

ABSTRACTS

R. A. REINERS, Editor. ABSTRACTORS: N. E. Bednarczyk, J. G. Endres, J. Iavicoli, K. Kitsuta, F. A. Kummerow, E. G. Perkins, T. H. Smouse, J. A. Thompson and R. W. Walker

• Fats and Oils

ISOLATION AND IDENTIFICATION OF A SERIES OF α,β -UNSATURATED ALDEHYDES FROM VALENCIA ORANGE PEEL OIL. M. G. Moshonas and E. D. Lund (Fruit and Vegetable Products Lab., U.S.D.A., Winter Haven, Fla. 33880). *J. Agr. Food Chem.* 17, 802-805 (1969). The isolation of a series of 7 α,β -unsaturated aldehydes, consisting of various self- and mixed-condensation products of octanal, nonanal, and decanal from orange peel oil is reported. The compounds possess the structure of dehydrated aldol condensation products, α,β -dialkyl acroleins. Five of the 7 compounds, not previously reported as constituents of natural products are: α -hexyl- β -heptyl, α -hexyl- β -octyl, α -heptyl- β -heptyl, α -octyl- β -heptyl, and α -hexyl- β -nonyl acrolein. The other two are probably α -octyl- β -octyl and α -heptyl- β -nonyl acrolein. Structure proof was accomplished by synthesis using the appropriate combinations of octanal, nonanal and decanal in a base-catalyzed aldol condensation; isolation of the various mixed and self-condensation products by gas-liquid chromatography; and proof of structure by ozonolysis and mass spectrometry. The possibility that these compounds are artifacts produced during the isolation procedure by condensation of the aldehydes is contradicted by the absence of appreciable amounts of the decanal self-condensation product.

SPLITLESS INJECTION ON CAPILLARY COLUMNS, PART I. THE BASIC TECHNIQUE; STEROID ANALYSIS AS AN EXAMPLE. K. Grob and G. Grob (Dept. of Organic Chem., Univ. of Zurich, Switzerland). *J. Chromatog. Sci.* 7, 584-586 (1969). A very simple procedure for splitless injection on capillary columns is discussed. In contrast to more sophisticated devices recently developed for the same purpose, the method described requires no additional equipment. The advantages of the method which broadens the use of capillary columns are discussed. Steroid analysis serves as an example of the applications.

ROLE OF LIPIDS IN FLAVORS. D. A. Forss (International Flavors & Fragrances, Union Beach, N.J. 07735). *J. Agr. Food Chem.* 17, 681-685 (1969). The best known contribution of lipids to flavor is as precursors. The compounds formed may be readily volatile and have appreciable odors—e.g., lower molecular weight aliphatic aldehydes, ketones and fatty acids. The intact lipid or low volatile breakdown products of lipids contribute to flavor largely through mouth stimulation. The taste of such compounds as well as their effects on the rheological properties of foods is discussed. In addition, lipids may modify the flavor of other compounds, particularly those of low polarity.

ANALYSIS OF CRUDE OIL CARBOXYLIC ACIDS AFTER CONVERSION TO THEIR CORRESPONDING HYDROCARBONS. W. K. Seifert, R. M. Teeter, W. G. Howells and M. J. R. Cantow (Chevron Oil Field Res. Co., Richmond 94802). *Anal. Chem.* 41, 1638-1647 (1969). A new and comprehensive approach has been developed for the identification of carboxylic acids in virgin crude oil. Knowledge of the structures of these acids is relevant to questions concerning the origin of petroleum and the origin of life. The identification involves initial conversion of acids, *via* the corresponding alcohols and their *p*-toluene sulfonate esters, to hydrocarbons. The hydrocarbons are separated by a combination of silica gel and gel permeation chromatography and identified by ultraviolet, infrared, and high resolution mass spectrometry. For the acid-hydrocarbon conversion, proof was obtained for the maintenance, on an average statistical basis, of most of the carbon skeletal structures of the original acids. The structure determination of the hydrocarbons and the retention of carbon skeleton during conversion permits identification of many classes of carboxylic acids not previously discovered in virgin crude oil.

ON THE NATURE OF HYDROCARBON CHAIN MOTIONS IN LIQUID CRYSTALS. M. C. Phillips, R. M. Williams, D. Chapman

(Molecular Biophysics Unit, Unilever, Frythe, Welwyn, England). *Chem. Phys. Lipids* 3, 234-244 (1969). Consideration of the thermodynamic parameters of the crystal to liquid crystal transition for lecithins and of the fusion of molecules containing hydrocarbon chains allows a quantitative estimate of the configurational freedom of hydrocarbon chains in the liquid crystalline states at the transition temperature. For lipids in general, it appears that the chain mobility is intermediate between that of β (or β') crystals and an *n*-alkane melt. The freedom of hydrocarbon chain motions is, however, greater in fully hydrated liquid crystalline systems than in anhydrous systems. These conclusions are pertinent to the much discussed question of hydrocarbon chain fluidity in thermotropic mesophases and certain biological systems.

A SYNTHESIS OF PROSTAGLANDIN F_{1a} AND RELATED SUBSTANCES. G. Just, C. Simonovitch, F. H. Lincoln, W. P. Schneider, U. Axen, G. B. Spero and J. E. Pike (Dept. of Chem., McGill Univ., Montreal). *J. Am. Chem. Soc.* 91, 5364-5371 (1969). The synthesis of prostaglandins F_{1a} , F_{1b} and a number of related products is described. The key step of the syntheses involves the acid-catalyzed opening and rearrangement of epoxybicyclo(3.1.0)hexanes.

A SPIN-LABELED LIPID FOR PROBING BIOLOGICAL MEMBRANES. A. S. Waggoner, T. J. Kingzett, S. Rottschaefer and O. H. Griffith (Dept. of Chem., Univ. of Oregon, Eugene 97403). *Chem. Phys. Lipids* 3, 245-253 (1969). A spin-labeled lipid has been synthesized in which the nitroxide is rigidly bound to a stearic acid chain at the 12-position. The spin-labeled lipid is sufficiently stable for general use in studying biological membranes and model membrane systems. Interactions of this new spin label with sodium dodecyl sulfate and lecithin micellar solutions have been investigated by electron spin resonance. The electron spin resonance spectra of the solubilized spin labeled lipid are sensitive to changes in micellar structure which affect the mobility of the label.

ANALYSIS OF VEGETABLE OIL MIXTURES BY GAS CHROMATOGRAPHY. M. Jernejeic and L. Premru (Chem. Inst. Boris Kidric, Ljubljana, Yugoslavia). *J. Oil Col. Chem. Assoc.* 52, 623-27 (1969). Ten samples of commercial vegetable oils were analyzed. Methyl esters of the fatty acids were prepared and their relative retention times determined on a column of 10 per cent LAC 446 when temperature programmed 150-200°C. The relative content of chosen components was determined by the area of their recorder peaks.

PROTECTED YEAST PROLONGS SHELF-LIFE. Food Eng. Staff (Chestnut and 56th Sts., Philadelphia, Pa. 19139). *Food Eng.* 40(2), 116-7 (1968). Emulsifiers of glycerol mono-stearate or fatty acid esters of sorbitan serve as antioxidant carriers to protect active dried yeast. Pre-mix bakery products now can have a shelf-life of a year.

DOES MULTI DUTY AT HIGH SPEED. R. Fritsche and L. E. Iverson (Miami Margarine Co., Cincinnati, Ohio). *Food Eng.* 40(3), 90-2 (1968). A new compact packager forms, fills, wraps, collates, packs ¼ pound margarine prints into cartons, and seals them at rates up to 300 per minute. It handles a wide range of product consistencies.

WHIPPED TOPPING, A COMPLEX EMULSION. W. G. Thalheimer (Atlas Chem. Ind., Inc., Wilmington, Del.). *Food Eng.* 40(5), 112-13 (1968). The role of vegetable fats, proteins, stabilizers, surfactants, and processing in the production of a desirable topping is discussed.

ENZYMES ENHANCE FLAVOR OF MILK SOLIDS. J. V. Ziemba (Sr. Assoc. Ed., Food Eng., Chestnut and 56th Sts., Philadelphia, Pa. 19139). *Food Eng.* 41(1), 105-10 (1969). Enzymatic hydrolyses of milk solids augment flavors of butterfat and milk proteins. Treated butterfat improves taste of butter-flavored products, and protein hydrolysates resemble beef stock.

INGREDIENT INNOVATIONS CHALLENGE PROCESSORS, PART I. L. Trauberman (Managing Ed., Food Eng., Chestnut and 56th Sts., Philadelphia, Pa. 19139). *Food Eng.* 41(7), 105-112 (1969). A survey of new and improved ingredients for reducing formulation costs without sacrificing product quality, for developing profitable new items, and for process simplification. Examples given include specialty fats for use in non-dairy whipped toppings and in one-step sponge cakes. Items for use in confections include a dry product containing 50 per

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cent vegetable fat and a new texture conditioner for soft candies.

THE ANALYSIS OF FATS CONTAINING CYCLOPROPENOID FATTY ACIDS, PART II: DETERMINATION WITH HYDROGEN BROMIDE. D. A. Rosie and G. G. Shone (Dept. of Chem., Kingston College of Tech., Kingston-upon-Thames). *Analyst* 94, 477-80 (1969). Methods for the determination of total fatty cyclopropenoids, by reaction with hydrogen bromide in a benzene medium, are described. Contrary to previous indications this reaction proceeds rapidly at room temperature. The methods presented obviate the undesirable need for the use of elevated temperatures in total cyclopropenoid determinations.

THE DETERMINATION OF POLYOXYETHYLENE EMULSIFIERS IN FOODS. J. M. Murphy and C. C. Scott (Ministry of Tech., Lab. of the Gov. Chem., Cornwall House, Stamford Street, London, S.E. 1). *Analyst* 94, 481-83 (1969). A preliminary investigation showed that polyoxyethylene emulsifiers contain substantial amounts of "free" polyethylene glycol. An improved method for determining these emulsifiers in foods is presented, in which the emulsifier is extracted with chloroform, "cleaned up" on an alumina column and analyzed by thin-layer chromatography with a modified Dragendorff reagent to spray the chromatogram. The method is at least ± 15 per cent accurate down to an emulsifier level of 0.01 per cent in fats and 0.001 per cent in baked foods and food mixes.

EMULSIFIERS. III. PERIODIC ACID ANALYSIS FOR 1-MONOGLYCERIDES. E. Distler and F. J. Baur (Procter & Gamble Co., Winton Hill Tech. Ctr., 6000 Center Hill Rd., Cincinnati, Ohio 45224). *J. Assoc. Offic. Anal. Chem.* 52, 602-607 (1969). Two collaborative studies were conducted to compare the official AOCS method, Cd 11-57, and a revised Miner procedure. The second study used known materials to obtain accuracy information. Studies showed that the revised Miner procedure is more accurate and more precise at low monoglyceride levels such as those encountered in commercial shortenings. It is recommended that the Miner procedure be adopted as official first action. It is further recommended that the joint AOAC-AOCS methods study of these two procedures be continued so that a single common method can be made official by both societies.

EXTRACTION OF PLANT STEROLS FROM ADULTERATED BUTTER OIL USING A DIGITONIN-IMPREGNATED CELITE COLUMN. D. E. La-Croix (Dairy Prod. Lab., Eastern Util. Res. and Dev. Div., Wash., D.C. 20250). *J. Assoc. Offic. Anal. Chem.* 52(3), 600-02 (1969). The phytosterol, β -sitosterol, can be used as an index of adulteration of butter oil with vegetable oils. A rapid, semimicro method for the analysis of plant sterols in butter oil adulterated with vegetable oils is described. The sterols are removed from the adulterated butter oil by passing the sample through a digitonin-impregnated Celite 545 column, eluted with dimethyl sulfoxide and analyzed for β -sitosterol by GLC, using a 3% JXR column. As little as 3 mg β -sitosterol/100 g butter oil can be readily detected by this method.

SYNTHETIC GLYCOSYLGlycerIDES IN BREADMAKING. Y. Pomeranz and H. P. Wehrli (Crops Res. Div., ARS, USDA, Kansas State Univ., Manhattan, Kansas 66502). *Food Technol.* 23, 1213-15 (1969). The addition of natural and synthetic polar lipids restored breadmaking potentialities of petroleum-ether-defatted wheat flour. Among the synthetic glycosylglycerides, cellobiosyl derivatives were more effective improvers than monogalactosyl derivatives. The optimum chain length for monogalactosyl glycerides was octanoic acid; for cellobiosylglycerides it was decanoic acid. Linoleyl derivatives were more effective than stearyl derivatives, though double bonds are not essential for the effect of glycolipids. Glycolipids, but not phospholipids, increased loaf volume of bread nutritionally-improved with 5% commercial soy flour.

REDUCTION OF UNSATURATED FATTY ACIDS AND THEIR ESTERS TO UNSATURATED FATTY ALCOHOLS BY SELECTIVE CATALYTIC

HIGH PRESSURE HYDROGENATION. III. METALLIC AND METAL OXIDE COPPER-CHROMIUM CATALYSTS. H. Bertsch, H. Reinheckel and K. Haage (Inst. for Fat. Chem., Ger. Acad. of Sci. of Berlin, Berlin-Adlershof, Ger.). *Fette Seifen Anstrichmittel* 71, 357-362 (1969). Influence of catalyst composition and reaction conditions on the high pressure reduction of unsaturated fatty acids and their esters is described for metallic and mixed metal oxide copper-chromium catalysts. From these investigations and from a comparison with the efficacy of other metals in hydrogenation, a hypothesis on the mechanism of the catalytic reaction in the reduction of carboxyl group and saturation of C-C double bonds is developed. The optimum reaction data of oleic acid and methyl oleate, which were used as test substances, are given.

