

ABSTRACTS

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

• Fats and Oils

CHANGES IN LIPID COMPOSITION IN WHEAT DURING STORAGE DETERIORATION. R. D. Daftary and Yeshajuhu Pomeranz (Dept. of Flour and Feed Milling Industries, Kansas State Univ., Manhattan, Kan.). *J. Agr. Food Chem.* 13, 442-46 (1965). Titratable acidity was about 7% higher in benzene extracts than in petroleum ether extracts of wheat. Benzene extracts of moistened wheat contained more free fatty acids than did extracts of wheat redried to 11 to 12% moisture. Changes in lipid composition during grain deterioration were followed by qualitative and quantitative thin-layer chromatography (TLC) and fractionation on silicic acid columns. Deterioration of wheat was accompanied by formation of at least four unidentified compounds that showed autofluorescence under ultraviolet light. Grain deterioration was accompanied by lowering of polar lipids and rapid disappearance of at least five ninhydrin- or Dragendorff-reagent positive polar lipids. The breakdown of polar lipids was more rapid and more intensive than formation of free fatty acids or disappearance of triglycerides.

LIPIDS ASSOCIATED WITH ACID-PRECIPIATED CASEIN. J. Cerbulis and C. A. Zittle (Eastern Regional Res. Lab., Phila., Pa.). *J. Dairy Sci.* 48, 1154-56 (1965). Acid-precipitated casein contains 4.5-7% of lipids on dry weight basis. About 40-50% of the total lipid is extractable with petroleum ether and is presumably free fat. The remaining 50-60% of the lipids are more strongly associated with the casein. Phospholipids appear mainly in the latter fraction; ratio of phospholipid to total lipids is greater in casein than in whole milk lipids.

CONSISTENCY OF ISOBARIC BINARY VAPOUR/LIQUID EQUILIBRIUM DATA: METHYL ESTERS OF FATTY ACIDS. K. A. Naik, A. Husain and K. S. Chari. *Indian J. Tech.* 2 (8), 255-8 (1964). The visual methods proposed for testing the consistency of isothermal or isobaric binary vapour/liquid equilibrium data have been found applicable to binary mixtures of fatty acid esters even when the difference between the boiling points of the components is $> 30^{\circ}\text{C}$. The composition-resolution method with the use of excess free energy function has been found to provide a convenient basis for smoothing and extrapolating isobaric experimental data when the heat of mixing between the components can be neglected. (Rev. Current Lit. Paint Allied Ind. #278).

DETERMINATION OF ESTERIFIED OLIVE OIL DERIVED FROM ORUJO OIL BY MEANS OF PANCREATIC LIPASE. PRELIMINARY NOTE. A. M. Vela (Inst. de la Grasa y sus Derivados, Sevilla, Spain). *Grasas y Aceites* 16, 69-72 (1965). A method is proposed for identifying esterified olive oil derived from orujo oil (sulfur oil) and for determining the amount of esterified oil in mixtures with natural olive oil. This method is based on the low proportion of saturated fatty acid chains in the β position of natural olive oil and the much larger amount of saturated fatty acids in this position in esterified olive oil. The composition of the fatty acids in the β position is determined using pancreatic lipase. The lipase hydrolyzes the external (α) fatty acid chains leaving unaffected the central ones, thus producing free fatty acids and beta-monoglycerides. A formula is given relating the concentration of saturated fatty acids in the β position to esterified oil content.

STRUCTURE OF AN OPTICALLY ACTIVE ALLENE-CONTAINING TETRAESTER TRIGLYCERIDE ISOLATED FROM THE SEED OIL OF SAPIUM SEBIFERUM. H. W. Sprecher, R. Maier, M. Barber and R. T. Holman (Hormel Inst., Univ. Minn., Austin, Minnesota). *Biochemistry* 4, 1856-63 (1965). The optically active lipid isolated from the seed oil of the Chinese tallow tree, *Sapium sebiferum*, previously considered to be a triglyceride containing 2,4-deca-dienoic acid esterified on one of the primary hydroxyls of glycerol, is shown to be tetraester triglycerides (I). Mass spectral analysis showed that in the completely hydrogenated I stearic acid is esterified to one primary and the secondary

hydroxyl group of glycerol. The other primary hydroxyl of glycerol is esterified to 8-hydroxyoctanoic acid, and its ω -hydroxyl group, in turn is esterified to decanoic acid. In I the predominant common fatty acids are linoleic and linolenic acids. The eight-carbon hydroxy acid is 8-hydroxy-5,6-octadienoic acid.

ISOLATION OF 11-CYCLOHEXYLUNDECANOIC ACID FROM BUTTER. J. C. M. Schogt and P. Haverkamp Begemann (Unilever Res. Lab., Vlaardingen, The Netherlands). *J. Lipid Res.* 6, 466-70 (1965). After fractionation using distillation, recrystallization, urea treatment, hydrogenation, and repeated urea treatment, a hitherto unknown acid has been isolated from butter fatty acid methyl esters. Gas-liquid chromatography followed by mass and infrared spectroscopy showed that the acid is saturated and it contains 17 carbon atoms and a terminal cyclohexyl group. Synthetic 11-cyclohexylundecanoic acid appeared to be identical with the isolated acid. Butter contains at least 0.01% of this acid.

PURIFICATION OF NEUTRAL LIPID FRACTIONS BY THIN-LAYER CHROMATOGRAPHY ON ALUMINUM OXIDE. J. M. Lederkremer and R. M. Johnson (Inst. of Nutr. and Food Technol., The Ohio State Univ., Columbus, Ohio). *J. Lipid Res.* 6, 572-74 (1965). Two-stage thin-layer chromatography on Aluminum Oxide G has been used to separate triglycerides, cholesterol, cholesterol esters, and diglycerides from contaminating fatty acids present in the lipid mixtures. Negligible hydrolysis of glycerol esters occurs. The method is particularly useful in metabolic experiments conducted *in vitro*, in which radioactive fatty acids are employed as precursors.

LARGE-SCALE SEPARATION OF FATTY ACID METHYL ESTERS BY COLUMN CHROMATOGRAPHY ON ACID-WASHED FLORISIL IMPREGNATED WITH SILVER NITRATE. R. L. Anderson and E. J. Hollenbach (The Procter and Gamble Co., Miami Valley Labs., Cincinnati, Ohio). *J. Lipid Res.* 6, 577-78 (1965). Rapid, large-scale separation of fatty acid methyl esters has been accomplished with acid-washed Florisil impregnated with silver nitrate. Recovery of material was quantitative and the peaks contained only one type of fatty acid; i.e., saturated, monoenoic, or dienoic acid.

TRANS UNSATURATED FATTY ACIDS IN ANIMAL FATS. R. Guillaumin (Inst. Corps Gras, Paris). *Fette Seifen Anstrichmittel* 66, 907-9 (1964). The depot and organ fats of ruminants sometimes reaches as high a content of *trans* acids as 11%. In non-ruminants *trans* isomeric fatty acids are rarely found.

STABILITY OF OILS AND FATS DURING LONG STORAGE IN PACKAGES MEANT FOR SALE. E. Winter (Bundesforschung Anstalt Lebensmittelrischhaltung, Karlsruhe). *Fette Seifen Anstrichmittel* 67, 124-30 (1965). The storage properties of groundnut oil, soybean oil, sunflower oil, coconut oil and biskin in the usual commercial packages were investigated at 0 and 25°C. Changes in PV, TBA, and acid values were measured and compared with changes in odor, taste, and consistency over a 3-year period. A significant improvement in the storage stability of oils and fats was found when they were sealed under vacuum or nitrogen.

PAPER CHROMATOGRAPHIC SEPARATION OF CRITICAL PAIRS OF HIGHER FATTY ACIDS FROM THE PRODUCTS BY THE PROCEDURE OF MARGOSCHES FOR THE DETERMINATION OF THE IODINE VALUE OF FATS. G. Rankoff and D. Rankoff (Inst. Org. Chem., Bulgarian Acad. Sci., Sofia). *Fette Seifen Anstrichmittel* 66, 912-15 (1964). The paper-chromatographic method when used on the iodinated, unsaturated fatty acids from olive, sunflower and groundnut oils showed the presence of diastereoisomers of the erythro and threo-iodinated hydroxy acids.

CHANGES IN METHYL ESTERS OF FATTY ACIDS ON HEATING OXIDATION PRODUCTS. K. H. Ney (Lab. Margarine Union, Hamburg, Bahrenfeld). *Fette Seifen Anstrichmittel* 67, 190-94 (1965). The hydroperoxide of methyl oleate reacts on heating to form the methylepoxystearate. Methyl epoxystearate upon heating is converted into the methyl ester of keto stearic acid. These reactions do not alter the number of carbon atoms in the molecule. Other reactions take place in which medium chain aldehydes and ketones are formed.

RECOVERY OF NICKEL AND FAT FROM SPENT NICKEL CATALYST. D. Rebello and K. D. Mukherjee (Dept. of Chem. Technol., Univ. Bombay, India). *Fette Seifen Anstrichmittel* 67, 81-85 (1965). Spent nickel catalyst is treated with mineral acid in

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the presence of a fat-dissolving, water immiscible solvent such as hexane. The solutions are then separated to yield the pure nickel salt and the hardened fat.

THIN LAYER CHROMATOGRAPHY IN THE FIELD OF FATS XVI: EXPERIMENTS ON THE TWO DIMENSIONAL ANALYSIS WITH THE HELP OF PHOTOMETRY. H. P. Kaufmann and K. D. Mukherjee (Inst. Indust. Fettforsch., Münster). *Fette Seifen Anstrichmittel* 67, 183-87 (1965). Methyl esters and glycerides of mixtures of fatty acids as well as cholesterol esters were separated using thin-layer chromatography on silver nitrate-treated silica gel G. The chromatograms were assayed photometrically after charring.

RUBBER COMPOUNDED WITH A RELEASING COMPOSITION CONTAINING A BRANCHED CHAIN ALCOHOL AND STEARIC ACID. E. Avon (Technical Processing, Inc.). U.S. 3,201,361. The described composition consists of a homogeneous mixture of an oleate salt of a metal such as zinc, magnesium, lead or calcium in an amount such that the oleic acid equivalent amounts to 11-16% of the total mixture; 50-60% of a lubricant (petroleum jelly or paraffin wax); 4-6% monohydric branched-chain alkylol having from 8 to 18 carbon atoms; 4.5-6.7% potassium stearate; and 14.26% free stearic acid.

