broken eggs, as well as blood spots. A reduced total blood calcium was observed in the deficient hens having secondary sex characteristics indicative of the laying condition, but there was no effect in typical cull hens. A vitamin D deficiency did not lower percentage tibia bone ash.

STUDIES IN EXPERIMENTAL DIABETES. IV. FREE FATTY ACID MOBILIZATION. M. E. Tarrant, R. Mahler and J. Ashmore (Dept. of Pharmacology, Indiana Univ. School of Med., Indianapolis, Ind.). *J. Biol. Chem.* 239, 1714–9 (1964). Rats, mice, and dogs were made acutely insulin-deficient by injection of anti-insulin serum. The concentration of free fatty acids in their plasma rose within 1 hour. This rise appeared to be due to release of free fatty acids from adipose tissue and not to any defect in their utilization. The free fatty acid release could not be correlated with any impairment of glucose metabolism or fatty acid esterification, either *in vitro* or in the intact animal. Glycerol release by isolated adipose tissue from insulin-deficient rats was elevated, and was reduced towards normal by insulin in the absence of glucose. It is suggested that free fatty acid mobilization in the early stages of insulin deficiency is due to increased lipolysis of adipose tissue triglycerides.

EFFECT OF DIET ON AORTIC RUPTURES IN TURKEYS INDUCED BY DIETHYLSTIBESTROL. C. F. Simpson and R. H. Harms (Florida Ag. Expt. Sta., Gainesville, Fla.). *Poultry Sci.* 43, 681–5 (1964). Data have been presented which indicate that treatment of male Broad-Breasted Bronze turkeys with diethylstilbestrol (DES) caused high mortality as a consequence of aortic rupture. Aortic ruptures were produced when poults were treated with DES weekly and fed three diets which varied in composition. A high incidence of ruptures occurred in birds fed 2 different diets and treated with 30 or 60 mg of liquid DES. Birds treated similarly and fed a third diet did not develop as many aortic ruptures. Although poults from one source grew more rapidly than poults from a second source, no difference was observed in incidence of aortic rupture among birds treated and fed similarly. Rate of weight gain did not appear to be the sole limiting factor associated with incidence of aortic rupture.

BIOSYNTHESIS OF 5α-CHOLESTAN-3β-OL IN THE RABBIT AND GUINEA PIG. S. Shefer, S. Milch and E. H. Mosbach (Dept. of Lab. Diagnosis, Public Health Res. Inst. of N. Y. City and Bureau of Lab., N. Y. City Dept. of Health, N. Y. 9). *J. Biol. Chem.* 239, 1731–6 (1964). The biosynthesis in vivo of 5α-cholestan-3β-ol was demonstrated in the rabbit and the guinea pig. Potential precursors of 5α-cholestan-3β-ol labeled

with C^{14} were administered to rabbits and guinea pigs, and 5α-cholestan-3β-ol was isolated from liver, intestinal wall, and adrenals at the end of 24 hours. In the guinea pig, labeled mevalonate gave rise to labeled 5α-cholestan-3β-ol in all three of these tissues, while in the rabbit this conversion could be demonstrated only in the adrenals. After the administration of labeled Δ^4-cholestenone to a rabbit, 90 to 95% of the total sterol radioactivity was present in the 5-α-cholestan-3β-ol fraction of liver, adrenals, and intestinal wall. The other tracers tested (cholesterol, desmosterol, and acetate) did not produce significant quantities of labeled 5α-cholestan-3β-ol in the rabbit under the conditions of these experiments.

TERPENE METABOLISM IN THE RAT TESTIS. I. THE CONVERSION OF ISOPENTENYL PYROPHOSPHATE TO SQUALENE AND STEROLS. R. A. Salokangas, H. C. Rilling and L. T. Samuels (Dept. of Biological Chemistry, Univ. of Utah College of Med., Salt Lake City, Utah). *Biochemistry* 3, 833–7 (1964). Certain aspects of the terpenoid biosynthetic pathway in testis have been investigated. It has been shown that C^{14} isopentenyl pyrophosphate is readily converted to squalene and to a much lesser extent to lanosterol and cholesterol. The accumulation of radioactive squalene in rat testis homogenates cannot be accounted for by either a lack of cofactors or the presence of a large endogenous pool of squalene. The squalene content of rat testis has been found to be 9 μg/g wet tissue.

THE CONVERSION OF GLUTAMATE CARBON TO FATTY ACID CARBON VIA CITRATE. I. THE INFLUENCE OF GLUCOSE IN LACTATING RAT MAMMARY GLAND SLICES. J. Madsen, S. Abraham and I. L. Chaikoff (Dept. of Physiology, Univ. of Calif., Berkeley 4, Calif.). *J. Biol. Chem.* 239, 1305–09 (1964). The stereospecific cleavage of citrate by the citrate cleavage enzyme was confirmed in lactating rat mammary gland and a role for this enzyme was assessed. When lactating rat mammary gland slices were incubated with glutamate-2-C^{14} or glutamate-5-C^{14} in the presence of glucose, a metabolic pathway via α-ketoglutarate → isocitrate → *cis*-aconitate → citrate to acetyl-CoA and oxaloacetate (a backward reaction of the Krebs cycle) was suggested by (a) the finding of appreciable amounts of C^{14} in the isolated fatty acids and (b) the location of the C^{14} in these fatty acids. A method for estimating the extent of this backward reaction is presented. It was calculated that, of the total amount of glutamate metabolized via the Krebs cycle in the presence of glucose, 20 to 30% proceeds by way of this backward pathway. In the absence of glucose, a much smaller fraction of the glutamate metabolized via the Krebs cycle goes by the backward reaction.

STUDIES WITH ACIDULATED COTTONSEED-OIL SOAPSTOCK. 1. ITS USE AS A FAT SUPPLEMENT IN PRACTICAL BROILER RATIONS. B. Lipstein and S. Bornstein (Div. of Poultry Husbandry, The National and Univ. Inst. of Ag., Rehovot, Israel). *Poultry Sci.* 43, 686–93 (1964). The value of acidulated cottonseed-oil soapstock and the tolerance level of gossypol, in diets of broilers, were tested in one 5-week and two 10-week battery trials involving 2,314 chicks. The results obtained in these trials indicate that 0.10% dietary gossypol (supplied by this soapstock) is the safe upper tolerance level. Up to this limit there is no difference between acidulated cottonseed- and acidulated soybean-oil soapstock in terms of chick weight gains and feed/gain ratio. Levels of gossypol above 0.10% depress growth rate by means of reduced feed consumption, with a relatively smaller effect on the feed/gain ratio. At concentrations which exceed 0.20%, gossypol causes high mortality. In one trial, lysine supplementation did not affect the gossypol tolerance level.

2. ATTEMPTS TO REDUCE ITS GOSSYPOL CONTENT. *Ibid.*, 694–701. The data presented indicate that the gossypol content of acidulated cottonseed-oil soapstock is influenced by the fol-

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