CONTRIBUTION TOWARDS QUANTITATIVE DETERMINATION OF PHOSPHATIDES. U. Beiss (Inst. for Sugar Beet Res., Gottingen, Ger.). *Fette Seifen Anstrichmittel* 71, 363-365 (1969). A procedure is described by which the percentage distribution of individual phosphatides in a lipid extract of natural origin can be determined.

OXY ACIDS IN THERMALLY TREATED FATS. J. Wurziger and U. Solzer (Chem. and Nutr. Lab., Hygiene Inst., Freien and Hansestadt, Hamburg, W. Ger.). *Fette Seifen Anstrichmittel*, 71, 365-367 (1969). The usual values such as peroxide, aldehyde and epoxide values are insufficient for the assessment of the degree of oxidation of thermally treated fats. Characteristics such as acid value, coloration with alkali and oxy acid concentration offer a good insight into the degree of oxidation of a fat. According to this work, a good correlation exists between the alkali coloration and the oxy acid concentration so that the determination of either suffices. The measurement of alkali coloration is preferred since it is simple and rapid.

AUTOXIDATION OF SATURATED FATTY ACIDS. VI. ISOLATION AND IDENTIFICATION OF CLEAVAGE PRODUCTS FORMED BY THE OXIDATION OF LAURIC, MYRISTIC AND STEARIC ACIDS AND THEIR METHYL ESTERS. H. Thaler and H.-J. Kleinau (Inst. for Nutr. Chem. Technol., Univ. Braunschweig, W. Ger.). *Fette Seifen Anstrichmittel* 71, 261-264 (1969). In addition to previously reported primary oxidation products, other substances considered to be secondary products were also identified in the oxidation of saturated fatty acids and their methyl esters. A large number of methyl ketones, aldehydes and carboxylic acids were detected.

COMPARISON OF A FEW METHODS FOR THE DETERMINATION OF OXIRANE OXYGEN CONTENT OF OILS AND FATS. H. Karstens (Unilever Res. Lab., Hamburg, W. Ger.). *Fette Seifen Anstrichmittel* 71, 267-269 (1969). The accuracy and reproducibility of four methods were compared by multiple determinations of oxirane oxygen contents of 9,10-epoxystearic acid, alone or in conjunction with varnish grade linseed oil, or epoxidized oil, as well as of heat treated oxidized oil. Besides the direct method employing HBr/glacial acetic acid, three indirect procedures were tested. The differences among the above methods lie in titrating agents and end point indication. Especially with regard to attainable accuracy, the method employed involving potentiometric end point indication is distinctly advantageous as compared to colored indicators. None of the methods tested is applicable for the determination of oxirane oxygen content at a level $< 0.01\%$.

A HYPOTHESIS FOR CAROTENE DECOLORIZATION BY A FREE RADICAL MECHANISM IN FATTY ESTERS AND HYDROCARBONS. P. Venkatarao and K. T. Achaya (Reg. Res. Lab., Hyderabad-9, India). *Fette Seifen Anstrichmittel* 71, 270-272 (1969). Under oxidizing conditions, carotene is decolorized slowly in unsaturated and rapidly in saturated fatty esters. In unsaturated and saturated hydrocarbons, the reverse is observed. An explanation is offered based on the following hypotheses: a) That carotene is decolorized by hydrogen abstraction which upsets the entire conjugated chromophoric systems and b) that the degree of internal stability of the substrate-derived peroxy-free radicals, which are the hydrogen-abstracting agencies, is inversely related to the life of carotene in that substrate.

PHOSPHOLIPID CONSTITUTION OF EGYPTIAN VEGETABLE OILS. I. SAFFLOWER, GROUNDNUT AND GHUFA OILS. F. Osman, A. E. Ashour and A. M. Gad (Fats and Oils Dept., Nat. Res. Center, Dokki, Cairo, Egypt, (UAR)). *Fette Seifen Anstrichmittel* 71, 262-266 (1969). The phosphatidyl components of

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three Egyptian vegetable oils, namely safflower, groundnut and chufa, were identified using thin-layer chromatography (TLC). The fatty acids of the total phosphatides as well as those of lecithin and cephalin fractions of each sample were determined by gas-liquid chromatography.

COMPARATIVE STUDIES ON THE LIPIDS IN HUMAN TISSUES, ORGANS AND SERUM AS WELL AS IN FEMALE MILK. R. Tarjan, M. Kramer and K. Szotyori (Inst. for Nutr. Res., Budapest, Hungary). *Fette Seifen Anstrichmittel* 71, 272-275 (1969). The fatty acid composition of the total lipids, as well as the individual lipid fractions of the fat tissues, heart, liver and brain in 31 cases where death occurred due to accident was determined. The lipid composition of serum and female milk was also determined.

THERMAL OXIDATION AND THE CHEMISTRY OF FRYING FATS. J. P. Freeman (Unilever Res. Lab., Colworth, Welwyn, Herts, U.K.). *Food Process. Marketing* 38, 303-306 (1969). The chemistry associated with the oxidation of frying fats is reviewed.

TRIGLYCERIDE STRUCTURE OF HUMAN MILK FAT. W. C. Breckenridge, L. Marai and A. Kuksis (Dept. of Biochem. and Banting and Best Dept. of Med. Res., Univ. Toronto, Toronto, Ontario, Canada). *Can. J. Biochem.* 47, 761-769 (1969). The triglycerides of the fat globules of human milk were resolved by thin-layer chromatography on silica gel impregnated with silver nitrate. The chemical structure of the glycerides was determined by gas chromatography. The major triglycerides of human milk fat contained 48-54 acyl carbons which were made up of C₁₂-C₂₈ acids of various degrees of unsaturation. The fully saturated triglycerides (6-8%) contained mainly *sn*-glycerol 1-stearate 2-palmitate esterified at position 3 with C₁₄-C₁₈ acids. The monoenoic triglycerides (25-27%) were comprised mainly of *sn*-glycerol 1-oleate 2-palmitate esterified at position 3 with C₁₂-C₁₈ acids, as well as their racemates. The dienes (24-33%) were made up largely of *sn*-glycerol 1-stearate 2,3-dioleate and 2-palmitate 1,3-dioleate. The trienes (18-20%) contained mostly *sn*-glycerol 1,2,3-trioleate and 1-oleate 2-palmitate 3-linoleate. The tetraenes (7-21%) were identified as mainly *sn*-glycerol 1,2-diolate 3-linoleate and 1-palmitate 2,3-dilinoleate. Arachidonic acid was found in the polyene fraction (6%) in the 2-position, while the linolenic acid was preferentially associated with position 3 of the tetraenes.

ADDITION OF HYPOIODOUS ACID TO UNSATURATED FATTY ACIDS UNDER THE CONDITIONS EMPLOYED IN THE RAPID METHOD OF MARGOSCHES FOR THE DETERMINATION OF THE IODINE VALUE OF FATS. II. ERUCIC, BRASSIDIC, PETROSELINIC AND PETROSELALDIC ACIDS. D. Rankoff and B. Ivanova (Inst. for Organic Chem, Bulgarian Acad. of Sci., Sofia, Bulgaria). *Fette Seifen*

Anstrichmittel 71, 209-211 (1969). The addition of hypoiodous acid to the double bonds of erucic, brassidic, petroselinic and petroselaidic acids was investigated under the conditions employed in the quick procedure of Margosches for the determination of iodine value of fats. In all the cases, the reaction does not proceed stereospecifically, which leads to the formation of both the diastereomeric iodohydroxy derivatives. The configuration of these compounds was determined by conversion into compounds of known structure. In the conversion of iodohydroxy derivatives to the corresponding hydroxy-acetoxy compound employing silver- or sodium acetate in acetic acid, the corresponding keto acid is formed as well.

REACTION OF LONG CHAIN EPOXIDES WITH HYDROGEN SULFIDE. W. Umbach, R. Mehren and W. Stein (Basic Res. Lab., Henkel and Cie, Dusseldorf, W. Ger.). *Fette Seifen Anstrichmittel* 71, 199-203 (1969). Base catalyzed reactions of hydrogen sulfide with long-chain terminal or internal epoxides for the preparation of hydroxyalkyl mercaptans and/or *bis*-(hydroxyalkyl)-sulfides are reported. The influence of temperature, pressure, solvent, and nature and concentration of catalyst on the composition of the product is thoroughly examined.

THERMAL DECOMPOSITION OF SOME ACETOXYOCTADECENOATES AND ACETOXYOCTADECADIENOATES. Y. Toyama and K. Takeoka (Dept. Applied Chem., Fac. of Eng., Toyo Univ., Kawagoe-shi, Saitama-ken/Japan). *Fette Seifen Anstrichmittel* 71, 195-199 (1969). The rates of the thermal decomposition (dehydroacetoxylation) and the activation energies of the following three samples have been determined: a) acetylated methyl ricinoleate; b) methyl ester mixture of vicinally unsaturated acetoxyoctadecenoates prepared by reacting methyl oleate and mercuric acetate in acetic acid; c) methyl ester mixture of vicinally unsaturated acetoxyoctadecadienoates prepared by reacting methyl linoleate and mercuric acetate in acetic acid. The thermal decomposition is shown to be a first-order reaction.

QUANTITATIVE DETERMINATION OF NONPOLAR DIMERIC FATTY ACIDS IN FATS AND OILS. G. Billek and O. Heisz (Unilever Res. Lab., Hamburg, W. Ger.). *Fette Seifen Anstrichmittel* 71, 189-195 (1969). Fatty acids, obtained by saponification of fats followed by removal of unsaponifiables and petroleum ether-insoluble oxidized fatty acids, were treated to give urea-adducts. The mixture of fatty acids which did not form adducts was esterified and separated by thin-layer chromatography. It was shown by mass-spectrometry that the methyl esters of nonpolar dimeric fatty acids occupied a definite zone in the chromatogram. The concentration of methyl esters of nonpolar dimeric fatty acids can be estimated by charring the chromatogram under standardized conditions and quantitating the zone by densitometry. A test substance is co-

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chromatographed for comparison. The relative standard deviation in the chromatography and densitometric determination was approximately 10%. The lower limit of detection was at 0.005% of the methyl ester of nonpolar dimeric fatty acid. Using dimeric fatty acids labelled with radioactive carbon, it was proved that approximately 90% of the nonpolar dimeric fatty acids can be detected by this method.

• Biochemistry and Nutrition

UPTAKE OF GLUCOSE-1-¹⁴C BY PULLULARIA PULLULANS. E. Merdinger (Biochem. Res. Lab., Chem. Dept., Roosevelt Univ., Chicago, Ill. 60605). *J. Bacteriol.* 98, 1021-1025 (1969). Of the total glucose-1-¹⁴C activity added to the culture medium, 24.8% appeared in the neutral lipid fraction and 2.1% in the phospholipid fraction. The greatest amount of the activity (16.5%) was found in the free sterols. Among the phospholipids the largest amount of activity (1.1%) appeared in phosphatidylserine and phosphatidylethanolamine. The second largest amount of the total ¹⁴C was found in trehalose (9.6%) followed by carbon dioxide (7.3%).

PHOSPHOLIPID METABOLISM IN FERROBACILLUS FERROOXIDANS. S. A. Short, D. C. White and M. I. H. Aleem (Dept. Biochem., Univ. of Kentucky, Lexington, Ky. 40506). *J. Bacteriol.* 99, 142-150 (1969). In the chemoautotroph *Ferrobacillus ferrooxidans*, fatty acids represent 2% of the cell dry weight and 88% of the lipid fraction; 25% of the fatty acids of the latter fraction are associated with the diacyl phospholipids. The phospholipids were identified as phosphatidylmonomethylethanolamine (42%), phosphatidylglycerol (23%), phosphatidylethanolamine (20%), cardiolipin (13%), phosphatidylcholine (1.5%) and phosphatidyl dimethylethanolamine (1%).

EVIDENCE OF HYDROGENATION OF CIS-2 DOUBLE BONDING ON A WAY OF β -OXIDATION BY ESCHERICHIA COLI. M. Uchiyama, M. Yonaha and M. Mizugaki (Inst. of Pharmacy, Tohoku Univ. of School of Med., Sendai, India). *J. Biochem. (Tokyo)* 65, 977-8 (1969). Investigation using 13-hydroxyundecanoic acid as substrate suggested that the 5-hydroxy acid derived from homoricinoleic acid and accumulated in the medium was not the *cis*-2 enoic acid but the corresponding saturated acid. The 5-hydroxy acid derived from either homoricinoleic acid or 13-hydroxyundecanoic acid showed exactly the same retention time by gas chromatographical analysis with diethyleneglycol succinate as adsorbent. To confirm the hydrogenation of the *cis*-2 double bonding, 5-hydroxyundec-*cis*-2-enoic acid- δ -lactone was chemically synthesized by the method described earlier. Chemical oxidation of the synthesized δ -lactone gave 3-hydroxyundecanoic acid showing the α,β -unsaturation in the δ -lactone. The hydrogenation of 5-hydroxyundec-*cis*-2-enoic acid to 5-hydroxyundecanoic acid by growing the resting cells of *E. coli* proceeded efficiently. The bioreduction takes place specifically on *cis*- α,β -double bonding of hydroxy acid.