PROCESS FOR THE CONTINUOUS REMOVAL OF BREAK (MUCILAGINOUS PRODUCTS) FROM, AND PURIFICATION OF, VEGETABLE OILS AND FATS. R. Raffaeta. U.S. 3,206,487. The process comprises the steps of: (a) confining a mixture of about equal volumes of oil and water in a closed circuit; (b) continuously circulating the mixture in the closed circuit; (c) continuously emulsifying the mixture during the course of its circulation; (4) continuously removing oil and water from the closed circuit; (e) introducing oil and water into the closed circuit at a total flow rate equal to the total flow rate at which oil and water are removed; and (f) centrifuging the removed oil and water to separate the purified oil from the water.

• Fatty Acid Derivatives

THERMAL CONDUCTIVITY OF CASTOR OIL-BASED RIGID URETHANE FOAMS. C. K. Lyon, V. H. Garrett, D. R. Black, T. H. Applewhite and L. A. Goldblatt (West. Reg. Res. Lab., West. Utilization Res. and Dev. Div., U.S. Dept. of Agri., Albany, Calif.). *Ind. Eng. Chem. Product Res. Dev.* 4, 189-91 (1965). The effect of aging on thermal conductivities of comparable castor oil-based and polyether-based rigid urethane foams was determined. Uncut samples of both foam types (representative of foam-in-place applications) maintained equivalent low thermal conductivities throughout test periods of 6 to 12 months. With cut samples, commonly used for testing, thermal conductivities increased at different rates on aging. Mass spectroscopic studies showed that the relatively greater initial increase in thermal conductivity of cut samples of castor oil-based foams was due to a higher rate of diffusion of air into the foam cells.

SOME REACTIONS OF UNSATURATED FATTY ACIDS AND THEIR DERIVATIVES IN MOLTEN ALKALIES. M. F. Ansell, A. N. Radziwill, D. J. Redshaw, I. S. Shepherd, D. Wallace and B. C. L. Weedon (Queen Mary College, Mile End Road, London E. 1, England). *J. Agr. Food Chem.* 13, 399-401 (1965). The labeling pattern of saturated fatty acids formed by treatment of unsaturated fatty acids with molten potassium deuterioxide indicates that reactions of the Varrentrapp type proceed by an initial sequence of discrete, reversible, prototropic rearrangements. The principal reactions during the alkali fusion of aleuritic acid have been identified. Alkali fusion of epoxy derivatives of unsaturated fatty acids has been shown to involve initial opening of the epoxide ring in at least three ways: hydrolysis, reduction or rearrangement, and β -elimination. Reactions of the latter type predominated in the alkali fusion of ω -alkoxy fatty acids.

SYNTHESIS OF NATURALLY OCCURRING UNSATURATED FATTY ACIDS BY STERICALLY CONTROLLED CARBONYL OLEFINATION. L. D. Bergelson and M. M. Shemyakin (Inst. Chem. of Nat. Prods., USSR Acad. Sci., Moscow USSR). *Angew. Chem. Internat. Edit.* 3, 250 (1964). A review. Carbonyl olefination can be sterically controlled to obtain *cis*-olefins. This reaction may be used to synthesize unsaturated and polyunsaturated fatty acids with differing positions and sequences of *cis*- and *trans* double bonds and to ω -hydroxy and branched chain fatty acids.

AUTOXIDATION OF METHYL LINOLEATE IN WATER I. CONTRIBUTION: ISOLATION AND IDENTIFICATION OF N-AMYLHYDROPEROXIDE AND 2-OCTENAL. W. Klopffer, H. Esterbauer and E. Schauen-

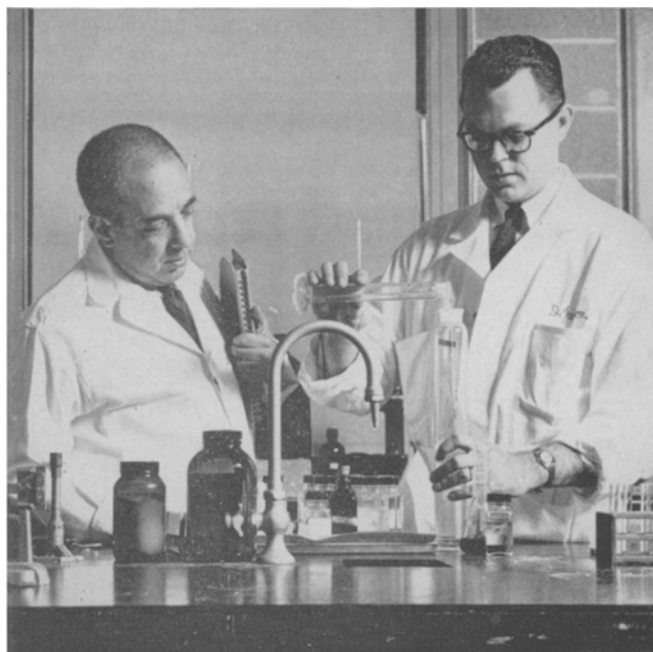
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stein (Lehrkanzel Biochem., Univ. Graz, Austria). *Fette Seifen Anstrichmittel* 67, 198-203 (1965). From the water soluble reaction products formed during the autoxidation of methyl linoleate two compounds were isolated by means of adsorption chromatography. Infrared and ultraviolet spectrophotometry and polarography were used to characterize the compounds. The reduced n-amyldihydroperoxide (pentanol -1) was analyzed in the form of its 4,4'-nitroazo-benzenecarbonic acid ester and the 2-octenal as its 2,4-dinitrophenylhydrozone. The mechanism of the formation of these compounds is discussed.

BIFUNCTIONAL FATTY ALCOHOLS III. LINOLENIC ACID AS STARTING MATERIAL FOR THE PREPARATION OF LONG CHAIN ALKANDIOLS-1,4. K. Thewalt and W. Rudolph (Lab. Chem. Works, Witten, Witten/Ruhr). *Fette Seifen Anstrichmittel* 67, 187-89 (1965). Unsaturated ketoesters of carboxylic acids with 9 to 13 carbon atoms are formed by the δ -condensation of linolenic acid ester with aldehydes. Hydrogenation of the same in the presence of copper chromite catalysts at 230-240C yields 55-85% of theoretical amounts of primary mono and primary secondary bifunctional fatty alcohols with hydroxyl groups in the γ -position.

TETRAALKYL DIMERIC FAT AMINES. R. Fisher, L. R. Vertnik, K. E. McCaleb and N. M. Le Bard (General Mills, Inc.). *U.S. 3,201,471*. A compound is described having the structure $R_1R_2NCH_2DCH_2NR_3R_4$ in which $R_1, R_2, R_3,$ and R_4 are selected from the group consisting of aliphatic hydrocarbon radicals with 1 to 18 carbon atoms and hydroxy substituted aliphatic hydrocarbon radicals with 1 to 18 carbon atoms, and D is a dimeric fat radical.

• Biochemistry and Nutrition

ANTAGONISM BETWEEN VITAMINS A AND K IN THE GERM-FREE RAT. B. S. Wostmann and P. L. Knight (Lobund Lab. Dept. of Bio., Univ. of Notre Dame, Notre Dame, Indiana). *J. Nutr.* 87, 155-60 (1965). Weanling germ-free male rats were fed a semi-purified diet complete except for being free of vitamin K and containing only approximately 0.4 IU of vitamin A activity/g. Vitamins A and K₁ were administered separately, vitamin A to the amount of zero, 5, 50, 200 and 200 IU/day, and vitamin D first at a level of 2 μ g/day, later to the amount of 0.5 μ g/day. A daily intake of 0.5 μ g/day allowed only a limited lifespan before the rats died with all the symptoms of the hemorrhagic syndrome typical of a vitamin K deficiency. Compared with the survival time observed with vitamin A intakes usually considered adequate, lifespan was shortened by a daily intake of 2000 IU/day, possibly shortened at an intake level of 200 IU, and definitely prolonged without vitamin A supplementation. This last group of rats demonstrated reduced growth associated with the low vitamin A intake, presumably leading to a lower requirement for vitamin K. It was concluded that in the usual range of vitamin A intake for germ-free rats fed the diets commonly used in germ-free experimentation, no effect of vitamin A intake upon vitamin K requirement is demonstrable.

EFFECT OF DIETARY CHOLESTEROL ON MAN'S SERUM LIPIDS. F. Grande, J. T. Anderson, C. Chlouverakis, M. Proja and A. Keys (Lab. of Physiol. Hygiene, Univ. of Minn., Minneapolis, Minn.). *J. Nutr.* 87, 52-62 (1965). Serum cholesterol levels in man were found to be proportional to the square root of dietary cholesterol level. The change of serum cholesterol (mg/100 ml) produced by substituting a diet containing Z₂ mg of cholesterol/1000 kcal for another containing Z₁ mg of cholesterol/1000 kcal is given by the formula $\Delta \text{Chol} = 1.5 (Z_2 - Z_1)$. This formula is based on experiments in which the daily cholesterol supplement was dissolved in 100 g of oil which was in turn incorporated into the diet or given in the form of dry egg yolk mixed with the same amount of oil, but it also applies to other results reported in the literature with dietary cholesterol ranging from zero to about 3000 mg/day. The effect of dietary cholesterol on serum cholesterol and phospholipids was independent of the degree of saturation of the dietary fat. Conversely, the effect on the serum lipids of changing the composition of the dietary fat was independent of the cholesterol content of the diet at either 50 or 1500 mg/day.

LIPID BIOSYNTHESIS IN AORTIC INTIMA FROM NORMAL AND CHOLESTEROL-FED RABBITS. A. F. Whereat (School of Med., Univ. Penn., Phila., Pa.). *J. Atheroscler. Res.* 4, 272-82 (1964). The atherosclerotic intima differs from the normal in the rates of certain metabolic functions. Oxygen consumption is accelerated in the atherosclerotic intima. Succinate stimulates oxygen consumption to a much greater degree than any other substrate tested. Succinate oxidation is incomplete, and fumarate and malate are the only quantitatively significant products. We were unable to demonstrate incorporation of either acetate or mevalonate into cholesterol in either the control or atherosclerotic intima under the conditions of our experiments. Fatty acid synthesis from acetate is significantly increased in the atherosclerotic aortic intima. Succinate oxidation stimulates the fatty acid synthesis from acetate but is not itself incorporated into the lipid. The rate of fatty acid synthesis is greatest where the lesions are most advanced.