THE MECHANISM OF HEPARIN STIMULATION OF RAT ADIPOCYTE LIPOPROTEIN LIPASE. R. L. Patten and C. H. Hollenberg (McGill Univ. Med. Clinic, The Montreal Gen. Hosp., Montreal, Quebec, Canada). *J. Lipid Res.* 10, 374-382 (1969). Free fat cells and stromal-vascular cells were prepared from rat adipose tissue by incubation with collagenase. $\text{NH}_4\text{OH}-\text{NH}_4\text{Cl}$ extracts of acetone-ether powders prepared from fat cells contained lipoprotein lipase activity but extracts of stromal-vascular cells did not. Intact fat cells released lipoprotein lipase activity into incubation medium, but intact stromal-vascular cells did not. The lipoprotein lipase activity of the medium was increased when fat cells were incubated with heparin, and this was accompanied by a corresponding decrease in the activity of subsequently prepared fat cell extracts. Heparin did not release lipoprotein lipase activity from stromal-vascular cells. The lipoprotein lipase activity of $\text{NH}_4\text{OH}-\text{NH}_4\text{Cl}$ extracts of fat cell acetone powders is increased by the presence of heparin during the assay. This increase is not due to preservation of enzyme activity, but to increased binding of lipoprotein lipase to chylomicrons. Protamine sulfate and sodium chloride have little effect on the binding of lipoprotein lipase to chylomicrons, but they inhibit enzyme activity after binding to substrate has occurred. These inhibitors do, however, inhibit the stimulatory effect of heparin on enzyme-substrate binding.

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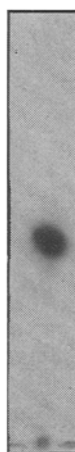
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Left: Purity check A of phosphatidyl glycerol U-C¹⁴. Solvent system: Chloroform/methanol/water, 70/25/4 by volume. Adsorbent: ADSORBOSIL-5.



Right: Purity check B of phosphatidyl glycerol U-C¹⁴. Solvent system: Chloroform/methanol/ammonium hydroxide, 60/30/5 by volume. Adsorbent: ADSORBOSIL-5.

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DIFFERENTIATION OF PHOSPHOLIPASES A IN MITOCHONDRIA AND LYSOSOMES OF RAT LIVER. M. Waite, G. L. Scherphof, F. M. G. Boshouwers and L. L. M. Van Deenen. (The Bowman Gray School of Med. of Wake Forest Univ., Winston-Salem, North Carolina 27103). *J. Lipid Res.* 10, 411-420 (1969). Highly purified mitochondria from rat liver contain a phospholipase A that catalyzes removal of 2-fatty acids, with a pH optimum above pH 8.0. Lysosomal preparations appeared to have two phospholipases A associated with them, one with a pH optimum at about pH 4.0, the second between pH 6.0 and 7.0. Mitochondrial phospholipase A hydrolyzed exogenous phospholipid as fast as or faster than endogenous phospholipid. The difference in specific radioactivity of ¹⁴C-ethanolamine-labeled endogenous mitochondrial phospholipid before and after incubation indicates that a fraction of mitochondrial phosphatidyl ethanolamine is hydrolyzed more rapidly than the mitochondrial phospholipids as a whole.

EARLY EFFECTS OF FEEDING EXCESS VITAMIN A: MECHANISM OF FATTY LIVER PRODUCTION IN RATS. V. N. Singh, M. Singh and T. A. Venkatasubramanian (Dept. of Biochem., Vallabhbhai Patel Chest Inst., Univ. of Delhi, Delhi-7, India). *J. Lipid Res.* 10, 395-401 (1969). Oral administration of vitamin A (30,000 IU daily for 2 days) to young rats caused a marked increase in hepatic glycogen, cholesterol and glycerides, while hepatic phospholipid content remained almost unaltered. It was found that more glucose-¹⁴C was incorporated into liver lipids in vitamin A-fed rats, where incorporation of glucose-¹⁴C and DL-glycine-¹⁴C into the liver protein remained unaltered. The increase in glucose-¹⁴C incorporation was confined to the glyceride-glycerol portion of the lipids; incorporation into liver fatty acids was inhibited. Plasma free fatty acid concentrations were elevated. It is postulated that in the vitamin A-fed rats, increased accumulation of lipids in the liver is caused by a stimulation of fatty acid mobilization from adipose tissue and enhanced formation of glycerophosphate through glycolysis, with consequent increase in the glyceride synthesis in the liver.

TRIGLYCERIDE FORMATION AND HYDROLYSIS BY TOAD BLADDER EPITHELIUM. A. A. Rosenbloom and P. Elsbach (Dept. of Med., New York Univ. School of Med., New York 10016). *J. Lipid Res.* 10, 406-410 (1969). Triglycerides of toad bladder epithelium have been labeled *in vitro* with either palmitate-¹⁴C or linoleate-¹⁴C, during incubation of bladders that had been cut in halves. Hydrolysis with pancreatic lipase of triglycerides labeled in this fashion revealed that palmitate-¹⁴C appeared predominantly in the 1- and 3-position, whereas half of linoleate-¹⁴C was located in the 2-position. The hydrolysis of palmitate-¹⁴C or linoleate-¹⁴C labeled triglycerides was examined in homogenates of isolated bladder mucosal cells. Lipase activity was evident from pH 3.5 to 8.0, but clearly greatest at pH 4.5. Below pH 6.0 the products of hydrolysis were fatty acid and monoglyceride and the 1- (or 3-) position was preferentially attacked; above pH 6.0 complete deacylation occurred. Acid-optimum hydrolysis of triglycerides with production of monoglycerides was linear for about 30 min. After 2 hr most of the labeled triglycerides were hydrolyzed. Repeated freezing and thawing of the homogenate enhanced lipase activity. Added Ca⁺⁺, previously shown to be required for phospholipase A activity in toad bladder, had no effect on hydrolysis of triglycerides.

THE RAPID DETERMINATION OF DIAZINON AND ITS OXYGEN ANALOGUE IN ANIMAL TISSUES BY GAS CHROMATOGRAPHY. A. F. Machin and M. P. Quick (Ministry of Agr., Fisheries and Food, Central Vet. Lab., New Haw, Weybridge, Surrey). *Analyst* 94, 221-25 (1969). A rapid method for the simultaneous gas-chromatographic determination of diazinon and its oxygen analogue (diazoxon) in blood, fat, liver, muscle and brain is described. The use of a selective thermionic phosphorus detector makes clean-up unnecessary. Preparation of the sample before injection consists in trituration with sand and sodium sulphate, elution with methanol or ether and concentration. Quantitative measurements are made by comparison with an internal standard. The method is satisfactory for the determination of 0.05 parts per million of diazinon and 0.2 parts per million of diazoxon in a 0.1 gram sample.

(Continued on page 682A)

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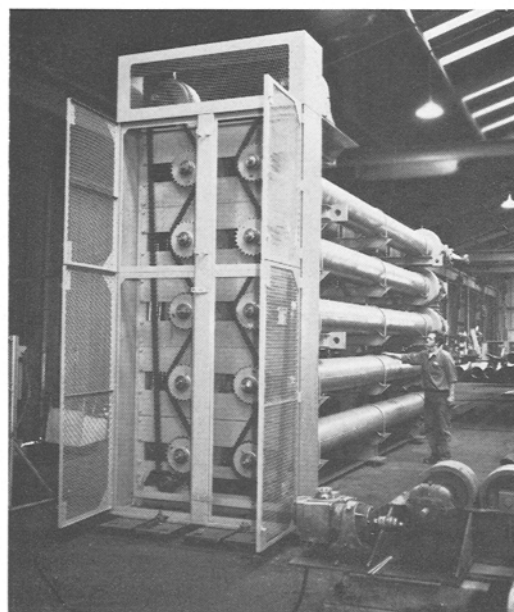
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• Local Section News . . .

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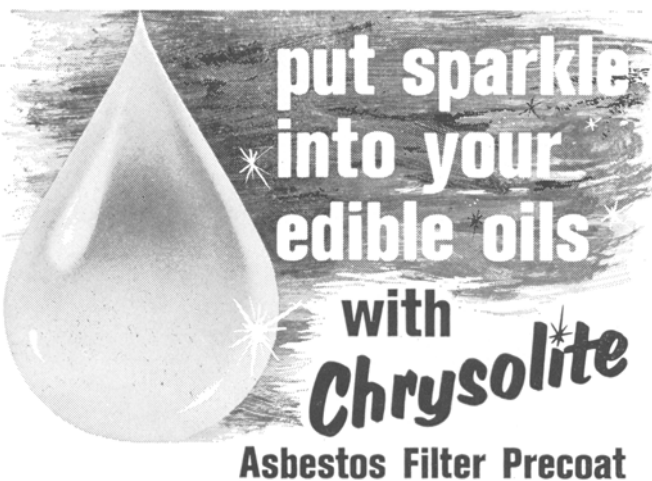
was suggested that the time-sharing computer as now available to even very small organizations is an ideal and inexpensive tool for this kind of calculation.

The after-dinner speaker, Art Apostol from General Electric Corp., was introduced by Program Chairman, R. Anderson of Swift and Co. Mr. Apostol's talk was on "In Process Control Computer Applications." The speaker traced the evaluation of the modern day computer, beginning with the ancient abacus. The first real computer contained 18,000 vacuum tubes and was built at the University of Illinois. This computer was followed by a number of improved versions. The first modern-day computer called UNIVAC, was produced in 1951. Today we are using "Fourth Generation" computers which are 100-1,000 times faster than those in use eight years ago.

After presenting the audience with a list of definitions on computer terminology, Mr. Apostol discussed the different types of software and hardware along with their industrial applications for processing control. The meeting was adjourned after a question and answer period.

A very happy and healthy holiday season to all. We hope to see you again at our next meeting, January 21, 1970. Our featured after-dinner speaker will be Donald C. Malins who is associated with the Food Service Pioneer Research Laboratory of the U.S. Fish and Wildlife Service. The topic of his talk will be "Lipids—An Oceanic Resource."

Members of the NCS wishing to contribute local section news items should contact Louis P. Goodman, Publicity Chairman, Kraftco Corp., Glenview, Illinois 60025.



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ABSTRACTS: BIOCHEMISTRY AND NUTRITION

(Continued from page 677A)

ASSAY OF ACYL-CoA:MONOGLYCERIDE ACYLTRANSFERASE FROM RAT SMALL INTESTINE USING CONTINUOUS RECORDING SPECTROPHOTOMETRY. J. B. Rodgers, Jr. (Dept. of Med. (Gastroenterology), Albany Med. College, Albany, N.Y.). *J. Lipid Res.* 10, 427-432 (1969). Acyl-CoA:monoglyceride acyltransferase in microsomal preparation from the small intestine of the fat has been measured by means of continuous recording spectrophotometry. The reaction of 5,5'-dithiobis (2-nitrobenzoic acid) with CoA has been employed for this assay and optimal conditions for the reaction have been defined. One of the substrates, palmitoyl-CoA, inhibits the reaction even in modest concentrations. This inhibition is largely prevented by the addition of bovine serum albumin to the incubation medium.

BACTERIAL 7-DEHYDROXYLATION OF CHOLIC ACID AND ALLOCHOLIC ACID. V. Bokkenheuser, T. Hoshita and E. H. Mosbach (Dept. of Microbiol., St. Luke's Hosp. Center, New York, N.Y.). *J. Lipid Res.* 10, 421-426 (1969). An obligate anaerobic organism capable of dehydroxylating cholic acid to deoxycholic acid and allocholic acid to allodeoxycholic acid was isolated from feces of the rabbit. It was a member of the bacteroides group (Gramvariable, nonsporulating anaerobes). The growth of the organism was inhibited by neomycin, 10-20 µg/ml. The existence of this organism affords a satisfactory explanation for the development of gallstones in the cholestanol-fed rabbit and for their absence in rabbits simultaneously treated with neomycin.

NEW TECHNIQUE FOR ENZYMIC HYDROLYSIS OF GLYCOSPHINGOLIPIDS. G. Dawson and C. C. Sweeley (Dept. of Biochem., Mich. State Univ., E. Lansing, Mich.). *J. Lipid Res.* 10, 402-405 (1969). A method is described for the study of glycosyl ceramide glycosyl hydrolases. Problems arising from the limited solubility of glycosyl ceramides in aqueous media were overcome by coating the substrate on a filter paper disc that had been treated with phosphatidyl choline. A comparison between the disc method and conventional dispersion of the substrate by detergent was made with two enzymes, galactosylgalactosyl-glucosyl ceramide galactosyl hydrolase (trihexosyl ceramide galactosyl hydrolase) from lysosomes of human and rat small intestine and human spleen, and D-galactose oxidase. In both cases enzymatic activity was greater with the paper disc method than it was with substrates dispersed by detergents. The galactose liberated by the glycosyl hydrolase was determined as the trimethylsilyl derivative of the free sugar by gas-liquid chromatography.

METABOLISM OF CHOLESTEROL IN THE TISSUES AND BLOOD OF THE CHICK EMBRYO. W. E. Connor, R. Johnston and D. S. Lin (Cardiovascular Res. Labs., Dept. of Internal Med., Univ. of Iowa College of Med., Iowa City, 52240). *J. Lipid Res.* 10, 388-394 (1969). Three artificially inseminated laying White Leghorn hens were given 35-50 µg of cholesterol-4-¹⁴C intravenously. Their subsequently produced eggs contained cholesterol-¹⁴C-labeled yolks. Some of the fertilized eggs were analyzed for cholesterol content and radioactivity. Other eggs were incubated until hatching. The specific activity of the cholesterol contained in the serum and tissues of newly hatched chicks was determined and compared with that of yolk sac, which was taken as representative of egg yolk cholesterol before its metabolic transfer into the chick embryo. The specific activities of cholesterol in intestine, liver, serum, heart and skeletal muscle and the whole chick were 95-58% of that in yolk sac, but that of brain cholesterol was only 11% of this value. These results indicate that whereas most of the cholesterol in the chick originated from the egg yolk, cholesterol biosynthesis was active in the brain and provided about 90% of its cholesterol content.

CHARACTERIZATION OF LIPOPROTEIN PARTICLES ISOLATED FROM THE GOLGI APPARATUS OF RAT LIVER. R. W. Mahley, R. L. Hamilton and V. S. Lequire (Depts. of Pathology & Anatomy, Vanderbilt Univ. School of Med., Nashville, Tenn. 37203). *J. Lipid Res.* 10, 433-439 (1969). It has been proposed that particles within tubules and vesicles of the Golgi apparatus of liver cells are precursors of very low density lipoproteins in blood plasma. To characterize these particles a cell fraction rich in Golgi apparatus and associated particles was isolated from rat liver in quantities sufficient for analysis. Particles freed from the membranes of the Golgi apparatus and floated at $d = 1.006$ were studied by the chemical analysis, immunodiffusion and paper electrophoresis. The lipid composition of the

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Golgi particles was similar to that of very low density lipoproteins from the same rats. The protein content was about 10% of dry weight for both the Golgi particles and plasma very low density lipoproteins. It was concluded that the lipoproteins in the Golgi apparatus are the precursors of plasma very low density lipoproteins.

METABOLISM OF $1\text{-}^{14}\text{C}$ PALMITIC ACID IN GOATS IN VARIOUS METABOLIC STATES. S. Yamdagni and L. H. Schultz (Dept. of Dairy Sci., Univ. of Wisconsin, Madison). *J. Dairy Sci.* 52, 1278-88 (1969). Palmitate- $1\text{-}^{14}\text{C}$ albumin complex was constantly infused intravenously through jugular catheter for four hours in goats in three metabolic states: I, 72-hr fasted, phlorizinized, and nonlactating; II, normally fed and lactating; III, 72-hr fasted, phlorizinized and lactating. The ^{14}C activity was measured in ketone bodies and various lipid fractions of plasma at various time intervals, and quantitation of these fractions was carried out. The estimated percentage of C atoms of plasma ketone bodies derived from plasma free fatty acids were 89, 41 and 103 in the respective three trials. The ratio between β -hydroxybutyrate and acetone + acetoacetate changed from 4:1 in Trial I to 1.4:1 in Trial III; at the same time the concentration of total ketone bodies was three times higher in Trial III. The turnover rates of free fatty acids were 4.2, 4.3, and 7.9 mg/kg/min for Trials I, II, and III, respectively. The corresponding turnover times were 4.6, 1.8, and 3.1 min. Production of ketone bodies from free fatty acids and free fatty acid turnover rate were directly related to the stress conditions imposed on the animals. In all metabolic states the major ^{14}C activity of plasma was recovered in the free fatty acid fraction, whereas in liver most of the activity was in glycerides. Adipose tissue samples in all the trials contained only a small amount of ^{14}C activity.

THE SIDE-CHAIN CLEAVAGE OF CHOLESTEROL AND CHOLESTEROL SULFATE BY ENZYMES FROM BOVINE ADRENOCORTICAL MITOCHONDRIA. D. G. Young and P. F. Hall (Russell Grimwade School of Biochem., Univ. of Melbourne, Parkville, Victoria,

Australia). *Biochemistry* 8, 2987-2997 (1969). The requirements for the conversion of cholesterol into pregnenolone and of cholesterol sulfate into pregnenolone sulfate (called side-chain cleavage) by fractions from bovine adrenocortical mitochondria have been examined and found to be the same for both substrates. The requirements are reduced triphosphopyridine nucleotide, a reduced triphosphopyridine nucleotide-cytochrome P-450 reductase, nonheme iron, cytochrome P-450 and molecular oxygen. It is likely that some additional factor(s) may be required. Optimal conditions for side-chain cleavage were obtained with both substrates and the rates of cleavage with the two substrates were compared using three solvents or suspending agents, namely, N,N-dimethylformamide, bovine serum albumin and Tween-80. Values for apparent K_m and V_{max} with standard errors were calculated by least-squares fit. The apparent K_m for cholesterol sulfate (18 μM) is less than that for cholesterol (40-50 μM). Anomalous results were observed when cholesterol sulfate was suspended in Tween-80 and when cholesterol was added in N,N-dimethylformamide, probably as the result of inadequate saturation of the enzyme. Values for V_{max} with both substrates were of the same general order (1.5-5.9 $\mu\text{moles/min per mg of protein}$). The sum of the cleavage of the two substrates incubated separately approximately equals the total cleavage of both substrates incubated together. These observations suggest that under optimal conditions the two substrates are cleaved at about the same rate and that cholesterol and cholesterol sulfate are cleaved by separate enzymes.

LIPID PEROXIDE FORMATION IN MICROSOMES. THE ROLE OF NON-HAEM IRON. E. D. Wills (Dept. Biochem., Med. Coll. St. Bartholomew's Hosp., London E.C.1. England). *Biochem. J.* 113, 325-32 (1969). Metal ion-chelating agents such as EDTA, o-phenanthroline or desferrioxamine inhibit lipid peroxide formation when rat liver microsomes prepared from homogenates made in pure sucrose are incubated with ascorbate or NADPH. Microsomes treated with metal ion-chelating agents do not form peroxide on incubation unless inorganic iron

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5. F. A. Hughes and E. N. Harvey, Jr.
6. D. Dutton
7. K. K. Carroll
8. H. Schlenk
9. L. A. Horrocks
10. Participants of First Symposium on Ether-Linked Lipids. Randall Wood; G. A. Thompson, Jr.; Fred Snyder; F. Spencer; D. C. Malins; L. A. Horrocks; W. E. M. Lands; G. B. Ansell; Hildegard Debuch; Howard Goldfine; and Morris Kates.
11. S. Usui, M. Hedberg, J. M. Hartlage, W. L. Freyberger and D. W. Frommer
12. R. O. Feuge
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17. A. O. Lenghi
18. U. Varnasi
19. F. Spencer
20. H. W. Knoche
21. Y. Hirano

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(Fe²⁺ or Fe³⁺) in a low concentration is added subsequently. No other metal ion can replace inorganic iron adequately. Microsomes prepared from sucrose homogenates containing EDTA (1mM) do not form lipid peroxide on incubation with ascorbate or NADPH unless Fe²⁺ is added. Washing the microsomes with sucrose after preparation restores most of the capacity to form lipid peroxide. Lipid peroxide formation in microsomes prepared from sucrose is stimulated to a small extent by inorganic iron but to a greater extent if adenine nucleotides, containing iron compounds as a contaminant, are added. The iron contained in normal microsome preparations exists in haem and in non-haem forms. One non-haem component in which the iron may be linked to phosphate is considered to be essential for both the ascorbate system and NADPH system that catalyze lipid peroxidation in microsomes.

LIPID PEROXIDE FORMATION IN MICROSOMES: RELATIONSHIP OF HYDROXYLATION TO LIPID PEROXIDE FORMATION. *Ibid.*, 333-341. Aminopyrine strongly inhibits NADPH-induced lipid peroxide formation in rat liver microsomes, but ascorbate-induced peroxidation is inhibited to a smaller extent. Aminopyrine oxidation is stimulated by Mg²⁺ but inhibited by Ca²⁺. Concentrated solutions (10mM) of iron-chelating agents inhibit aminopyrine oxidation, but the more dilute solutions (0.5mM) of chelators that block lipid peroxide formation do not inhibit aminopyrine oxidation. Induction of lipid peroxide formation in microsomes by incubation with ascorbate or NADPH or by treatment with ionizing radiation leads to a sharp decline in the ability of microsomes to oxidize aminopyrine or hydroxylate aniline. It is considered that the two processes of hydroxylation and lipid peroxide formation are closely linked in microsomes. They probably depend on the same electron-transport chain, and peroxide formation, which involves membrane disintegration, may be part of the normal membrane remodelling process.

MEMBRANES OF ANIMAL CELLS. IV. LIPIDS OF THE L CELL AND ITS SURFACE MEMBRANE. D. B. Weinstein, J. B. Marsh, M. C. Glick and L. Warren (Dept. Biochem., School Dental

Med., Univ. Pa., Philadelphia, Pa. 19104). *J. Biol. Chem.* 244, 4103-11 (1969). Surface membranes were isolated by the fluorescein-mercuric acetate and Tris procedures from mouse fibroblasts (L cells) grown in suspension culture. Membranes isolated by the fluorescein-mercuric acetate method contained approximately 4.7% of the protein and 13.8% of the lipid of the whole cell. Approximately 95% of the plasma membrane lipid was accounted for. Comparison of lipid-protein ratios for whole cells and surface membranes shows a 3-fold enrichment of lipid in the surface membrane. Neutral lipids make up 40% of the total membrane lipid, with cholesterol and triglycerides accounting for 20 and 13%, respectively. The high cholesterol content of the surface membrane is reflected in a cholesterol-phospholipid molar ratio of 0.69 while the corresponding ratio for total cell lipid is 0.26. Fifty-nine per cent of the membrane phospholipids contain choline, and the surface membranes contain more sphingomyelin and less lecithin and phosphatidylethanolamine than the total cell lipid. Phosphatidylserine and sphingomyelin show a 2- to 3-fold enrichment of unsaturated and long chain fatty acids relative to other membrane phospholipids. The lipid composition of the surface membranes of L cells was compared to the composition reported in the literature for surface membranes of animal cells. The comparison indicates that the lipid compositions of L cell and liver cell surface membranes are basically similar. Lipid composition may be useful in classifying surface membrane preparations.

CHARACTERIZATION OF RAT TISSUES BY GAS-LIQUID CHROMATOGRAPHY WITH THE ELECTRON-CAPTURE DETECTOR. M. Theiner and L. Friedner (Dept. of Nutr. and Food Sci., Mass. Inst. Technol., Cambridge 02139). *Proc. Soc. Exp. Bio. Med.* 131, 1073-6 (1969). The unsaponifiable lipids of rat tissue include a number of components which possess moderate affinities for electrons with thermal energies. These can be separated and analyzed by gas-liquid chromatography coupled with simultaneous detection by flame ionization and electron capture. Different rat tissues yield characteristic chromatographic patterns.

THE EFFECT OF GUANINE DERIVATIVES ON PALMITATE-1-¹⁴C INCORPORATION INTO RAT EPIDIDYMAL ADIPOSE TISSUE. M. M. Szabo and M. S. Goldstein (Div. of Metabolism and Endocrinology, Dept. of Med., Michael Reese Hosp. and Med. Center, Chicago 16, Ill.). *Proc. Soc. Exp. Bio. Med.* 131, 1055-1059 (1969). The incorporation of palmitate-1-¹⁴C into rat epididymal adipose tissue lipids was markedly enhanced by the *in vitro* additions of guanosine and its 5'-phosphate esters and to a lesser extent by adenosine, uridine and cytidine. The magnitude of the response to guanosine depended on the glucose concentration of the incubating medium and the nutritional state of the donor animal; little or no stimulation was obtained when esterification was already enhanced by the presence of glucose in the medium or when fat pads of fasted-refed rats were employed.

THE ISOLATION AND IDENTIFICATION OF 25-HYDROXYERGOCALCIFEROL. T. Suda, J. F. DeLuca, J. K. Schnoes and J. W. Blunt (Dept. of Biochem., Univ. of Wisconsin, Madison, Wis.). *Biochemistry* 8, 3515-20 (1969). A polar metabolite of vitamin D₂ (314 μg) has been isolated in pure form from the blood of four pigs given 500,000 IU of ergocalciferol/day for 26 days. It has been identified as 25-hydroxyergocalciferol by means of ultraviolet spectra, gas-liquid partition chromatography, nuclear magnetic resonance spectra, mass spectra and mass spectra of its trimethylsilyl ether derivative. It is about 1.5 times more active than vitamin D₃ or D₂ in curing rickets in rats.

FATTY ACID OXIDATION, CITRIC ACID CYCLE ACTIVITY AND MORPHOLOGY OF MITOCHONDRIA IN DIABETIC RAT LIVER. Y. Harano, R. G. DePalma and M. Miller. (Depts. of Med. and Surgery, Case Western Reserve Univ., Cleveland, Ohio 44106). *Proc. Soc. Exp. Biol. Med.* 131, 913-917 (1969). Palmitate and oleate oxidation in isolated mitochondria from alloxan diabetic rat liver were shown to be elevated two or threefold over normal. These increases were accompanied by enlargement of mitochondria as observed by electron microscopy. These metabolic and structural changes were reversed by treating the diabetic rat with insulin. The activities of citric acid cycle enzymes in isolated mitochondria were unchanged in the diabetic state. It is suggested that insulin regulates long chain fatty acid oxidation in the liver by an effect on mitochondria structure and function.