ACCUMULATION OF LIPID AND NONLIPID CONSTITUENTS IN RABBIT ATHEROMA. H. A. I. Newman and D. B. Zilversmit (Dept. of Physiology, Univ. of Tenn., Memphis, Tenn.). *J. Atheroscler. Res.* 4, 261-71 (1964). Six groups of 10 rabbits each were fed 1% cholesterol for periods up to 4 months. Free and esterified cholesterol of the thoracic intima were observed to increase exponentially early in the experiment. The rate of cholesterol accumulation was found to depend on time of exposure to atherogenic diet as well as on serum cholesterol level. The data suggest that height of serum cholesterol level will not substitute for length of exposure to cholesterol feeding. Aortic phospholipids also increased exponentially after an initial lag period. Aortic triglycerides, DNA and protein showed relatively minor changes during the same period. It is concluded that these changes are consistent with the view that aortic phospholipids increase in response to cholesterol infiltration and that the increase does not reflect cellular proliferation.

ATHEROMATOUS PLAQUES IN AORTAS OF ESSENTIAL FATTY ACID DEFICIENT RATS. S. Kahn (Squibb Inst. Med. Res., New Brunswick, N.Y.). *Proc. Soc. Exp. Biol. Med.* 119, 1035-36 (1965). Gross atherosclerotic plaques were produced in the abdominal

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aortas of female rats maintained for one year on a diet deficient in essential fatty acids but with an elevated content of cholesterol and saturated fat. Male rats were not similarly affected.

EFFECT OF NICOTINIC ACID ON SERUM TRIGLYCERIDE, CHOLESTEROL AND CHYLOMICRONS IN RATS. R. S. Jacobs, M. S. Grebner and D. L. Cook (Dept. of Pharmacology, G. D. Searle and Co., Chi., Ill.). *Proc. Soc. Exp. Biol. Med.* 119, 1117-20 (1965). The effects of nicotinic acid on serum lipids have been studied in rats. Significant decreases in serum triglyceride and cholesterol were observed in fasted rats 4 hours after oral administration of nicotinic acid. Greater reductions in triglyceride occurred than in cholesterol. In addition, in another study, oral or parenteral administrations of nicotinic acid significantly inhibited fat-induced chylomionemia.

ABSORPTION OF DIETARY FATS BY THE RAT IN CHOLESTYRAMINE-INDUCED STEATORRHEA. R. W. Harkins, L. M. Hagerman and H. P. Sarett (Dept. of Nutr. Res., Mead Johnson Res. Center, Evansville, Indiana). *J. Nutr.* 87, 85-92 (1965). Cholestyramine, a bile acid-sequestering resin, impaired fat absorption in weanling rats fed a diet containing 15% corn oil. With no cholestyramine, 94% of the fats were absorbed. Absorption decreased with increasing amounts of resin, and with 10% cholestyramine in the diet, only 35% of the fat was retained. In a second study, 5% cholestyramine decreased the retention of a variety of dietary fats as follows: (in per cent) medium-chain triglycerides, 2; coconut oil, 7; safflower oil, 16; soy oil, 17; corn oil, 20; olive oil, 27; butterfat, 28; and lard, 37. Two per cent cholestyramine had little effect upon net calcium absorption although 5% decreased absorption with all dietary fats. The type of dietary fat had little effect on the fecal excretion of bile acids. Two per cent cholestyramine increased the fecal excretion of bile acids 30-fold over control levels although 5% cholestyramine did not further increase bile acid excretion. Net retention of the medium-chain fatty acids and linoleic acid was high even with marked steatorrhea, whereas the long-chain saturated fatty acids and oleic acid were poorly absorbed.

INCIDENCE OF TERATOGENY INDUCED BY VITAMIN E DEFICIENCY IN THE RAT. R. A. Gortner, Jr. and Joanne Ekwurtzel (Shanklin Lab. of Biology, Wesleyan Univ., Middletown, Conn.). *Proc. Soc. Exp. Biol. Med.* 119, 1069-71 (1965). Holtzman and Sherman strain female albino rats were raised on tocopherol-free or tocopherol-supplemented diets for 3 months, then bred to normal males. On the 9th to 11th day of gestation most of the depleted females received a tocopherol supplement by stomach tube. All animals were sacrificed on the 21st day of gestation and the uteri and fetuses examined for resorptions and congenital malformations. Of the 92 developed fetuses produced by those rats receiving the delayed supplements, only 4, or less than 5% of the total, showed any gross abnormalities.

EFFECTS OF NICOTINIC ACID (NA) AND NICOTINAMIDE (NAM) ON SERUM CHOLESTEROL AND ERYTHROCYTE NICOTINAMIDE ADENINE DINUCLEOTIDE (NAD) LEVELS OF RABBITS. F. Fontenot, H. Redetzki and R. Deupree (Louisiana State Univ. of Med., New Orleans). *Proc. Soc. Exp. Biol. Med.* 119, 1053-55 (1965). The influence of NA and NAM administration on erythrocyte NAD and serum cholesterol of cholesterol fed rabbits was investigated. NAM was more effective than NA in both elevation of erythrocyte NAD concentrations and lowering of serum cholesterol. The results were discussed in support of the hypothesis of Altschul that a significant relationship exists between the effects of NA and NAM on serum cholesterol and erythrocyte NAD levels.

INFLUENCE OF CARBOHYDRATE-TO-FAT RATIO ON METABOLIC CHANGES INDUCED IN RATS BY FEEDING DIFFERENT CARBOHYDRATE-FAT COMBINATIONS. Catherine Carroll and Elizabeth Bright (Dept. of Home Economics, Agr. Expt. Station, Univ. of Arkansas, Fayetteville, Ark.). *J. Nutr.* 87, 202-10 (1965). The study was designed to determine whether changing the relative proportions of carbohydrate and fat in the diet would influence metabolic responses in rats to different sources of the 2 nutrients. Four carbohydrate-fat combinations (glucose and fructose each with corn oil (CO) and with hydrogenated coconut

oil (HCO)) were combined in high carbohydrate:low fat and low carbohydrate:high fat diets. Protein and caloric values of all diets were equivalent. Eight groups of male, weanling rats were each fed one of the experimental diets for 2 to 4 weeks. Reducing the carbohydrate-to-fat ratio from 64:5 to 19:25 (by weight) resulted in the following changes in liver functions: 1) marked reduction or complete elimination of responses of the glucose-6-phosphatase and fructose diphosphatase enzyme systems to dietary fructose; 2) significant increase in response of glucose-6-phosphatase to dietary HCO; 3) decreases in liver glycogen, to a different extent with different carbohydrate-fat combinations; 4) striking increases in total lipid in rats fed CO or HCO with glucose; and 5) increases in cholesterol in rats fed CO, and in phospholipid in rats fed HCO.

EFFECT OF PHYSICAL FORM, COMPOSITION AND LEVEL OF INTAKE OF DIET ON THE FATTY ACID COMPOSITION OF THE SHEEP CARCASS. A. Bensadoun and J. T. Reid (Dept. of Animal Husbandry, Cornell Univ., Ithaca, N.Y.). *J. Nutr.* 87, 239-44 (1965). The fatty acid composition of the mixed lipids was determined in the ground, whole carcasses of 45 sheep, each of which had ingested for 196 days one of 3 diets at one of 3 levels of intake. The sheep were of 2 ages (27 and 15 months at the time of slaughter). The proportions of the major fatty acids were not different for the sheep ingesting the 3 diets (chopped hay; pelleted, ground hay; and a pelleted mixture of 45% of corn meal and 55% of ground hay). There was, however, a significantly lower proportion of heptadecanoic and heptadecenoic acids in the carcasses of sheep fed the corn-containing diet than in those ingesting the other diets. Carcasses of sheep on the low level of intake contained 18% more of oleic acid than did those of sheep receiving the high level of intake. This significant increase ($P < 0.05$) was at the expense mainly of stearic acid. The carcass lipids of 27- and 15-month-old sheep adjusted to the same body weight had essentially the same fatty acid composition. The proportions of branched heptadecanoic acid were significantly greater ($P < 0.05$) in the 27-month-old sheep. In this study, body weight was the most important factor influencing the fatty-acid composition of the carcass.

BIOCHEMICAL AND HISTOCHEMICAL CHANGES IN AORTA OF CHICKS FED VEGETABLE OILS AND CHOLESTEROL. S. Banerjee, P. Narasimha Rao and S. K. Ghosh (Dept. of Physiology, S.M.S. Med. College, Jaipur, India). *Proc. Soc. Exp. Biol. Med.* 119, 1081-86 (1965). Portions of aorta of chicks fed coconut oil, sesame oil, mustard oil and hydrogenated groundnut oil, with or without simultaneous administration of cholesterol, for 8 weeks were analyzed for cholesterol, phospholipids, hexosamine and hydroxyproline. The distribution of elastic tissue, acid mucopolysaccharides and lipids in the aorta was also studied by histochemical methods. The severity or degree of the different changes was neither related to the plasma level of cholesterol nor to the saturation or unsaturation of the vegetable oils used.

EFFECT OF DIETARY LINOLEIC ACID, VITAMIN E AND ETHOXYQUIN ON FERTILITY OF MALE CHICKENS. G. H. Arscott, J. E. Parker and E. M. Dickinson (Dept. of Poultry Sci. and Veterinary Med., Oregon State Univ., Corvallis, Ore.). *J. Nutr.* 87, 63-68 (1965). Adult male chickens were fed diets low in linoleic acid, vitamin E and ethoxyquin or high in linoleic acid with or without added vitamin E or ethoxyquin for 25 weeks. Fertilizing capacity and sperm concentration of semen were adversely affected with males fed the high linoleic acid diet without vitamin E or ethoxyquin. The addition of vitamin E or ethoxyquin to the high linoleic acid diet overcame these adverse effects. The low linoleic acid diet without vitamin E and ethoxyquin had no adverse effect on fertilizing capacity and concentration of semen. No differences were evident for males fed any of the diets as far as semen volume, hatchability of fertile eggs, body or testes weights or feed consumption were concerned. During the 26th and 27th weeks increasing the number of sperm inseminated failed to improve fertility of the males fed the diet high in linoleic acid without vitamin E. The results indicate that diets high in linoleic acid but without vitamin E affect fertilizing capacity as well as the number of sperm produced.

EFFECTS OF NON-ESSENTIAL ACIDS ON ESSENTIAL FATTY ACID DEFICIENCY. R. B. Alfin-Slater, R. S. Morris, H. Hansen and J. F. Proctor (School of Public Health, Univ. of Calif., Los Angeles, Calif.). *J. Nutr.* 87, 168-72 (1965). The effects of the methyl esters of fatty acids of chain length C-4 through C-18, including oleic, elaidic, and linoleic, on essential fatty acid deficiency were studied in feeding tests with rats. Using

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depressed growth response as a criterion, all of the fatty acid esters except oleate and linoleate accentuated essential fatty acid deficiency, with the esters of fatty acids C-4 through C-10 and elaidate exhibiting a much greater effect than the longer-chain fatty acid esters. The highest mortality was observed in females fed caproate, caprylate and laurate and in males fed caprylate, laurate and stearate. When reproductive performance is judged, methyl esters of myristate, palmitate, stearate, oleate and linoleate improved the reproductive performance of female rats fed the fat-free diets and, therefore, partially alleviated the essential fatty acid deficiency. When cholesterol levels are used as criteria, supplementation with laurate and oleate resulted in an increase in the accumulation of hepatic cholesterol esters over that obtained when an unsupplemented fat-free diet was fed, indicating that laurate and oleate accentuate essential fatty acid deficiency in this respect. It is concluded that several metabolic pathways and physiological responses affected by essential fatty acid deficiency can be influenced by the concomitant presence of non-essential fatty acids. It is further concluded that evaluation of essential fatty acids status should involve more than one measurement.

IDENTIFICATION OF SOME LIPID PEROXIDES BY THIN-LAYER CHROMATOGRAPHY. K. Oette (The Rockefeller Inst., New York, N.Y.). *J. Lipid Res.* 6, 449-54 (1965). Solvent systems are described which permit class separations of various peroxidized lipids by thin-layer chromatography. This procedure has proved useful in biological studies and for testing the deterioration of lipids. Proof is given that the least polar peroxides of methyl esters, glycerides, and fatty acids are monoperoxides; it is then assumed that the least polar peroxides in the other tested groups (cholesterol, cholesterol esters, and phospholipids) are also monoperoxides. The more polar peroxides probably represent highly peroxidized and polymeric forms.

QUANTITATIVE ANALYSIS OF PHOSPHOLIPIDS AND PHOSPHOLIPID FATTY ACIDS FROM SILICA GEL THIN-LAYER CHROMATOGRAMS. F. Parker and N. F. Peterson (Div. of Dermatology, Dept. of Med., Univ. of Wash. School of Med., Seattle, Wash.). *J. Lipid Res.* 6, 455-60 (1965). An improved procedure for the quantitative assay of phospholipids separated by TLC is described, in which a specially washed Silica Gel H and a newly designed chromatography unit are employed. Analysis of phospholipid phosphorus and phospholipid fatty acids was accomplished in the presence of silica gel scraped directly from the chromatoplates. Recovery of phosphorus ranged from 96 to 100%. The washing of the Silica Gel H with chloroform-methanol-formic acid resulted in extremely low phosphorus blanks on the silica gel, and also removed impurities which were found to cause various peaks on GLC.

INFLUENCE OF SEVERAL PHYSICAL ACTIVITIES ON SERUM CHOLESTEROL CONCENTRATIONS IN YOUNG MEN. D. E. Campbell (Dept. of Physical and Health Education, Univ. of Texas, Austin, Texas). *J. Lipid Res.* 6, 478-80 (1965). An attempt has been made to determine the influence of several physical activities upon the serum cholesterol of 133 young adult males, who were randomly selected to participate in 10-week programs of cross-country running, golf, tennis, tumbling-gymnastics, wrestling, and weight training, and whose cholesterol values were compared with those of a control group. The findings as examined by analysis of variance suggest that different types of physical activity influence cholesterol concentrations in different degrees: subjects who participated in a vigorous and dynamic type of activity showed a significant decrease, whereas subjects who participated in a vigorous but static type of activity experienced no significant reduction during the experimental period.

LIPID COMPOSITION OF HEART MUSCLE HOMOGENATE. L. W. Wheelodon, Z. Schumert and D. A. Turner (Biochem. Res. Div., Sinai Hospital of Baltimore, Inc., Baltimore, Maryland). *J. Lipid Res.* 6, 481-89 (1965). Using cytochrome oxidase and esterase assays as a guide, mitochondria and microsomes were prepared from ox heart homogenate with about 25% cross-contamination of phospholipid. By the same criteria, the lipid complement of well washed myofibrils was essentially microsomal in origin. Approximately 60% of the phospholipid of

the whole homogenate was found to be associated with microsomes, about half of this being firmly bound to myofibrils.

Mitochondrial lipids were characterized by a higher degree of unsaturation of the free fatty acids and higher contents of cardioliolipin, cholesterol, and coenzyme Q than in microsomes, where choline-containing phospholipid, especially sphingomyelin, formed a greater proportion of the total phospholipid than in mitochondria. The outstanding difference was the virtual localization of ethanolamine in microsomes, in contrast to the equal distribution of choline plasmalogen between mitochondria and microsomes. Myofibril lipids resembled more closely microsomal than mitochondrial lipids, but contained in addition phosphatidyl serine and phosphoinositide, which were not detected in mitochondria and microsomes.

PHOSPHOLIPID COMPOSITION OF CHICK BRAIN DURING DEVELOPMENT. T. J. Siek and R. W. Newburgh (Dept. of Chem., Oregon State Univ., Corvallis, Oregon). *J. Lipid Res.* 6, 552-55 (1965). The phospholipid composition of chick embryo brain was examined over the interval 4-18 days of incubation. Phosphatidyl ethanolamine accounted for about 25% of the total lipid phosphorus during this interval, while phosphatidyl choline decreased from 70 to 50%. Sphingomyelin formed a relatively low fraction of the total phospholipid. The sum of inositol phosphatides and phosphatidyl serine was about 5%, with slight variation during the interval studied. A phospholipid resembling cardioliolipin was detected. Its content increased to 4% during this incubation period.

ORIGIN OF PHOSPHOLIPIDS IN THE CHICK EMBRYO DURING DEVELOPMENT. *Ibid.*, 556-64. Inorganic-P³², injected into yolks of eggs incubated to produce embryos of different ages, was incorporated into all phospholipid fractions in both whole chick embryo and embryo brain. Specific activity values compared between individual phospholipids of the same incubation age and in eggs injected at the same time did not vary more than twofold between one another. Biologically prepared phosphatidyl-P³² choline and phosphatidyl-P³² ethanolamine, when injected into yolks of eggs, gave a very different pattern of incorporation into embryo brain from that given by inorganic P³². When the labeled choline phosphatide was injected, a phosphatidyl choline fraction was isolated whose specific activity was 30-40 times greater than those of other phospholipid fractions. Phosphatidyl-P³² ethanolamine injection gave a qualitatively similar result. Glycerol-1-3-C¹⁴ and acetate-1-C¹⁴ were incorporated to a much lesser extent than inorganic-P³². The hypothesis is advanced that as the embryo develops, *de novo* synthesis from inorganic phosphate decreases and intact phospholipid is transferred from the yolk to the embryo and its organs.

LIPID COMPOSITION OF THE NORMAL HUMAN BRAIN: GRAY MATTER, WHITE MATTER, AND MYELIN. J. S. O'Brien and E. Lois Sampson (Depts. of Pathol. and Med., Univ. of Southern Cal. School of Med., Los Angeles, Cal.). *J. Lipid Res.* 6, 537-44 (1965). Gray matter, white matter, and myelin were isolated from the frontal lobes of humans aged 10 months, 6 yr., 9 yr., and 55 yr., and the lipid compositions of each were determined. Myelin had a much higher lipid content (78-81% of the dry weight) than white matter (49-66%) or gray matter (36-40%). Myelin contained much higher molar percentages of cerebroside and cerebroside sulfate, slightly higher molar percentages of cholesterol, and lower molar percentages of ethanolamine glycerophosphatides and choline glycerophosphatides than gray matter. The molar percentages of serine glycerophosphatides and sphingomyelin were about the same in each tissue. The aldehyde content of glycerophosphatides, expressed as molar percentage of the total lipoidal residues in each lipid, were as follows: ethanolamine glycerophosphatides from myelin 40-50%; ethanolamine glycerophosphatides from gray matter 21-27%; serine glycerophosphatides from myelin 21-36%; serine glycerophosphatides from gray matter 0.3-3.8%. Choline glycerophosphatides from either tissue contained only traces of aldehydes. The extra-myelin portion of white matter had a lipid composition that was very similar to that of myelin, but quite different from that of gray matter. Assuming a molecular weight of 28,000 for myelin protein(s), it was calculated that for each protein molecule in human myelin there are 186 lipid molecules, 111 of which are polar lipids and 75 of which consist of cholesterol. The over-all molar ratios of the polar lipids are phosphatidyl ethanolamine: serine glycerophosphatides: choline glycerophosphatides: sphingomyelin: cerebroside: cerebroside sulfate: seramide: uncharacterized lipids 25:9:20:9:29:7:3:9. It was calculated that the molar ratio of protein amino acids to polar lipids in human myelin is 2.38 to 1.

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FATTY ACID AND FATTY ALDEHYDE COMPOSITION OF THE MAJOR BRAIN LIPIDS IN NORMAL HUMAN GRAY MATTER, WHITE MATTER, AND MYELIN. *Ibid.*, 545-51. Gray matter, white matter, and myelin were isolated from the frontal lobes of the brains of humans aged 10 months, 6 yr, 9 yr, and 55 yr. The major lipids, including ethanolamine glycerophosphatides (EGP) serine glycerophosphatides (SGP), choline glycerophosphatides (CGP), sphingomyelin, cerebroside, cerebroside sulfate, and ceramide were isolated by column chromatography and their fatty acid and fatty aldehyde compositions were determined by gas-liquid chromatography. EGP and SGP from myelin had a fatty aldehyde composition which differed from that of EGP and SGP from gray matter; octadecenaldehydes were present in much higher proportions in these lipids from myelin than in those from gray matter. EGP and SGP also contained high proportions of 20- and 22-carbon polyunsaturated fatty acids, whereas CGP contained small proportions of these acids. Each glycerophosphatide from gray matter contained approximately 3- to 6-fold higher proportions of polyunsaturated fatty acids than did the same glycerophosphatide from myelin. Sphingomyelin, cerebroside, cerebroside sulfate, and ceramide also differed in their fatty acid compositions depending upon their tissue source; each sphingolipid from myelin in the younger subjects contained 5- to 9-fold higher proportions of long-chain fatty acids (C_{18} - C_{26}) than did the same sphingolipid from gray matter. The lipids from myelin in the baby (10 months) were very similar to those from myelin in the adult, both with respect to their content of polyunsaturated fatty acids and to their content of long-chain fatty acids. These findings suggest that myelin in the baby is "chemically mature" in its lipid composition at an early age.