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DERIVATIVES OF ALDOSTERONE FOR GAS PHASE ANALYSIS. E. C. Horning and B. F. Maume (Inst. Lipid Res., Baylor Univ. College of Med., Houston, Texas 77025). *J. Chromat. Sci.* 7, 411-418 (1969). The preparation of derivatives of aldosterone for use in GLC separations was studied. Several derivatives with good gas chromatographic properties were prepared. Since this steroid hormone is usually present in biologic systems in very low concentration, the chief requirement is for a derivative or derivatives that may be used with an electron capture detector. The methoxime-heptafluorobutyryl ester (MO-HFB) derivatives (four isomers) separate as a single peak with a non-selective column, and these derivatives may be detected at nanogram and subnanogram levels. The structures of the derivatives prepared in this study were determined by mass spectrometry. Aldosterone reacts primarily as the C-18 hemiacetal although a tri-Mo structure may be prepared by prolonged reaction with methoxylamine. Derivatives of the hemiacetal structure have the greatest usefulness for GLC work.

ESTIMATION OF HERITABILITY AND REPEATABILITY OF MILK AND MILK FAT PRODUCTION WITH SELECTION EFFECTS REMOVED. D. F. Butcher and A. E. Freeman (Dept. of Animal Sci., Iowa State Univ., Ames, Iowa). *J. Dairy Sci.* 52, 1259-67 (1969). Two groups of data were used for this study. One group contained 110,084 and the other group 106,296 lactations on registered and grade Holstein cows. All records were analyzed as deviations from regressed adjusted herd-year-season averages. Estimates of the relationship between various pairs of lactations made by using five different methods gave essentially the same results. It was concluded that regressions or intraclass correlations, where all records of the pair are included, are much easier to compute and give essentially the same results as the more complicated procedures of Maximum Likelihood and construction of variances and covariances to try to remove the effects of selection. Heritability estimates obtained by using paternal half-sib analyses of variance were much lower than daughter-dam regression estimates for one group of data, but they were in good agreement in the other group of data. Estimates of heritability obtained by intrasire, intraherd regression of daughter on dam were slightly lower than intrasire regression, indicating that use of deviated records does not remove all variations due to herds from a daughter-dam regression. The derivation of a method to estimate heritability and repeatability by removing the effects of selection is presented.

THE EFFECT OF HORMONES ON HEPATIC CHOLESTEROL ESTER SYNTHESIS IN VITRO. J. S. Schweppe and R. A. Jungmann (Dept. Res., Chicago Wesley Memorial Hosp., and Depts. of Med. and Biochem., Northwestern Univ. Med. School, Chicago, Ill.). *Proc. Soc. Exp. Biol. Med.* 131, 868-870 (1969). The effects of hormones on the *in vitro* biosynthesis of cholesterol palmitate, oleate and linoleate by rat liver microsomes from cholesterol and fatty acids was studied. It appears that L-thyroxine and glucagon in general stimulate the synthesis of all three esters. Testosterone increases cholesterol palmitate and oleate formation. Its effect on cholesterol linoleate formation varies, being stimulatory at the highest concentration and inhibitory at the lowest concentration of hormone used. The action of 17 β -estradiol on cholesterol esterification is also dependent on the concentration of the hormone used. However, cholesterol oleate synthesis was markedly stimulated at all concentrations of 17 β -estradiol.

FRACTIONATION OF HUMAN SERUM HIGH DENSITY LIPOPROTEIN IN UREA SOLUTIONS. EVIDENCE FOR POLYPEPTIDE HETEROGENEITY. A. Seanu, J. Toth, C. Edelstein, S. Koga and E. Stiller (Depts. of Med. and Biochem., Univ. of Chicago, and Argonne Cancer Res. Hosp., Chicago, Ill.). *Biochemistry* 8, 3309-16 (1969). The lipid-free protein (apo-HDL₂) of immunologically pure human serum HDL₂ (d 1.063-1.125) was separated by Sephadex G-200 chromatography in 8M urea into four components: I, III, IV, and V with a per cent weight distribution of 5, 65, 22 and 8, and an apparent molecular weight of (10³) 52.0-55.0, 25.8-28.0, 16.4-17.6 and 11.2-11.8, respectively (calibrated Sephadex columns, and disc gel electrophoresis in sodium dodecyl sulfate). Equilibrium ultracentrifugation analysis corroborated the molecular weight data obtained by chromatography and electrophoresis. However, under appropriate conditions of speed and protein concentration, III exhibited a smaller molecular

The Lighter Side of Fall Meeting



Activities

1. R. H. Hagberg and Mrs. J. P. Krumbein
2. Mrs. & Mr. J. P. Hughes, Mrs. & Mr. L. E. Brown and Mr. & Mrs. R. T. O'Connor
3. J. C. Cowan, W. O. Lundberg, G. C. Cavanagh, J. C. Conan, R. C. Stillman and R. Reiser
4. J. Murphy, G. Colbert, Mrs. & Mr. A. E. Brust and R. G. Krishnamurthy
5. T. Lentz, Mrs. R. A. Hagberg and Mrs. & Mr. R. S. Smith
6. B. Boer, Mrs. A. A. Schmitz and Mrs. G. Lichtenwalter
7. Dr. & Mrs. O. E. Bell and Mrs. & Mr. O. N. Brenik
8. Mrs. J. C. Cowan, Mrs. M. F. Formo and Mrs. A. Privett
9. B. N. Lutsky, C. B. Hirschberg, J. A. Martin and F. R. Albright
10. P. E. Pfeffer, V. G. Martin and H. Rothbart
11. Mr. & Mrs. H. Hamilton
12. President's Cocktail Party
13. President's Cocktail Party
14. R. C. Christiansen and J. C. Lamping
15. Mr. & Mrs. J. Konen and D. S. Bolley
16. D. Dutton and L. H. Going

weight component of approximately 17,000. Fraction II was minor and only occasionally seen. Fraction I appeared as an aggregate of the other components. Fractions III, IV and V differed in immunological and spectral (circular dichroism and ultraviolet absorption spectroscopy) properties, electrophoretic mobility (disc gel electrophoresis) and amino acid composition. These three fractions all showed heterogeneity by disc gel electrofocusing. The results suggest that apo-HDL₂ contains three distinct classes of polypeptides which are totally (IV and V) or partially (III) dissociated in urea and are probably made up of subcomponents. The observed similarity in band pattern (disc gel electrophoresis in the presence or absence of urea) between HDL₂ and apo-HDL₂ was taken to support the hypothesis that HDL₂ is made up of lipoprotein subspecies with distinct peptide moieties.

THE RELATIVE VALUE OF RAPESEED AND SOYBEAN OILS IN CHICK STARTER DIETS. R. E. Salmon (Res. Station, Res. Branch, Canada Agr., Swift Current, Saskatchewan, Canada). *Poultry Sci.* 48, 1045-50 (1969). The response of male broiler chicks to 10% rapeseed oil (RSO), soybean oil (SBO), or combinations of the oils in starter diets containing 22 or 25% protein was studied to 41 days of age in two experiments. Diets containing 10% RSO alone or 2.5, 5 or 7.5% RSO with 7.5, 5 or 2.5% SBO depressed growth as compared with 10% SBO in Experiment 1. In Experiment 2, 10% RSO in either the low or high protein basal diet depressed the growth of chicks. Feed conversion to 13 days of age was significantly depressed by diets containing RSO in Experiment 1 but not in Experiment 2. No significant differences in feed conversion occurred during later periods of either experiment or in dietary metabolizable energy between oil treatments during the second or sixth week of either experiment. The failure of RSO to depress the growth of chickens in earlier studies is attributed to a higher saturated fatty acid content in the diets used in the previous studies. The abdominal depot fat contained relatively more palmitic, palmitoleic, stearic and oleic acids and less linoleic and linolenic acids

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than the dietary fat. The depot fat contained only 50% of the level of erucic acid contributed by diets containing RSO. Analysis of depot fat at weekly intervals revealed little change in fatty acid composition to 41 days of age.

STUDIES ON THE POSITIONAL INTEGRITY OF GLYCERIDE FATTY ACIDS DURING DIGESTION AND ABSORPTION IN RATS. S. S. Raghavan and J. Ganguly (Dept. of Biochem., Indian Inst. of Science, Bangalore 12, India). *Biochem. J.* 113, 81-87 (1969). Rats previously starved for 24 hr. were separately given by intraduodenal injections 0.5 ml of a dispersion containing 10 mg of sodium taurocholate, with 50 mg of glycerol 1,3-dioleate 2-palmitate-1-¹⁴C, glycerol 1,2-dioleate 3-palmitate-1-¹⁴C, a mixture of palmitic acid-1-¹⁴C and triolein, or a mixture of palmitic acid-1-¹⁴C and oleic acid. At the end of 30 min, the net amounts and the radioactivity of the neutral lipid components recovered from the intestinal lumen and mucosa, and the position of the labelled palmitic acid in the mucosal triglycerides, were determined. When glycerol 1,3-dioleate 2-palmitate-1-¹⁴C was administered, most of the labelled acid was retained in the di- and mono-glycerides of the lumen; the triglycerides were the major components containing the radioactivity in the mucosa and 75-80% of the labelled acid was located at the β -position of these triglycerides.

ETHANOL-INDUCED ARTIFACTS IN THE METABOLISM OF ³H-VITAMIN D₃. G. Ponchon and H. F. DeLuca (Dept. of Biochem., Univ. of Wisconsin, Madison 53706). *Proc. Soc. Exp. Biol. Med.* 131, 727-731 (1969). The fate of vitamin D₃ in vitamin D-deficient rats was studied following the intravenous injection of 10 IU (0.25 μ g) of vitamin D₃-1,2-³H dissolved either in 0.05 ml of ethanol or 0.05 ml of blood plasma. Ethanol accelerates the disappearance of the radioactivity from the plasma compartment and decreases the characteristic rebound of plasma radioactivity occurring after 1-4 hr. The metabolic and toxic effects of ethanol are discussed, and the use of plasma as a vehicle for intravenous injections of vitamin D is advocated.

STERIC EFFECTS IN THE GLYCERALDEHYDE 3-PHOSPHATE DEHYDROGENASE CATALYZED HYDROLYSIS OF ACYL PHOSPHATES. AN EXAMPLE OF SUBSTRATE-INDUCED COOPERATIVITY. D. R. Phillips and T. H. Pife (Dept. of Biochem., Univ. So. Cal., Los Angeles, Cal.). *Biochemistry* 8, 3114-3119 (1969). The acyl phosphatase activity of glyceraldehyde 3-phosphate dehydrogenase has been examined at 25C. This activity can conveniently be divided into three categories depending upon the steric bulk in the acyl group of the substrate, that of: (1) acetyl phosphate which follows apparent second-order kinetics, K_m being too large to measure; (2) propionyl, butyryl, isobutyryl, and isovaleryl phosphate which obey normal Michaelis-Menten kinetics; and (3) the highly branched compounds, trimethylacetyl and 3,3-di-methylbutyryl phosphate, which are not substrates. Arsenate increases the rate of hydrolysis of the compounds in group 2, showing the rate-determining step for those compounds to be deacylation of the acyl enzyme intermediate. Methyl phosphate, however, was found to be a poor catalyst for deacylation of the enzyme. This and other observations support a general base mechanism for deacylation of glyceraldehyde 3-phosphate dehydrogenase. The highly branched compounds bind to the enzyme since they inhibit acetyl phosphatase activity. In addition, compounds in groups 2 and 3 inhibit the dehydrogenase activity. This inhibition was found to be sigmoidal and was related to a cooperativity effect on binding to the enzyme. A plot of $\log k_3$ (deacylation) vs. E_s , the Taft steric effects constants, had a slope of 1.1.

INPUT-OUTPUT ANALYSIS FOR TOTAL INPUT RATE AND TOTAL TRACED MASS OF BODY CHOLESTEROL IN MAN. W. Perl and P. Samuel (Cardiorespiratory Res. Lab., Goldwater Memorial Hosp., Welfare Island, N.Y., N.Y. 10017). *Circ. Res.* 25, 191-9 (1969). The Stewart-Hamilton theorems for flow and volume are generalized to yield total input rate and total traced mass in multiple input, steady-state systems with partially labeled input. Application is made to existing decay curves of tracer cholesterol in human serum measured under a control steady state and again under a steady state of neomycin administration which lowered the serum cholesterol level. The effect of neomycin on the total traced mass of body cholesterol was to reduce it by 38, 40, 32, and 24 g., corresponding to 34, 40, 25, and 33%, in four patients studied.