FRACTIONATION OF NATURALLY OCCURRING LECITHINS ACCORDING TO DEGREE OF UNSATURATION BY THIN-LAYER CHROMATOGRAPHY. Gosta A. E. Arvidson (Dept. of Physiological Chem., Univ. of Lund, Lund, Sweden). *J. Lipid Res.* 6, 574-77 (1965). Intact, naturally occurring lecithins were fractionated according to degree of unsaturation by thin-layer chromatography on silica gel plates impregnated with silver nitrate and heated at 175-180C for several hours.

RAPID METHOD FOR DETERMINING FREE AND ESTERIFIED CHOLESTEROL IN PLASMA EXTRACTS. B. B. Zeitman (Physiology Branch, Environmental Bio. Div., NASA, Ames Res. Center, Moffett Field, Cal.). *J. Lipid Res.* 6, 578-80 (1965). A method has been developed for the quantitative measurement of free and esterified cholesterol in extracts of rat plasma after separation by thin-layer chromatography. As little as 0.2 ml of plasma may be used in the determinations.

EFFECT OF CHAIN LENGTH ON RATES OF UPTAKE OF FREE FATTY ACIDS DURING IN VITRO INCUBATIONS OF RAT ADIPOSE TISSUE. J. L. Knittle and J. Hirsch (The Rockefeller Univ., New York, N.Y.). *J. Lipid Res.* 6, 565-71 (1965). The incorporation into triglycerides of $1-C^{14}$ -labeled saturated free fatty acids (FFA) of different chain lengths (C_2 - C_{16}) was studied using incubation of rat epididymal fat pads *in vitro*. The results indicate that the rate at which adipose tissue incorporates FFA is dependent on chain length. With very short-chain acids a small amount is directly esterified into triglycerides, but the major portion is elongated and esterified. Analyses by means of factice chromatography showed that acids with a chain length of 10 or longer were esterified without elongation.

METABOLISM OF ARACHIDONIC ACID- $1-C^{14}$ IN THE RAT. R. H. Coots (The Procter & Gamble Co., Miami Valley Labs., Cin., Ohio). *J. Lipid Res.* 6, 494-97 (1965). The metabolism of arachidonic acid- $1-C^{14}$ has been studied in the rat. The acid was fed as a component of randomly rearranged soybean oil; its absorption was greater than 96%. The rate of catabolism of arachidonic acid was significantly lower than that previously seen for linoleic acid and other long-chain fatty acids. A marked tendency for arachidonic acid to be incorporated into phospholipids was observed. Arachidonic acid-containing phospholipids have been shown to have a slower turnover rate than linoleic and palmitic acid-containing phospholipids. It is proposed that this slower turnover rate is a reflection of the "essential"

character of arachidonic acid and is a rational explanation for the slower catabolic rate of this acid.

DEVELOPING RAT BRAIN: CHANGES IN CHOLESTEROL, GALACTOLIPIDS, AND THE INDIVIDUAL FATTY ACIDS OF GANGLIOSIDES AND GLYCEROPHOSPHATIDES. Y. Kishimoto, W. E. Davies and N. S. Radin (Mental Health Res. Ins., Univ. of Mich., Ann Arbor, Mich.). *J. Lipid Res.* 6, 532-36 (1965). Groups of brains from rats of various ages (7 to 275 days) were analyzed for their contents of galactolipids (cerebroside + cerebroside sulfate), cholesterol, and the individual fatty acids of the gangliosides and glycerophosphatides. Ganglioside stearate was found to accumulate at a steady pace during the first 20 days of life, then more slowly for at least 32 days more, then to decrease. Ganglioside arachidate, on the other hand, accumulated steadily throughout the period covered. The glycerophosphatide acids showed several inversions in the ratios between selected pairs of acids, the trend being toward increasing unsaturation and chain length. Higher contents of 22:6 and 20:4 acids than previously reported were found, presumably because of improved methods. The galactolipids and cholesterol were deposited at very similar rates after about 15 days, the molar ratio of the deposits (cholesterol/galactolipids) being about 2.2, the value found for purified myelin.

TURNOVER OF THE FATTY ACIDS OF RAT BRAIN GANGLIOSIDES, GLYCEROPHOSPHATIDES, CEREBROSIDES, AND SULFATIDES AS A FUNCTION OF AGE. Y. Kishimoto, W. E. Davies and N. S. Radin (Mental Health Res. Inst., Univ. of Mich., Ann Arbor, Mich.). *J. Lipid Res.* 6, 525-31 (1965). Rats of three different ages (7, 13, and 22 days) were given a single injection of acetate- $1-C^{14}$, then sacrificed at intervals (4 hr, 2 days, 10 days, and 30 days). The fatty acids of brain gangliosides, glycerophosphatides, cerebroside, and sulfatides were isolated and counted. Some were decarboxylated and the resultant carbon dioxide was counted. The data indicate that the palmitate of gangliosides, and glycerophosphatides is made *de novo* from acetate, but that the stearate of these lipids is made by elongation of palmitate. The palmitate used for elongation is not the freshly synthesized acid but rather the acid recently liberated by breakdown of the complex lipids. Lignocerate also appears to be made by elongation, but it is possible that the stearate of cerebroside is made *de novo*. No qualitative difference was seen in the turnover curves or modes of biosynthesis in the three rat groups. The turnover rates for ganglioside fatty acids were similar to those for the phosphoglycerides. Turnover data for sphingosine in cerebroside and sulfatides are presented.

NEW METHOD FOR PARTIAL DELIPIDIZATION OF SERUM LIPOPROTEINS. A. Gustafson (Cardiovascular Sect., Oklahoma Med. Res. Ins., Okla. City, Oklahoma). *J. Lipid Res.* 6, 512-17 (1965). A new method of partial delipidization of serum lipoproteins has been developed which removes neutral lipids preferentially and yields phospholipid-protein residues in soluble form without the use of detergents. Purified serum lipoprotein fractions were lyophilized in the presence of insoluble starch, which protected the proteins against damage during freezing, and the dry lipoproteins were partially delipidized by consecutive extractions with n-heptane. All neutral lipids and some phospholipids, mainly lecithin, were removed. Soluble phospholipid-protein residues were recovered by extraction with buffer solutions in yields ranging from 50 to 90%. The characteristic differences in the phospholipid-protein ratios between individual lipoprotein fractions are probably determined by the binding properties, dependent on primary structure, of the corresponding protein moieties.

TRANSPORT OF LYSOLECITHIN BY ALBUMIN IN HUMAN AND RAT PLASMA. S. Switzer and H. A. Eder (Dept. of Med., Albert Einstein College of Med., Bronx, N.Y.). *J. Lipid Res.* 6, 506-11 (1965). Lysolecithin comprises 9.6 and 21.5% of the plasma of man and rat, respectively. Ultracentrifugal and gel filtration studies showed that the major portion of the lysolecithin is not found together with the other phospholipids in the plasma lipoproteins. By zone electrophoresis, gel filtration, and ammonium sulfate fractionation, it was found that lysolecithin was consistently associated with albumin fractions. Immunoelectrophoretically homogeneous rat albumin was prepared. It contained 0.5 mg of lipid phosphorus per g of protein; 98.3% of this lipid was lysolecithin. It is concluded that lysolecithin is transported in plasma bound to albumin.

SERUM LIPOPROTEINS IN RATS WITH CARBON TETRACHLORIDE-INDUCED FATTY LIVER. B. Lombardi and G. Ugazio (Dept. of Path., Univ. of Pittsburgh School of Med., Pitts, Pa.). *J. Lipid Res.* 6, 498-505 (1965). After the administration of CCl_4 to male rats, liver triglycerides began to increase after a lag period of about 1 hr; the level of serum triglycerides fell

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sharply during the first 30 min of intoxication. Three classes of serum lipoproteins were isolated by flotation in the ultracentrifuge and their concentrations and chemical compositions were determined. Within 4 hr of the administration of CCl₄ the level of the very low density (VLD-) lipoproteins fell to 25% of that in the control rats. Smaller decreases in the levels of the other two classes of lipoproteins were evident. The serum concentration of all the components of the VLD-lipoproteins were reduced, but proportionally more lipids were bound to the protein moiety in the CCl₄-treated rats than in the controls. The concentrations of protein and triglycerides of the VLD-lipoproteins declined most steeply during the first hour of intoxication. The results are interpreted as further evidence that the fatty liver induced by CCl₄ is due to a block in the release of hepatic triglycerides to the plasma, the primary lesion being, very probably, inhibition of the synthesis of the protein moiety of serum lipoproteins.

OCCURRENCE OF METHYL ESTERS IN THE PANCREAS. E. Leikola, E. Nieminen, and E. Salomaa (Dept. of Pharmacy, Univ. of Helsinki, Helsinki, Finland). *J. Lipid Res.* 6, 490-93 (1965). By column chromatography two lipid fractions, x₁ and x₂, of which there are only traces in the lipids of other organs, have been isolated from the pancreatic lipids. The lipid fraction x₂, which forms a very considerable proportion of the total lipids of the pancreas, has been found to be composed of the methyl esters of lauric, myristic, tetradecenoic, palmitic, palmitoleic, stearic, oleic, linoleic, and arachidic acids. Fraction x₁ was not analyzed.

ISOLATION AND CHEMICAL CHARACTERIZATION OF PHOSPHATIDYL GLYCEROL FROM SPINACH LEAVES. F. Haverkate and L. L. M. Van Deenen (Dept. of Biochem., Lab. of Org. Chem., The State Univ., Utrecht, The Netherlands). *Biochim. Biophys. Acta* 106, 78-92 (1965). Pure phosphatidyl glycerol was obtained from spinach leaves after repeated chromatography on silica columns. Ascertainment of the configuration of the hydrolysis products formed by the action of phospholipases C (EC 3.1.4.3) and D (EC 3.1.4.4) demonstrated that this phospholipid is identical with 1,2-diacyl-glycerol-3-phosphoryl-1'-glycerol. Fatty acid analysis of several lipid fractions showed that the Δ⁵-trans-hexadecenoic acid, present in the leaves, is concentrated almost exclusively in phosphatidyl glycerol. Degradation experiments with phospholipase A (EC 3.1.1.4) showed that this acid is located preferentially at the 2-ester position. A subfractionation of phosphatidyl glycerol was accomplished by thin-layer chromatography on silica plates impregnated with silver nitrate. A breakdown of the two fractions obtained with phospholipase A allowed the recognition of several molecular species, and 1-linolenyl,2-Δ⁵-trans-hexadecenyl-glycerol-3-phosphoryl-1'-glycerol appeared to be the major species. The results were confirmed by hydrolysis of phosphatidyl glycerol with phospholipase C and separation on impregnated adsorbents of the diglycerides formed.

CHANGES EFFECTED BY DIETARY COCONUT OIL ON THE STRUCTURE OF TRIGLYCERIDES IN THE DIGESTIVE SYSTEM AND THE ADIPOSE TISSUE OF THE RAT. J. Clément, J. Bézard and E. Courel (Lab. de Physiol. Animale, Faculté des Sciences, Dijon, France). *Biochim. Biophys. Acta* 106, 25-33 (1965). In this work the structure of triglycerides has been investigated. The technique used is based upon the specificity of pancreatic lipase (EC 3.1.1.3). In coconut oil, most of the lauric acid is located in the β-position of the triglycerides. After administration of a diet containing 10 or 20% coconut oil to the rat, the fatty acid composition and the structure of the triglycerides were investigated in the intestinal lumen, the mucosa, the lymphatic chylomicrons and the perirenal adipose tissue. The following facts were established: (a) The proportion of lauric acid in the β position in the triglycerides decreases progressively from the intestinal lumen towards the adipose tissue, where it predominates in external positions, (b) In the glycerides of adipose tissue, lauric acid seems to substitute for the unsaturated acids, particularly for oleic acid: consideration of the distribution of lauric and oleic acids between the α, α' and β positions, shows their proportions in the β position to vary inversely. (c) Myristic acid, which is preferentially located in the external positions

of the triglyceride molecules of coconut oil, remains predominantly in that position in the triglycerides present at the various other sites studied.

SYNTHESIS AND ENZYMIC HYDROLYSIS OF AN O-ALANYL ESTER OF PHOSPHATIDYL GLYCEROL. P. P. M. Bonsel, G. H. De Haas and L. L. M. Van Deenen (Dept. of Biochem., Lab. of Org. Chem., Utrecht, The Netherlands). *Biochim. Biophys. Acta* 106, 93-105 (1965). A racemic o-alanyl ester of phosphatidyl glycerol, containing one saturated and one unsaturated fatty acid, was synthesized by a reaction between silver benzyl-(γ-oleoyl-β-palmitoyl)-DL-α-glycerol phosphate and DL-α-iodo-β-tert-butyl-γ-(N-tert-butoxycarbonyl)-DL-alanyl glycerol. The synthetic substance was hydrolysed by phospholipase A (EC 3.1.1.4), C (EC 3.1.4.3) and D. (EC 3.1.4.4). The results of the enzymic degradation and some other properties of this compound have been compared with those of amino acid derivatives of phosphatidyl glycerol from bacteria.

METABOLISM IN THE RAT OF CHYLE OBTAINED AFTER FEEDING HYDROGENATED COCONUT OIL LABELED WITH STEARIC ACID. J. Flovson, T. Olivecrona and P. Belfrage (Dept. of Physiol. Chem., Univ. of Lund, Lund, Sweden). *Biochim. Biophys. Acta* 106, 34-44 (1965). Chyle with a high content of saturated fatty acids, labeled with stearic acid-3,4-H³, was obtained from a thoracic duct cannulated rat fed hydrogenated coconut oil. The chyle was injected intravenously into carbohydrate re-fed rats and the blood and the tissue radioactivity studied. The rate of disappearance from the blood and the tissue distribution of the label was quite similar to that obtained in experiments with corn oil chyle. For a short time most of the label in the liver was in non-phospholipid stearic acid. Then increasing amounts appeared in the phospholipids, and label appeared in oleic acid in both phospholipids and non-phospholipids. By 160 min. the relative proportion of labeled oleic to labeled stearic acid was 4 in the liver non-phospholipids and 0.28 in the liver phospholipids. These figures are quite similar to those obtained in experiments with stearic acid administered as free fatty acid. It is concluded that the chyle triglycerides are removed by the liver without immediate breakdown. The trapped chyle is then the substrate for a system that liberates stearic acid in a form rendering its subsequent metabolism involving desaturation and distribution between ester fractions indistinguishable from that of stearic acid injected as the albumin complex.

METABOLISM OF LIPOPROTEIN LIPID IN THE ISOLATED PERFUSED RAT HEART. H. K. Delcher, M. Fried and J. C. Shipp (Dept. of Biochem. and Med., Univ. of Florida College of Med.). *Biochim. Biophys. Acta* 106, 10-18 (1965). The disappearance from the perfusate, localization within heart lipids, and oxidation of C¹⁴-labeled lipid from three ultracentrifugally separated lipoprotein fractions were studied in the isolated perfused rat heart. With d < 1.006 lipoprotein there was a net disappearance of triglyceride fatty acid from the perfusate. Heart lipids, increased free fatty acids and CO₂ accounted for all triglyceride fatty acid uptake. CO₂ formation and restoration of heart lipids in the perfused tissue accounted for 90% of the expected respiration. A net increase of free fatty acids in the perfusate suggested lipoprotein lipase activity either at or near the myocardial cell membrane, or lipase release into the perfusate. With d 1.006-1.070 lipoprotein, C¹⁴O₂ and C¹⁴-label in the heart lipids accounted for all of the triglyceride fatty acid uptake. The final concentration of triglycerides in the perfusate was very low and no net increase in free fatty acid in the buffer was observed. The amount of triglyceride fatty acid recovered in CO₂ was less than, and that in heart lipids equal to, that observed with d < 1.006 lipoprotein. A larger fraction of heart lipid was recovered as phospholipid. With d 1.070-1.210 lipoprotein, the lack of change in concentration or specific activity of phospholipid with perfusion suggested that phospholipids were not extracted, or metabolized by the perfused heart. The C¹⁴-labeled lipid extracted was primarily free fatty acid; the major non-oxidative fate of this free fatty acid was incorporation into tissue phospholipids.

CONVERSION OF GLUCOSE-1-C¹⁴ TO LIPID BY MACROPHAGES IN VITRO. A. J. Day and N. H. Fidge (Dept. of Human Physiol. and Pharmacol., Univ. of Adelaide, Adelaide, Australia). *Biochim. Biophys. Acta* 106, 19-24 (1965). Glucose-1-C¹⁴ was incubated *in vitro* with macrophages, obtained from the peritoneal cavity of rabbits, in order to investigate the uptake and incorporation of glucose into lipid by these cells. The macrophages were found to convert an appreciable amount of the labelled glucose to lipid. Fractionation of this lipid on silicic acid columns showed the label to be incorporated mainly into triglyceride and phospholipid, and to a lesser extent into cholesterol, mono- and diglyceride. Of the glucose-1-C¹⁴ converted to lipid, 94-

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96% was incorporated into the lipid glycerol moiety, only 4-6% being incorporated into the cholesterol and fatty acid. The incorporation of glucose-1-C¹⁴ into lipid varied from 17.4 to 33% of that incorporated into all cell components; in addition considerable oxidation of glucose to C¹⁴O₂ was brought about by the macrophages.

THE ENZYMATIC ACYLATION AND HYDROLYSIS OF LYSOLECITHIN. J. F. Erbland and G. V. Marinetti (Biochemistry Department, The Univ. of Rochester, School of Medicine and Dentistry, Rochester, N.Y.). *Biochim. Biophys. Acta* 106, 128-138 (1965). The present experiments show that lysolecithin is converted to lecithin by rat-liver preparations which are free from cell particles. In this process lysolecithin is esterified directly rather than being degraded to simpler compounds which are subsequently incorporated into lecithin. Two separate reactions are postulated whereby lysolecithin is converted to lecithin. These reactions are as follows: (1) 2 lysolecithin → lecithin + glycerophosphoryl-choline (2) lysolecithin + acyl-CoA → lecithin Reaction 1 is from a quantitative standpoint the more important reaction although Reaction 2 has a faster rate. The Michaelis-Menten plot of lecithin synthesis vs. lysolecithin concentration gave support for Reactions 1 and 2 since the rate curve was anomalous, being composed of two separate parts which gave K_m values of 0.08 mM (Reaction 2) and 3.3 mM (Reaction 1). The value of K_m obtained for lysolecithin hydrolysis was 1.03 mM. When lysolecithin-P³² was used as substrate, the synthesis of lecithin as measured by either P³²-incorporation or by chemical phosphorus was the same and either method could be used to quantitate lysolecithin esterification or lysolecithin hydrolysis. The studies given here show that the rate of metabolism of lysolecithin was essentially constant at pH values between 6.2 and 6.9 but at values greater than 6.9 there was suppression of both lecithin synthesis and lysolecithin hydrolysis. Iodoacetic acid failed to inhibit either lecithin synthesis or lysolecithin hydrolysis whereas HgCl₂ inhibited both. CN⁻ inhibited lecithin synthesis but not lysolecithin hydrolysis. Stearyl-glycollecithin was found to be inactive as a substrate for lysolecithinase in the rat-liver supernatant fluid but inhibited lysolecithin hydrolysis and lecithin synthesis 73 and 80% respectively. Transesterification has been shown to occur by the use of doubly-labeled lysolecithin.

THE METABOLISM OF LYSOLECITHIN IN RAT-LIVER PARTICULATE SYSTEMS. *Ibid.*, 139-144. The following reactions occur in the rat liver preparations studied: (1) lysolecithin → glycerophosphorylcholine + fatty acid, (2) lysolecithin + acyl-CoA → lecithin, (3) 2 lysolecithin → lecithin + glycerophosphorylcholine. In enzyme systems containing microsomes, lecithin synthesis from lysolecithin occurs primarily by Reaction 2 which is ATP and CoA dependent. In the particle-free cytoplasmic fluid Reaction 1 is predominant. This reaction is not influenced by added ATP or CoA. Triglyceride, diglyceride, phosphatidylethanolamine, inositol phosphatide and cholesterol esters do not act as acyl donors for the esterification of lysolecithin in rat-liver homogenates. The formation of lecithin in the microsomal system by Reaction 2 is about 6 times greater than the synthesis of lecithin in the cytoplasmic fluid by Reaction 3.

THE EFFECTS OF TRANS, TRANS-LINOLEATE UPON THE METABOLISM OF LINOLEATE AND LINOLENATE AND THE POSITIONAL DISTRIBUTION OF LINOLEATE ISOMERS IN LIVER LECITHIN. Z. Selinger and R. T. Holman (The Hormel Institute, Univ. of Minnesota, Austin, Minn.). *Biochim. Biophys. Acta* 106, 56-62 (1965). 9-trans,12-trans-Linoleate, when fed to the fat-deficient rat together with linoleate or linolenate, suppresses the levels of poly-unsaturated fatty acids arising from these precursors. Gas-liquid chromatography of the fatty acids of liver lecithin revealed the presence of *trans,trans*-linoleate. Unlike the naturally occurring *cis,cis* isomer, *trans,trans*-linoleate has a distribution pattern between the α- and β-positions in lecithin, which resembles that of saturated fatty acids; 80% of it was found in the α position.