Social Events of Minneapolis Meeting



1. H. Birnbaum, R. D. Wood, R. J. Reiser, R. Reiser, Mrs. R. Reiser, E. M. Loyd and Mrs. R. J. Reiser
2. R. D. Feuge, Mr. & Mrs. L. E. Brown, G. Sumrelo and N. V. Lovegren
3. R. M. Burton, G. Rouser, G. B. Ansell, R. Anderson and R. D. Wood
4. R. B. Wettstrom, E. Marshack and M. Mattikow
5. J. Bunn, Jr., G. Grady, W. M. Barger, W. H. Jennings, N. Brinkmeyer and G. Wadlington
6. Mr. & Mrs. A. G. Johanson and D. S. Fritz
7. Overall view of the Banquet
8. Mr. & Mrs. L. V. Anderson
9. Mrs. R. J. Reiser, H. Schlenk and R. J. Reiser
10. O. Privett, K. E. Guyer and S. Ramachandran
11. Howe-Baker Cocktail Party
12. T. Stovall and L. T. Pyle
13. Mr. & Mrs. T. Stovall, Jr.
14. President's Cocktail Party
15. D. W. Launden and Mrs. M. F. Formo
16. Mrs. Greenwalt and D. S. Fritz
17. Drew Chemical Cocktail Party
18. Mrs. F. A. Hughes
19. Mr. & Mrs. J. C. Cowan
20. R. H. Potts
21. Drew Chemical Cocktail Party

The present analysis utilizes only the area and the first time moment of the plasma decay curve. It is applicable to decay curves of more general shape than those that can be fitted by a small number of exponentials. The analysis does not require the assumption of compartments.

THE EFFECT OF ASCORBIC ACID ON THE CARBOHYDRATE METABOLISM OF VITAMIN A-DEFICIENT CHICKS. M. Perok and J. Kendler (Dept. of Animal Hygiene and Poultry Sci., The Hebrew Univ., Jerusalem, Israel). *Poultry Sci.* 48, 1101-04 (1969). Blood glucose levels, determined at 19 and 33 days of age, showed a strong hyperglycaemic response in the vitamin-deficient chicks as compared to their controls (320.5 and 292.0 vs. 168.1 and 198.4 mg./100 ml., respectively). Liver glycogen concentrations in the avitaminotic chicks were reduced to less than half as compared to the control levels (0.51 vs. 1.09 and 1.26 vs. 3.56 per cent, respectively).

PREVENTION OF VITAMIN A DEFICIENCY. Sheila M. Pereira and A. Begum (Dept. Nutr. Res., Christian Med. College and Hosp., Vellore, South India). *Am. J. Clin. Nutr.* 22, 858-862 (1969). A single massive oral dose of vitamin A (100,000 μ g) given to normal preschool children maintained higher serum levels for about 13 weeks compared to the controls. The same dose given to vitamin A-depleted children maintained adequate serum levels for about 15 weeks.

BASIC STUDIES ON THE MECHANISM OF ACTION OF VITAMIN D. A. W. Norman, M. R. Haussler, T. H. Adams, J. F. Myrtle, Patricia Roberts and K. A. Hibberd (Dept. Biochem., Univ. Cal., Riverside, Cal.). *Am. J. Clin. Nutr.* 22, 396-411 (1969). Studies on the cellular and subcellular localization of vitamin D and its metabolites have led to the suggestion that the vitamin is first metabolized to an as yet uncharacterized more polar compound that then associates stereospecifically with a receptor in the chromatin or DNA-containing portion of the mucosal cells. This in turn activates the biochemical expression of genetic information, which ultimately results in the physiological responses characteristic of vitamin D.

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STRUCTURE OF CELL WALL LIPOPOLYSACCHARIDE FROM *SALMONELLA TYPHIMURIUM*. Hiroshi Nikaido (Biochem. Res. Lab., Mass. Gen. Hosp., Boston, Mass.). *J. Biol. Chem.* 244, 2835-45 (1969). Lipopolysaccharide of wild type *Salmonella typhimurium* is believed to contain 0 side chains, whose reducing terminal sugar, galactose, is linked to the R core. Two oligosaccharides were isolated by periodate oxidation, NaBH₄ reduction, and mild acid hydrolysis of such a lipopolysaccharide. They were also isolated from the lipopolysaccharide of a mutant synthesizing very short 0 side chains each containing only one "repeat unit," but not from that of a mutant totally lacking 0 side chains. The degradation of a lipopolysaccharide, which was synthesized partially *in vitro* and contained ¹⁴C only in the reducing terminal galactose residue of the 0 side chain, also resulted in the isolation of these two oligosaccharides, labeled with ¹⁴C. These results indicate that the oligosaccharides were derived from the linkage region between the 0 side chain and R core. Moreover, one of the oligosaccharides was identified as O-D-galactosyl-O-D-glucosyl-(1→2)-glyceraldehyde; the other appeared to be a cyclic acetal formed from the former oligosaccharide and glycerol. These and other results show that the reducing terminal galactose of 0 side chains is not linked to the nonreducing terminal sugar of the R core, i.e. N-acetyl-D-glucosamine, but to a glucose residue within the R core, through a 1→4 linkage if the sugars are in pyranose forms. This glucose in turn is linked, through a 1→2 linkage, to a galactose residue in the R core.

VITAMIN E DEFICIENCY AND THE ACCUMULATION OF AMINO ACIDS IN SKELETAL MUSCLE. G. E. Nicholals, R. R. Jones, J. F. Diehl and C. D. Fitch (Depts. of Biochem., Pathol. and Med., Univ. of Arkansas School Med., Little Rock, Ark.). *J. Nutr.* 99, 27-33 (1969). The movements of ¹⁴C-labeled glycine and α-aminoisobutyric acid (AIB) in and out of skeletal muscle slices from control and vitamin E-deficient rabbits were studied. Vitamin E deficiency increased ¹⁴C-amino acid accumulation at 60 minutes of incubation without having any demonstrable effect either on the early phase of entry (zero to 30 minutes) or on the efflux of ¹⁴C-amino acids. Inhibition of ¹⁴C-glycine incorporation into protein by puromycin had no effect either on the early phase of entry or on the accumulation of ¹⁴C-glycine by control or vitamin E-deficient muscle. The early phase of ¹⁴C-glycine entry followed first-order kinetics; its rate constant in sec⁻¹ was 5.5 × 10⁻⁴. Whereas steady-state distribution ratios of ¹⁴C-amino acids were reached by control muscle in about 30 minutes, dystrophic muscle continued to accumulate the ¹⁴C-amino acids for 60 minutes or more. Small differences in influx, efflux, or both, can result in significant differences in net accumulation with extended time. Thus, we suggest that vitamin E-deficient muscle has one or more compartments capable of increasing the accumulation of ¹⁴C-glycine and ¹⁴C-AIB due to a change either in influx or efflux which is too small to demonstrate. These compartments may be located in regenerating muscle cells, or in other cell types more abundant in the dystrophic muscle.

CHEMISTRY AND BIOLOGY OF PHOSPHOLIPIDS FROM AN UNCLASSIFIED MYCOBACTERIA, P6. M. Motomiya, A. Mayama, M. Fujimoto, H. Sato and S. Oka. (Res. Inst. for Tuberculosis, Leprosy, and Cancer, Tohoku Univ., Sendai, Japan). *Chem. Phys. Lipids* 3, 159-167 (1969). Crude phospholipid fraction from P6 (Scotochromogen) prepared by extraction with chloroform:methanol (2:1), followed by removal of non-lipid contaminants by Folch's procedure and extraction with acetone, yielded three fractions by column chromatography with silicic acid. Fraction I was identified as diphosphatidyl glycerol (cardiolipin) and was antigenic in flocculation test for syphilis and in latex agglutination test for lepromatous leprosy. Fraction II was identified as phosphatidyl ethanolamine and Fraction III as phosphatidyl inositol monomannoside.

IN VITRO INCORPORATION OF FORMATE-¹⁴C, GLYCEROL-1,3-¹⁴C, CHOLINE-1,2-¹⁴C, SERINE-1-¹⁴C, AND ETHANOLAMINE-1,2-¹⁴C INTO THE MAJOR PHOSPHOLIPIDS OF HUMAN PERIPHERAL ARTERIES. R. J. Morin (Dept. Pathol., Los Angeles County Harbor Gen. Hosp., Torrance, Calif. 90509). *Proc. Soc. Exp. Biol. Med.* 131, 880-2 (1969). The relative degrees of *in vitro* incorporation of ¹⁴C labeled precursors into surgical specimens of human femoral, tibial and popliteal arteries were choline > ethanolamine > serine > formate > glycerol. Formate ap-

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peared to be incorporated primarily into phosphatidyl serine, probably by fixation to glycine. In addition to *de novo* synthesis, the pathway of transmethylation of phosphatidyl ethanolamine to phosphatidyl choline seemed to be operative in these arterial preparations.

KETOGENESIS AND CHOLESTEROL SYNTHESIS IN NORMAL AND NEOPLASTIC TISSUES OF THE RAT. J. D. McGarry and D. W. Foster (Dept. of Internal Med., Univ. of Tex. Southwestern Med. School at Dallas, Dallas, Tex. 75235). *J. Biol. Chem.* 244, 4251-56 (1969). Enzymes of ketogenesis were measured in homogenates of liver, kidney, brain, intestine, heart, muscle and three transplantable hepatomas of the rat. Cholesterol synthesis from acetate-2-¹⁴C was studied in slices from these tissues. The results show that high levels of β-hydroxy-β-methylglutaryl coenzyme A synthase are unique to liver and the well differentiated hepatomas, and thus provide a biochemical basis for liver's role as the primary ketogenic organ. The ability to synthesize cholesterol from acetate was similar in liver, intestine and immature brain, all of which contained adequate β-hydroxy-β-methylglutaryl-CoA synthase to support the observed rates of cholesterologenesis. The failure of the undifferentiated hepatoma 3924A to synthesize acetoacetate or cholesterol from acetate was consistent with its lack of β-hydroxy-β-methylglutaryl-CoA synthase.

SOME SELENIUM RESPONSES IN THE RAT NOT RELATED TO VITAMIN E. K. E. M. McCoy and P. H. Weswig (Dept. of Agr. Chem., Oregon State Univ., Corvallis, Oregon). *J. Nutr.* 98, 383-89 (1969). Rats fed a low selenium ration containing Torula yeast with adequate vitamin E grew and reproduced normally. Their offspring were almost hairless, grew more slowly and failed to reproduce. Supplemental DL-methionine, sodium sulfate or increased dosage of vitamins was without effect. Supplementing with 0.1 ppm selenium as sodium selenite restored haircoat, growth and reproductive capabilities. Some usual biochemical tests for selenium or vitamin E deficiency status, or both, were inconclusive when compared with rats fed normal rations. Rats fed low selenium corn protein also had sparse hair and poor growth in the second generation. A low selenium ration from ewe muscle, however, supported rats for three generations without any abnormalities.

NUCLEAR MAGNETIC RESONANCE STUDIES OF SERUM LOW DENSITY LIPOPROTEINS (LDL₂). R. B. Leslie and D. Chapman (Molecular Biophysics Unit, Unilever Res. Lab., The Frythe, Welwyn, Hertfordshire, Great Britain) and A. M. Scanu. *Chem. Phys. Lipids* 3, 152-158 (1969). These N.M.R. results indicate that in LDL₂ the polar head group of the phospholipid is quite free and probably in an aqueous environment, but that the non-polar aromatic amino acid residues of the protein are somehow immobilized probably by apolar interactions with lipids. On the other hand the lipid hydrocarbon chains are quite freely mobile, and the molecular freedom of the cholesterol nucleus in the cholesterol esters increases with increasing temperature.

LIPID PEROXIDATION IN VITRO BY ISOLATED FAT CELLS OF RATS. CORRELATION WITH TOTAL LIPOLYSIS, GLUCOSE UTILIZATION, AND DIETARY TOCOPHEROL. V. R. Lavis, A. E. Kitabchi and R. H. Williams (Univ. of Wash., Dept. of Med., Div. of Endocrinology, Seattle, Wash. 98105). *J. Biol. Chem.* 244, 4382-86 (1969). Lipid peroxidation *in vitro* by isolated rat adipose



ABSTRACTS: BIOCHEMISTRY AND NUTRITION

tissue was measured by the thiobarbituric acid reaction. Incubation of isolated fat cells with adrenocorticotrophic hormone or epinephrine stimulated total lipolysis as measured by glycerol release, but not lipid peroxidation. Epinephrine inhibited the lipid peroxidation produced by ascorbate. However, prior treatment with epinephrine caused increased lipid peroxidation by epididymal fat pads. The isolated fat cells of vitamin E-deficient rats contained no detectable tocopherol. These fat cells showed no significant differences in lipid peroxidation, total lipolysis, or basal or insulin-stimulated glucose carbon incorporation into CO_2 and total lipids, as compared to fat cells from vitamin E-replete animals. Enhancement of lipid peroxidation *in vitro* by incubation of isolated fat cells with ascorbate caused no change in glucose-carbon utilization or total lipolysis. We conclude that the thiobarbituric acid test, applied to fat cells, is neither an indicator of total lipolysis, nor of vitamin E deficiency. Fat cell glucose utilization and lipolysis are not altered by either vitamin E deficiency or increased lipid peroxidation.

BIOSYNTHESIS OF FATTY ACIDS IN OBESE MICE IN VIVO. II. STUDIES WITH DL-MALATE-2- ^3H -3- ^{14}C , SUCCINATE-2,3- ^3H -2,3- ^{14}C , AND DL-ISOCITRATE-2- ^3H -5,6- ^{14}C . E. Lamdin, W. W. Shreeve, R. H. Slavinski and N. Oji (Div. Biochem., Med. Res. Center, Brookhaven Nat. Lab., Upton, N.Y.). *Biochemistry* 8, 3325-3331 (1969). Biosynthesis of fatty acids in the liver and in other tissues of the remaining carcass of obese hyperglycemic mice and their lean siblings has been investigated by isolation and counting of radioactivity in total fatty acids

of mice sacrificed 90 min. after intraperitoneal injection of trace amounts of malate, succinate or isocitrate specifically labeled with tritium and carbon-14. All ^3H - and ^{14}C -labeled carbohydrates in the present study, like those previously tested, were converted into hepatic fatty acids of obese mice in several-fold higher extent than to those of lean mice, whereas conversion into total fatty acids of the carcass was only moderately higher in the obese mice.

EVIDENCE FOR A NONABSORPTIVE ANTIHYPERCHOLESTEROLEMIC ACTION OF PHYTOSTEROLS IN THE CHICKEN. J. E. Konlande and H. Fisher (Dept. of Nutr., Rutgers Univ., New Brunswick, N.J.). *J. Nutr.* 98, 435-42 (1969). Soy sterols were administered orally (1% of diet) and subcutaneously (20 mg/day) to chicks with hypercholesterolemia induced by feeding a low protein, cholesterol-containing diet (8% protein, 5% medium-chain triglycerides (MCT) and 0.5% cholesterol). Sterol levels and patterns in plasma, tissues and excreta were determined by spectrophotometric and gas-liquid chromatographic analysis. Strong evidence for a nonabsorptive antihypercholesterolemic action of soy sterols included: similar liver cholesterol reductions for both oral and subcutaneous soy sterol administration, and reduction by subcutaneous soy sterol injection of endogenous hypercholesterolemia due to ova resorption induced by feeding 0.04% Nicarbazine to laying hens. A comparison of the oral administration of 1% soy sterols and 1% wheat germ sterols to chicks given a hypercholesterolemic diet (25% whole egg powder) resulted in a greater antihypercholesterolemic response from soy sterols which contain more campesterol (36%) than from wheat germ sterols (25% campesterol). Campesterol appears to be the major active component of

soy and wheat sterols in relation to their antihypercholesterolemic activity.

RELATIONSHIP OF PEARL MILLET TO MILK FAT DEPRESSION IN DAIRY COWS. I. CATION FERTILIZATION. J. P. Harner, R. W. Hemken and J. H. Vandersall (Dairy Science Dept.) and N. A. Clark and B. A. Schneider (Agronomy Dept., Univ. of Maryland, College Park). *J. Dairy Sci.* 52, 1244-52 (1969). Pearl millet soilages grown with either a high or low level of calcium plus potassium fertilizer were compared with Sudangrass soilage grown with a high level of fertilization by feeding lactating dairy cows. Either 12 or 15 cows were used in each of three years to study the effect of the treatments on milk production, milk composition and rumen volatile fatty acids. During two of the three years the heavily fertilized pearl millet soilage caused a significant reduction of milk fat test of -0.33 and -0.45% as compared with unfertilized pearl millet depression of -0.19 and -0.23%. The depressions in fat tests were associated with a decrease in the molar percentage of acetic and butyric acids and an increase in the propionic acid. Depressions in fat tests have ranged from -0.09 to -0.79 percentage units for cows receiving pearl millet over an eight-year period and the correlation of the amount of rainfall and the fat test change was $r = +0.64$ ($P < 0.05$). Increased levels of calcium and potassium fertilizer and periods of low soil moisture both are associated with more severe fat depression when pearl millet is fed to lactating cows.

HYDROLYSIS AND ABSORPTION OF MEDIUM-CHAIN TRIGLYCERIDES AND SOY OIL. L. M. Hagerman, R. W. Harkins and H. P. Sarett (Dept. of Nutr. Res., Mead Johnson Res. Center, Evansville, Ind. 47721). *Proc. Soc. Exp. Biol. Med.* 131, 1028-1033 (1969). Studies were conducted on *in vitro* hydrolysis of MCT and soy oil emulsified in gum arabic and Tween 80 solutions by pancreatic lipase and *in vivo* hydrolysis and absorption of these emulsions in normal and washed isolated sections of the small intestine of rats. When instilled into the normal intestine, emulsions of MCT in gum arabic were more rapidly hydrolyzed and more completely absorbed than those of soy oil. Absorption of both oils from gum arabic emulsions was somewhat less in the washed intestine than in the normal intestine. However, Tween 80 almost completely inhibited hydrolysis and absorption of both oils from the washed intestine, suggesting that intraluminal hydrolysis of MCT is necessary for rapid absorption of this fat.

CRYSTALLINE CHOLESTEROL: EFFECT ON SERUM CHOLESTEROL LEVELS IN PATIENTS WITH HYPERLIPIDEMIA. J. H. Goldie, A. Simmons and J. A. Little (Depts. of Med. and Dietetics, St. Michael's Hosp., Univ. of Toronto, Toronto, Ontario). *Am. J. Clin. Nutr.* 22, 710-715 (1969). The effect of adding pure cholesterol to a formula diet with corn oil (35% of calories), as the only fat source, was studied in five patients with hyperlipidemia. It was found that 1000 mg cholesterol/day significantly interfered with the cholesterol-lowering effect of the diet. In spite of the ingestion of large amounts of corn oil by the hyperlipidemic subjects, the effect of the dietary cholesterol on serum cholesterol conformed with the predictive formula described by Keys and co-workers.

MYOCARDIAL METABOLISM IV. METABOLISM OF FREE AND ESTERIFIED CHOLESTEROL BY THE PERFUSED RAT HEART AND HOMOGENATES. S. L. Gartner and G. V. Vahouny (Dept. of Biochem., School of Med., The George Washington Univ., Wash. D.C. 20005). *Proc. Soc. Exp. Biol. Med.* 131, 994-999 (1969). Hepatic and myocardial cholesterol biosynthesis was investigated using mevalonic acid-2-¹⁴C lactone as precursor. Under conditions of optimal biosynthesis of cholesterol by liver homogenates, there was no detectable labelling in digitonin-precipitable sterols in heart homogenates. The present studies suggest that the low levels of myocardial cholesterol metabolism are probably related to structural requirements of this tissue and that the heart plays little or no role in regulating circulating sterol levels.

EFFECT OF DELAYED MATURITY AND CARCASS FAT ON REPRODUCTIVE PERFORMANCE OF BROILER PULLETS. H. L. Fuller, D. K. Potter and W. Kirkland (Dept. of Poultry Sci., Univ. of Georgia, Athens, Georgia). *Poultry Sci.* 48, 801-09 (1969). In an attempt to separate the effects of obesity from that of age at maturity, White Plymouth Rock pullets were subjected to restricted energy intake, decreasing daylength, or both during the growing period. The controls were full-fed

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and received increasing daylength. In two experiments, restricting the energy intake by one-third, reduced carcass fat and increased the age at maturity. This treatment also resulted in significantly more total eggs and fewer un-settable eggs thus greatly increasing the number of settable eggs per hen in both trials. This advantage was manifested by fewer small eggs and fewer oversized or double-yolked eggs early in the laying period. Decreasing daylength also delayed maturity, but pullets delayed in this manner were fatter at maturity than those receiving increasing daylength within each feeding treatment. The advantages of this method of delay were considerably less for pullets housed in the summer than for those housed in the winter, which was interpreted as a relationship between obesity and the effects of temperature or season. For winter-housed pullets decreasing daylengths resulted in greater total egg production than that of the increasing daylength groups within each feeding treatment. Thus, the advantages of decreasing daylength were additive to those of restricted energy in the one trial where the combination was tested. Average egg weight was shown not to be a good measure of settable egg production.

THE INDUCTION OF NEUROGENIC HYPERCHOLESTEREMIA. M. Friedman, S. O. Byers and S. R. Elek (Harold Brunn Inst., Mt. Zion Hosp. and Med. Center, San Francisco, Cal. 94115). *Proc. Soc. Exp. Biol. Med.* 131, 759-762 (1969). A chronic hypercholesteremia almost invariably results in the rat after placement of an electrolytic lesion which involves the fornix, medial part of the lateral hypothalamus and one of the two medial nuclei of the hypothalamus. This hypercholesteremia is not accompanied by any discernible changes in plasma triglyceride.

CONTROL OF GLUCONEOGENESIS IN LIVER. IV. DIFFERENTIAL EFFECTS OF FATTY ACIDS AND GLUCAGON ON KETOGENESIS AND GLUCONEOGENESIS IN THE PERFUSED RAT LIVER. J. G. Exton, J. G. Corbin and C. R. Park (Dept. of Physiol., Vanderbilt Univ., Nashville, Tenn. 36203). *J. Biol. Chem.* 244, 4095-4102 (1969). The possible roles of fatty acids in the regulation of gluconeogenesis and in the gluconeogenic action of glucagon were investigated in the isolated perfused rat liver. It is concluded that fatty acids probably do not play a physiological role in the rapid regulation of hepatic gluconeogenesis, and that glucagon does not stimulate gluconeogenesis by activating hepatic lipolysis. The physiological regulation of ketogenesis appears to involve primarily the control of fatty acid supply to the liver, i.e., the regulation of lipolysis in fat tissue. Regulation by glucagon at the hepatic level is of minor importance.

STERIODS IN NEWBORNS AND INFANTS. HYDROXYLATED CHOLESTEROL DERIVATIVES IN THE STEROID MONOSULPHATE FRACTION FROM MECONIUM. P. Eneroth and J.-A. Gustafsson (Dept. Chem. and Dept. Germfree Res., Karolinska Institutet, S-10401 Stockholm 60, Sweden). *FEBS Letters* 3, 129-132 (1969). In a series of publications from this laboratory the nature of sterol and steroid hormone metabolites in feces from

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man and animals has been described. From studies in germ-free and conventional rats the profound influence of intestinal bacteria on steroid metabolism has been demonstrated. It has also been shown that in man sterol metabolizing microorganisms in the intestinal tract become active within the first year of life and that the pattern of steroid hormone metabolites excreted in feces undergoes marked changes during the first six months after birth. In connection with these studies an investigation of the steroid composition in meconium was initiated. During this study a great number of C_{19} and C_{21} steroids and considerable amounts of monohydroxy ($C_{27}O_1$) and dihydroxy ($C_{27}O_2$) sterols have been found in the steroid monosulphate fraction. This communication deals primarily with the identification of the dihydroxy compounds.

STUDIES OF A PHOSPHOLIPID-REQUIRING BACTERIAL ENZYME. I. PURIFICATION AND PROPERTIES OF URIDINE DIPHOSPHATE GALACTOSE:LIPOPOLYSACCHARIDE α -3-GALACTOSYL TRANSFERASE. A. Endo and L. Rothfield (Dept. of Molecular Biol., Albert Einstein College of Med., Bronx, N.Y. 10461). *Biochemistry* 8, 3500-07 (1969). The uridine diphosphate galactose: lipopolysaccharide α -3-galactosyl transferase of *Salmonella typhimurium* requires phospholipid for activity and catalyzes one of the reactions involved in biosynthesis of the cell envelope lipopolysaccharide of the organism. The enzyme has now been purified approximately 6000-fold. The purified enzyme required phospholipid for activity and was inhibited in a competitive manner by closely related nonsubstrate lipopolysaccharides. Extraction of the purified enzyme with organic solvents removed a bound lipid-soluble component, which contained no phosphorus and is still unidentified. This component (lipid Y) caused aggregation of the enzyme, but no role of the lipid in the enzyme reaction itself has yet been established.

II. THE ROLE OF PHOSPHOLIPID IN THE URIDINE DIPHOSPHATE GALACTOSE:LIPOPOLYSACCHARIDE α -3-GALACTOSYL TRANSFERASE REACTION. *Ibid.* 3508-15. Phospholipid is required as a cofactor in the reaction catalyzed by uridine diphosphate galactose:lipopolysaccharide α -3-galactosyl transferase. The technique of equilibrium density gradient centrifugation was used to determine the ability of different phospholipids to form binary complexes with lipopolysaccharide or with enzyme. Certain phospholipids formed binary complexes with lipopolysaccharide but not with enzyme; others formed binary complexes with enzyme but not with lipopolysaccharide. Both of these types of phospholipid were inactive in the enzyme system. In contrast, phospholipids which were able to form binary complexes both with lipopolysaccharide and with enzyme were active. The data suggest a model in which the enzyme reaction involves binding of phospholipid to both the lipopolysaccharide substrate and to the enzyme protein.

A SIMPLIFIED HEMOLYSIS TEST FOR VITAMIN E DEFICIENCY. H. H. Draper and A. Saari Csallany (Div. of Nutritional Biochem., Dept. of Animal Sci., Univ. of Ill., Urbana, Ill.). *J. Nutr.* 98, 390-94 (1969). A bioassay procedure for vitamin E is described in which inhibition of spontaneous hemolysis of erythrocytes incubated in isotonic saline-phosphate buffer at 37C serves as the criterion of response. The method offers advantages over the conventional dialuric acid-induced hemolysis test with respect to simplicity, sensitivity and reproducibility.

A CONTROLLED CLINICAL TRIAL OF A DIET HIGH IN UNSATURATED FAT IN PREVENTING COMPLICATIONS OF ATHEROSCLEROSIS. S. Dayton, M. L. Pearce, S. Hashimoto, W. J. Dixon and U. Tomiyasu. *Circulation* 40, II, 1-63 (1969). The control diet was similar to the regular institutional diet, which is a standard American diet. The experimental diet provided 38.9% of calories as fat, with an iodine value of 102.4, and had a cholesterol content of 146 mg/1,000 calories (365 mg/day). Linoleic acid content of the two diets was 10% and 38% of total fatty acid, respectively. The experimental diet induced a prompt drop in serum cholesterol level and sustained a difference between the experimental and control groups amounting to 12.7% of the starting level. The difference in the primary end point of the study—sudden death or myocardial infarction—was not statistically significant.