METABOLISM OF RED-CELL LIPIDS. I. INCORPORATION IN VITRO OF FATTY ACIDS INTO PHOSPHOLIPIDS FROM MATURE ERYTHROCYTES. E. Mulder and L. L. M. Van Deenen (Dept. Biochem., Lab.

Org. Chem., The State Univ., Utrecht, The Netherlands). *Biochim. Biophys. Acta* 106, 106-117 (1965). Erythrocytes freed from leucocytes and reticulocytes were demonstrated to incorporate fatty acids into their phosphoglycerides. This ability was decreased in the order rat, rabbit, man, ox and sheep. Lysis of the cells caused an increase of the rate of incorporation thereby abolishing the differences between erythrocytes of different species. Addition of coenzyme A and adenosine 5'-triphosphate promoted the fatty acid uptake, particularly in the lysates. In rabbit erythrocytes linoleic, oleic and palmitic acid were incorporated to a greater extent than stearic, myristic and lauric acid. The unsaturated fatty acids were esterified predominantly at the 2-position, whereas palmitic acid was directed to the 1-position of lecithin. By contrast to L-α-glycerophosphate, lysolecithin stimulated the incorporation of linoleic acid into lecithin of lysates of rabbit erythrocytes. The possible relation between the fatty acid uptake *in vitro* and the fatty acid renewal of phosphoglycerides from circulating erythrocytes is discussed.

METABOLISM OF RED-CELL LIPIDS. II. CONVERSIONS OF LYSO-PHOSPHOGLYCERIDES. E. Mulder, J. W. O. Van Den Berg and L. L. M. Van Deenen (Dept. of Biochem., The State Univ., Utrecht, The Netherlands). *Biochim. Biophys. Acta* 106, 118-127 (1965). Lysolecithin is converted by lysed rabbit erythrocytes into lecithin and glycerylphosphorylcholine. The formation of lecithin by two reactions was demonstrated: by a transacylation requiring the addition of ATP and CoA, and by a dismutation of lysolecithin, not involving the incorporation of exogenous fatty acid but accompanied by the formation of glycerophosphorylcholine. Under conditions favorable for fatty acid incorporation the first-mentioned pathway was found to be predominant. In addition, lysolecithin appears to be degraded by a lysophospholipase (EC 3.1.1.5).

TRIGLYCERIDE BIOSYNTHESIS IN THE INTESTINAL MUCOSA. J. M. Johnston and G. A. Rao (Dept. of Biochem., Univ. of Texas, Southwest. Med. School, Dallas, Texas). *Biochim. Biophys. Acta* 106, 1-9 (1965). The chemical type of triglyceride synthesized by the enzymes of the hamster intestinal mucosa from monoglycerides and fatty acids have been examined employing AgNO₃-impregnated silica gel thin-layer plates. When 2-mono-palmitin and palmityl-CoA or oleyl-CoA or the combination of the CoA thioesters of the fatty acids were employed, a close-to-theoretical distribution of the fatty acids in the synthesized triglycerides was obtained. When the free fatty acids and an activating system were substituted for the CoA derivatives, a preferential utilization of palmitic acid was observed. The relationship of the reported findings to the overall absorption of triglyceride is discussed.

INTERRELATIONSHIPS BETWEEN THE MAJOR FATTY ACIDS PRESENT IN THE LECITHIN ISOLATED FROM THE PLASMA OF EXPERIMENTAL RABBITS. J. H. Moore, D. L. Williams and D. R. Westgarth (Nat'l Inst. for Res. in Dairying, Shinfield, Reading, Great Britain). *Biochim. Biophys. Acta* 106, 145-154 (1965). Six groups of rabbits were given diets for 36 weeks containing either 0.2, 0.3, 1.9, 4.9, 5.4 or 10.6% linoleic acid and the fatty acid compositions of the plasma total phospholipid fractions were then determined. Examination of the fatty acid compositions of the total phospholipid fractions revealed that there were significant positive rectilinear relationships between the concentrations of palmitic and oleic acids and between the concentrations of stearic and linoleic acids. Significant negative rectilinear relationships were found between the concentrations of palmitic and stearic acids and between the concentrations of oleic and linoleic acids. The total plasma phospholipid fractions obtained from each group of rabbits were pooled and six samples of pure lecithin were isolated from the pooled plasma phospholipids. The positional distribution of the various fatty acids in the pure lecithin samples was then determined. Significant positive rectilinear relationships were found between the concentration of palmitic acid in the α-position and the concentration of oleic acid in the β-position and between the concentration of stearic acid in the α-position and the concentration of linoleic acid in the β-position. It is suggested that the linoleic acid content of the diet exerts a direct effect on the composition of the fatty acids occupying the β-position and an indirect effect on the composition of the fatty acids occupying the α-position of the plasma lecithin.

PHOSPHOLIPIDS AND ACTIVE SODIUM TRANSPORT IN TOAD BLADDER. J. De Graeff, E. F. Dempsey, L. D. F. Lameyer and A. Leaf (Departments of Medicine, Harvard Medical School, and the Massachusetts General Hospital, Boston, Mass., and the Univ. Hospital, Leiden, The Netherlands). *Biochim. Biophys. Acta* 106, 155-170 (1965). A study has been made of the phospho-

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lipids of the urinary bladder of the toad, *Bufo marinus*. The phospholipids were found to contain some 22% of the total phosphate in this tissue. The major phospholipid fractions were identified chromatographically as phosphatidyl inositol, sphingomyelin, phosphatidyl choline, phosphatidyl ethanolamine, phosphatidyl serine and phosphatidic acid. The last named compound, although quantitatively one of the least of the phospholipids had the highest rate of incorporation of ^{32}P .

RADIOACTIVITY IN BLOOD AND LIVER PARTIAL GLYCERIDES, AND LIVER PHOSPHOLIPIDS AFTER INTRAVENOUS ADMINISTRATION TO CARBOHYDRATE-FED RATS OF CHYLE CONTAINING DOUBLE-LABELED TRIGLYCERIDES. P. Belfrage, J. Elovson and T. Olivecrona (Dept. Physiol. Chem., Univ. of Lund, Lund, Sweden). *Biochim. Biophys. Acta* 106, 45-55 (1965). A chromatographic method permitting fast and complete separation of all non-phospholipid fractions in rat blood and liver except for 1,2-diglycerides from cholesterol, and 1-from 2-monoglycerides is described. The level of radioactivity and the ratio C^{14}/H^3 were determined in partial glycerides and free fatty acids from blood and liver and in liver phospholipids at short time intervals after the intravenous injection into carbohydrate-fed rats of chyle-containing triglycerides labeled with glycerol- C^{14} and palmitic acid- H^3 . The ratios C^{14}/H^3 in blood partial glycerides showed that they were not formed by breakdown of chyle triglycerides in the circulating blood, but were probably original constituents of the chyle. The sum of H^3 radioactivity in partial glycerides and free fatty acids in the liver never exceeded one tenth of the total H^3 radioactivity in this organ. The ratios C^{14}/H^3 in the labeled liver partial glycerides indicated that these were mainly formed during the resynthesis of triglycerides and phospholipids. There was a progressive rise of fatty acid radioactivity in the liver phospholipids. Only a small amount of C^{14} -radioactivity was found in these—slightly more relatively in the phosphatidyl ethanolamine than in the phosphatidyl choline fraction.

UPTAKE OF FAT BY PHAGOCYTTIC CELLS. AN EXAMINATION OF THE ROLE OF PHAGOCYTOSIS I. RABBIT POLYMORPHONUCLEAR LEUKOCYTES. P. Elsbach (Dept. of Med., New York Univ. School of Med., NYC, N.Y.). *Biochim. Biophys. Acta* 98, 402-419 (1965). The object of this investigation was to examine the ability of polymorphonuclear leukocytes obtained from peritoneal exudates produced in rabbits to incorporate complex lipids *in vitro* and in particular, to assess the role of phagocytosis in the uptake of fat by this phagocytic cell. Uptake was studied of three kinds of isotopically labeled complex lipids: a) commercially obtained C^{14} -labeled triglycerides, b) biosynthetically prepared mixed phospholipids, c) C^{14} -labeled chylomicra collected by cannulation of the thoracic duct in humans fed radioactive palmitic acid. Radioactivity was found associated with the leukocytes during incubation with each of these labeled compounds. However, this association appeared largely independent of time of incubation and the metabolic activity of the cells. In addition the labeled fat was not broken down to an appreciable extent. These findings are consistent with the conclusion that the glycerol esters studied adhere to the surface of the leukocytes without gaining access to the metabolic pathways concerned with breakdown of lipids. A comparison was also made of uptake of the three complex lipid species by resting leukocytes and by cells actively engaged in phagocytosis of heat-killed bacteria. During phagocytosis there was also minimal or no metabolic utilization of the labeled lipid. In contrast to marked stimulation of O_2 uptake by leukocytes in the presence of bacteria, a suspension of chylomicra had no detectable influence on respiration. In view of the previous demonstration of avid incorporation of free fatty acids, these results indicate that incorporation of lipids by the leukocytes is selective and apparently restricted to free fatty acids. It is concluded that phagocytosis is of little or no quantitative importance in the uptake of fat by this phagocytic cell.

RABBIT ALVEOLAR MACROPHAGES. II. *Ibid.*, 420-431. In the accompanying communication it was shown that polymorphonuclear leukocytes do not incorporate glycerol esters to an appreciable extent. A similar study of uptake of lipid was carried

out with another phagocytic cell, the alveolar macrophage obtained from rabbit lungs. Alveolar macrophages incorporate C^{14} -labeled free fatty acids complexed to serum albumin into cell lipids. Inhibition of glycolysis markedly reduced incorporation of fatty acid, while interference with oxidative metabolism had little or no inhibitory effect. In contrast to the polymorphonuclear leukocytes, the macrophages also incorporate C^{14} -tripalmitate and C^{14} -labeled chylomicra. However, evidence was obtained indicating that C^{14} -phospholipids become associated with the cells by a non-metabolic process. Homogenates of macrophages were examined for the presence of lipid-splitting activity. High lipase activity was found with three different substrates. These activities manifested different pH optima. Phospholipase activity was also demonstrable as indicated by hydrolysis of lecithin and lysolecithin but was low compared to the lipase activity. Indirect evidence was obtained suggesting that lipolysis preceded uptake of glycerides. These results are discussed in the light of reports in the literature concerning uptake of intact glycerolesters by various tissues, presumably by a phagocytosis-like process. The hypothesis is presented that uptake of fat by cells is restricted to incorporation of free fatty acids. It is proposed that esterification takes place on or in the surface of the cell and that the apparent ability of a cell to incorporate glycerol esters depends upon the presence of lipid-splitting enzymes capable of acting near or on the surface membrane.

A DIETHER ANALOG OF PHOSPHATIDYL GLYCEROPHOSPHATE IN HALOBACTERIUM CUTIRUBRUM. M. Kater, L. S. Yengoyan and P. S. Sastry (Div. of Biosciences, Nat'l Res. Council, Ottawa, Canada). *Biochim. Biophys. Acta* 98, 252-268 (1965). The major phosphatide in the extremely halophilic bacterium, *Halobacterium cutirubrum*, has been isolated by a combination of solvent fractionation, precipitation through the barium salt, and final purification as the sodium salt. Analytical and degradative data showed the phosphatide to be a phosphatidyl glycerophosphate with two long-chain ether groups instead of fatty acid ester groups. Both long-chain groups were found to be identical and were shown by comparison of infrared, nuclear magnetic resonance, and mass spectra with those of dihydrophytyl derivatives to have the structure 3,7,11,16-tetramethylhexadecyl. The two long-chain groups are joined by ether linkages to the α,β -positions of L-glycerol. The structure proposed for the major phosphatide in *H. cutirubrum* is therefore: 2,3-di-O-(3',7',11',15'-tetramethylhexadecyl-glycerol-1-phosphoryl)-1''-(3'')-glyceryl-3''(1'')-phosphate, in the notation of Hirschmann. Possible biochemical and physiological functions of this unusual phosphatide in the halophile cell are discussed.

PREPARATION AND ANALYSIS OF LIPID EXTRACTS FROM MILK AND OTHER TISSUES. D. S. Galanos and V. M. Kapoulas (Lab. of Food Chem. of the Nat'l Univ. of Athens, Athens, Greece). *Biochim. Biophys. Acta* 98, 278-292 (1965). Simple and quick analytical methods for the determination of the phosphorus, nitrogen, long chain base-nitrogen, and carbohydrate contents of samples containing even very low quantities of polar lipids have been devised, in order to follow fractionation schemes of lipids from milk or other fat-rich tissues. With these analytical methods strong evidence was obtained indicating that a large proportion of the polar lipids present in milk consists of proteolipids, and mucolipids or other glycolipids. Aqueous washing of chloroform-methanol lipid extracts had destroying effects on these complex molecules, which are also partially broken down by lyophilization, as well as by acidic treatment, including silicic acid chromatography when the finest particles of the adsorbent are not completely removed. Possible explanations for these effects are given and bibliographic data presented supporting the statements. The commonly accepted procedures for manipulating lipids extracts are revised.

FRACTIONATION AND IDENTIFICATION OF MILK POLAR LIPIDS: TRIESTER GLYCOPHOSPHOLIPIDS. *Ibid.*, 293-312. A representative sample of the polar lipids of milk was isolated in a preparative scale from commercially spray dried milk using mild isolation procedures. By silicic acid chromatography at least 16 peaks of phospholipids (mainly glyceophospholipids), two peaks of cerebrosides, and three peaks of ganglioside-like mucolipids were identified. Strong evidence is presented supporting the view that the glyceophospholipid are phosphate triester derivatives, for which general formulas are tentatively proposed. As a first approximation, the composition of the polar lipids of milk is the following: cerebrosides, 8.8; polyglyceophosphatides, 1.6; phosphatidylethanolamine, 13.6; galactophosphatidylserine, 10.5; inositolphosphatides, 11.9, other "glycecephalins", 2.2; phosphatidylcholine, 9.1; galactophosphatidylcholine, 8.7; lactophosphatidylcholine, 8.9; other "leci-

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thins", 5.0; "sphingomyelin", 11.3, all expressed in moles/100 moles of lipid phosphorus. The presence of glycerol ether phospholipids in milk is also shown and their function discussed. It is suggested that all the glycerol ether lipids of milk are found in the phospholipid fraction. The idea that new, unknown lipids containing spermine or other similar bases, are probably present in milk is discussed. A large proportion of milk lipids was found in the form of proteolipids. New properties for proteolipids are deduced, and an attempt to explain their linkages between lipid and peptide, as well as the role of heavy metals in their formation is made.

MECHANISMS OF LIPID PEROXIDE FORMATION IN TISSUES. ROLE OF METALS AND HAEMATIN PROTEINS IN THE CATALYSTS OF THE OXIDATION OF UNSATURATED FATTY ACIDS. E. D. Willis (Dept. of Biochem., Med. College of St. Bartholomew's Hosp., London, Great Britain). *Biochim. Biophys. Acta* 98, 238-251 (1965). Oxidation of unsaturated fatty acids such as linoleic acid and linolenic acid is catalysed by metals at 37C in the pH range 4.5-7.5 with the formation of peroxides. Co^{2+} and Mn^{2+} are very active catalysts while Cu^{2+} , Fe^{3+} and Fe^{2+} are weakly active. The catalytic activity of Fe^{3+} can be strongly stimulated by addition of ascorbic acid or cysteine but both these substances delay oxidation catalysed by Co^{2+} or by haematin proteins. The pH optimum for oxidation catalysed by Fe^{3+} and ascorbic acid is 5.5, for Co^{2+} catalysis it is 6.5 but haemoglobin-catalysed oxidation is unaffected by pH over the range 4.5 to 8.0. o-Phenanthroline and 8-hydroxyquinoline powerfully inhibit Co^{2+} -catalysed oxidation but powerfully stimulate Fe^{3+} catalysed oxidation. Co^{2+} catalysed oxidation is unaffected by most amino acids but is strongly inhibited by histidine, by serum albumin and by some other proteins. It is considered that, *in vivo*, lipid peroxide formation is likely to be a result of oxidation of unsaturated lipids catalysed by Fe^{3+} and a reducing agent such as ascorbic acid or by haematin proteins.

A COMPARISON OF THE LIPID COMPOSITION OF BROWN ADIPOSE TISSUE FROM MALE AND FEMALE BATS (MYOTIS LUCIFUGUS) DURING HIBERNATING AND NON-HIBERNATING SEASONS. H. J. Wells, M. Makita, W. W. Wells and P. H. Krutzsch (Depts. of Biochem. and Anatomy, Univ. of Pittsburgh, School of Med., Pittsburgh, Pa.). *Biochim. Biophys. Acta* 98, 269-277 (1965). The lipids of the interscapular brown fat of bats, *Myotis lucifugus*, have been analyzed by silicic acid column chromatography and anion-exchange chromatography of the mild alkaline hydrolysis products of phosphatides. Brushite ($CaHPO_4$) chromatography has been introduced as an adjunct technique for the convenient separation of neutral from polar lipids and the removal of phosphatidylcholine from other phosphatides. Data from comparative studies of tissue obtained from male and female bats sacrificed during the hibernating and non-hibernating seasons are presented. The fatty acid composition of the cholesterol ester and triglyceride fraction is reported. The triglycerides, the major class of lipid, contain predominantly unsaturated fatty acids. Oleic acid is the principal individual fatty acid of this fraction. All brown fat preparations have high levels of cardiolipid presumably related to the rich content of mitochondria in brown adipose cells.

LINOLEIC ACID ACCUMULATION IN DEPENDENCE OF FEED FAT TYPE. K. H. Niesar (Inst. Physiol., Univ. Munich, Germany). *Fette Seifen Anstrichmittel* 67, 340-43 (1965). The linoleic acid accumulation in ten organs of fowls, calf and pig was measured after feeding low fat diets containing 0.2 or 1% linoleic acid based the total feed. The same measurements were made after feeding rations containing 15% trilaurin, tripalmitin or tristearin. Linoleic acid concentration in neutral fat and phospholipids decreased after trilaurin feeding. This was not due to an increased amount of higher fatty acids of the linoleic acid family.

INTERACTION OF INDIVIDUAL PHOSPHOLIPIDS BETWEEN RAT PLASMA AND ERYTHROCYTES IN VITRO. T. Sakagami, Osamu Minari and T. Orii (Dept. of Chem. and Biochem., Sapporo Medical College, Sapporo, Japan). *Biochim. Biophys. Acta* 98, 356-364 (1965). Lecithin, sphingomyelin and lysolecithin in erythrocytes were actively exchanged with these phospholipids in plasma. Individual phospholipids were not always exchanged at the same rate. The extent of exchange was greatest in lysolecithin. The exchanges of lecithin and sphingomyelin were less active, although the former was more active than the latter. The results obtained in experiments *in vitro* suggest that *in vivo* the phospholipids of mature circulating erythrocytes are metabolized predominantly through exchange with the plasma phospholipids rather than by the synthesis and breakdown *in situ*.

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GAS CHROMATOGRAPHIC SEPARATION AND DETERMINATION OF PENTAERYTHRITOL SYSTEM BY TRIMETHYLSILYL ETHER DERIVATIVES. R. R. Suchanec (Res. Center, Hercules Powder Co., Wilmington, Del.). *Anal. Chem.* 37, 1361-65 (1965). A new gas chromatographic method for analyzing the complete pentaerythritol system is presented. The method is based on the trimethylsilyl ether derivatives of these polyhydroxy compounds. This procedure is not only shorter and simpler than the best previous method but also makes possible a more detailed analysis of commercial grades of pentaerythritol. Using this method with an internal standard, mono-, di-, tri-, tetra-, and penta-pentaerythritol can be detected under easily obtainable conditions with a conventional instrument equipped with a thermal conductivity detector. Other components that have been definitely detected are pentaerythritol dicyclic diformal, pentaerythritol cyclic monoformal, and pentaerythrose. Additional peaks which were detected were tentatively assigned to the following derivatives: bis(pentaerythritol) monoformal, dipentaerythrose hemiacetal, pentaerythritol-dipentaerythritol monoformal, tris(pentaerythritol) diformal, and bis-dipentaerythritol) monoformal.

K. T. Holley Retires After 38 Years' Service

K. T. Holley (1945), head chemist of the Georgia Experiment Station, Experiment, Georgia, has retired after a career of service with the station that has extended over 38 years. His principal research in peanut curing and peanut utilization has resulted in an impressive total of 45 publications in this field. He is a fellow of the American Association for the Advancement of Science, and the American Institute of Chemists; he is a member of the American Chemical Society (Chairman of the Georgia Section), and the New York Academy of Science, in addition to his membership in AOCS.

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