STANDARDS OF SUBCUTANEOUS FAT APPLIED TO PERCENTILE NORMS FOR ELEMENTARY SCHOOL CHILDREN. C. B. Corbin (Dept. of Health and Physical Educ., Texas A & M Univ.,

Exhibitors



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College Station, Texas). *Am. J. Clin. Nutr.* 22, 836-841 (1969). The purpose of this investigation was to develop standards of subcutaneous fat (skin fold for American children, specifically the children of College Station, Texas, elementary schools). Subscapular and triceps skin-fold measures were taken on 1,176 elementary school boys and girls in the College Station school system in an effort to develop percentile standards for skin-fold measures for elementary school children, grades 1-6. As expected, the results indicate that fat levels increase with the age of the children and that girls possess more fat than boys at all elementary school ages.

ACETYL CoA CARBOXYLASE AND FATTY ACID SYNTHETASE ACTIVITIES IN LIVER AND ADIPOSE TISSUE OF MEAL-FED RATS. Krishna Chakrabarty and G. A. Leveille (Div. Nutr. Biochem., Dept. Animal Sci., Univ. Ill., Urbana, Ill. 61801). *Proc. Soc. Exp. Biol. Med.* 131, 1051-4 (1969). The activities of acetyl CoA carboxylase and fatty acid synthetase in liver and adipose tissue of meal-eating (limited to a single, daily, 2-hr meal) and nibbling (*ad libitum*-fed) rats were determined. The activities of both enzymes were significantly higher in adipose tissue but not liver of meal-eating as compared to nibbling animals. This observation is in accord with the concept that adipose tissue is the primary site of fatty acid biosynthesis and the major site of the lipogenic adaptive changes induced by meal-feeding. The activities of both enzymes were found to be of the same order of magnitude in liver and adipose tissue. In adipose tissue the activity of acetyl CoA carboxylase was actually greater than that of fatty acid synthetase. The significance of this observation with regard to the regulatory role of acetyl CoA carboxylase in fatty acid biosynthesis is discussed.

EFFECTS OF CORN OIL AND LYSINE ON GROWTH, FATTY ACID COMPOSITION AND PALATABILITY OF LARGE BROAD WHITE TURKEYS. C. W. Carlson, E. Guenther, K. C. Schneider, L. P. Guild, D. Deethardt and Y. A. Greichus (South Dakota State Univ., Brookings, S.D.). *Poultry Sci.* 48, 1027-33 (1969). Large Broad White turkey toms were grown to 24 weeks of age on typical corn-soy diets with the following treatments from 12 weeks: basal, 4% corn oil, 0.1% L-lysine and corn oil plus lysine. In the first experiment all treatments showed significant growth increases but these differences were not observed in the second experiment. Representative birds of each treatment were slaughtered, sawed into halves with one half for initial palatability observations and the other half retained in storage at -10C for similar observations after six months. No marked and consistent effects were noted by the taste panel for flavor, tenderness or juiciness, however the panel did prefer the basal and lysine-fed turkeys. Significant increases in shear press values were noted for the corn oil-fed turkeys. The corn oil treatments resulted in a 25% relative increase in linoleic acid content of carcass fatty acids with concomitant decreases in the palmitic, palmitoleic

(Continued on page 700A)

• New Members . . .

(Continued from page 658A)

Jacqueline T. Terranova, Graduate Student, Louisiana State University in New Orleans, New Orleans, La.
Larry W. Wilson, Student, University of Georgia, Athens, Georgia.

Corporate Associate

Extrin Flavors, Inc., Grant M. Sweet, President, Long Island City, N.Y.
Sweco, Inc., J. P. Miller, Mgr., Process & Development Engineering, Los Angeles, California.

• Glycerine Analysis Subcommittee Report . . .

(Continued from page 651A)

to have the official U.S. method differ from the ISO procedure since international trading might be affected.

2. Primary Standard for Caustic

The committee feels that potassium acid phthalate is the best primary standard for caustic solutions. The approach being considered by ISO, although basically sound, is quite involved and therefore subject to errors. It has been suggested that any reference to standardization of caustic could be worded to permit use of any reliable standard. This might be the best way to resolve the differences.

3. Ash

The ISO proposal that the ash on crudes be obtained by ashing at 750 C for 10 min has not been generally accepted by the committee. Most members feel that further testing of this procedure must be made before any decision can be made. One member suggested that the use of sulfated ash be considered.

MONG

The committee is divided on acceptance of the MONG approach. Some members object to the fact that it gives organic matter by difference. It is generally agreed that both the current AOCS and the MONG procedures have drawbacks. The AOCS approach is not attractive because it is a time-consuming method. It also suffers from the fact that losses may occur in the heating step (at 175 C) if the alkalinity or acidity is not adjusted carefully. The MONG procedure is quite simple since only total glycerine, ash and water need to be measured, the balance being designated MONG. It is useful therefore as a practical procedure, and results obtained, although empirical, should be as valid as those obtained by the AOCS method. Any decision on the MONG question must await further ISO action.

Arsenic

The committee believes that the colorimetric silver diethyldithiocarbamate method is the best method for the determination of arsenic. The ISO committee is still debating on this matter and we must await developments until their next meeting.

R. J. Houle
C. F. Smullen
S. P. Smock
E. K. Schultz
N. C. Schultze
T. M. Brye
R. Houston
R. M. Kelley, Chairman

ABSTRACTS: BIOCHEMISTRY AND NUTRITION

(Continued from page 696A)

and oleic acid fractions. This demonstrates once again the ease with which such changes in carcass fatty acid compositions can be made.

THE EFFECTS OF NUTRITIONAL AND HORMONAL FACTORS ON THE FATTY ACID SYNTHETASE LEVEL OF RAT LIVER. D. N. Burton, Janet M. Collins, A. L. Kennan and J. W. Porter (Lipid Metabolism Lab., Veterans Admin. Hosp., and the Depts. of Physiolog. Chem. and Gynecol. and Obstetrics, Univ. of Wisc., Madison). *J. Biol. Chem.* 244, 4510-16 (1969). The rise in fatty acid synthesizing capacity of the liver which is observed on realimentation of fasting rats with a fat-free diet has been studied. The rise to supranormal levels has been shown unequivocally to be the result of adaptive enzyme synthesis. Proof of this was obtained through the demonstration that ¹⁴C-leucine is readily incorporated *in vivo* into the purified fatty acid synthetase complex formed during refeeding. In addition, measurements of the absolute levels of the enzyme in the livers of fasting and refeed rats showed drastic changes in the content of this protein complex with changes in nutritional status. Similarly, alloxan diabetic and portacaval-shunted rats exhibited lowered levels of fatty acid synthetase in liver; and insulin treatment of alloxan diabetic animals caused a return of the level of this enzyme toward the normal range. The characteristics of the fatty acid synthetase isolated from the livers of rats refeed for varying periods of time after fasting have been investigated. Differences in homogeneity and variation in specific enzyme activity of these preparations were found which correlated with the length of time of refeeding.

HUMAN ADIPOSE TISSUE COMPOSITION AND AGE. G. L. Baker (Dept. of Pediatrics, Univ. of Iowa, Iowa City, Iowa 52240). *Am. J. Clin. Nutr.* 22, 829-835 (1969). Adipose tissue specimens obtained at necropsy from individuals ranging from birth to 86 years of age were analyzed for water, lipid, nitrogen, DNA and fatty acids. Both subcutaneous and perirenal samples were studied in each instance. The water, lipid, nitrogen and DNA content as well as the fatty acid proportions are age related. The lipid content of adipose tissue accounted for only about 40% of adipose tissue weight in the newborn period and increased with increasing age to 75% in the adult. Concentrations of water, nitrogen and DNA declined with age.

EFFECT OF FEEDING AND WITHDRAWAL OF MENHADEN OIL ON THE ω 3 AND ω 6 FATTY ACID CONTENT OF BROILER TISSUES. D. Miller, K. C. Leong and P. Smith Jr. (Bur. of Comm. Fish. Technol. Lab., College Park, Maryland 20740). *J. Food Sci.* 34, 136-141 (1969). Analysis was made of the fatty acid composition of liver, adipose fat, thigh and breast muscles of broilers fed corn-soy commercial-type of diets containing one of two levels of fish oil (2.5 or 5.0%). The oil was subsequently continued, withdrawn or replaced with yellow grease 2, 3 or 4 weeks before termination of the experiment at the 8th week. The tissue contents of four ω -3-type fatty acids (20:4, 22:5 and 22:6) were increased in relation to the number of weeks menhaden fish oil was included in the diet and were significantly correlated to organoleptic scores. Liver had highest total content of the ω -3 fatty acids; the adipose fat, the least; the muscles, intermediate.

TUMOR LIPIDS: METABOLIC RELATIONSHIPS DERIVED FROM STRUCTURAL ANALYSES OF ACYL, ALKYL, AND ALK-1-ENYL MOIETIES OF NEUTRAL GLYCERIDES AND PHOSPHOGLYCERIDES. R. Wood and F. Snyder (Oak Ridge Assoc. Univ., Oak Ridge, Tenn.). *Arch. Biochem. Biophys.* 131, 478-94 (1969). The composition of the hydrocarbon moieties of the 1-, 2- and 3-positions of triglycerides and glyceryl ether diesters (GEDE) and of the 1- and 2-positions of diacyl and alkyl acyl phosphatidyl cholines (PC) and diacyl, alkyl acyl and alk-1-enyl acyl phosphatidyl ethanolamines (PE) from Ehrlich ascites cells (EAC) were determined. The carbon number percentage distribution of triglycerides, GEDE, and the diglyceride-type acetates derived from each class of PC and PE was also determined by GLC of the intact lipids. The fatty acid compositions of the 1-, 2-, and 3-positions of the triglycerides are different and also differ from the composition of the corresponding positions of the GEDE, which are not the same. Both tri-

glycerides and GEDE exhibit a 1-random-2-random-3-random type of distribution. The choline-containing phosphatides consist of 66% diacyl PC and 33% alkyl acyl PC. Ethanolamine-containing phosphatides are composed of 55% diacyl PE, 30% alkyl acyl PE, and 15% alk-1-enyl acyl PE. The carbon chain of the 1-position of each PC and PE class is predominantly saturated and the 2-position is dominated by polyunsaturated acids. All PC and PE classes except the alk-1-enyl acyl PE show a 1-random-2-random distribution. The similarities in composition at both the 1- and 2-positions between triglycerides and diacyl PC and between GEDE and alkyl acyl PC suggest a loss of acyl CoA:lysophosphatide acyl transferase enzymes which are present in normal tissue. Metabolic relationships derived from structural analyses of acyl, alkyl, and alk-1-enyl moieties of glycerides and phosphoglycerides and from established pathways for acylation reaction led to the proposal of a metabolic pathway for the biosynthesis of lipids containing glyceryl ethers.

STRUCTURAL STUDIES OF NEUTRAL GLYCERIDES AND PHOSPHOGLYCERIDES OF RAT LIVER. R. Wood and R. D. Harlow (Oak Ridge Assoc. Univ., Oak Ridge, Tenn.). *Arch. Biochem. Biophys.* 131, 495-501 (1969). The distribution of fatty acids esterified at each position of glycerol in triglycerides (TG), phosphatidyl choline (PC), and phosphatidyl ethanolamine (PE) of rat liver was determined. The carbon number distribution of TG and diglyceride acetates derived from PC and PE was also determined. Each of the TG positions showed a distinct distribution of fatty acids that was not arranged randomly, indicating pairing of some acids. The composition of the fatty acids esterified at the 1-position of PC and PE was the same, but they differed from that of TG; the 2-position differed in all three classes. The carbon number distribution of PC diglyceride acetates was different from that of PE diglyceride acetates and neither agreed with the random distribution values calculated from the analyzed compositions of the 1- and 2-positions of each class, indicating preferential pairing of some acids in these lipid classes also. Selectivity of diglycerides used for the biosynthesis of TG or phospholipids or both is substantiated by the lack of agreement between the TG carbon number distribution determined experimentally and that calculated from values of PC or PE diglyceride acetates plus the values of the 3-position of the TG. These findings for rat liver are the opposite of those reported in a companion paper in which tumor cells did not show selectivity of diglycerides used for TG and PC biosynthesis.

THE EFFECT OF CLOFIBRATE ON THE SERUM TRIGLYCERIDE CONCENTRATION IN NORMAL MALES FED HIGH-SUCROSE DIETS. D. Zakim and R. H. Herman (Metabolic Div., Fitzsimons Gen. Hosp., Denver 80240). *J. Atherosclerosis Res.* 10, 91-95 (1969). The effects of clofibrate administration on the serum triglyceride response to high caloric sucrose diets has been investigated in normal males. In each subject the sucrose diet increased the serum triglyceride concentration. Clofibrate treatment of normal individuals eating an *ad libitum* diet did not decrease the serum triglyceride concentration. Clofibrate therapy did not block the hypertriglyceridemic response to the sucrose diet.

ACTIVITY OF SELECTED GLUCONEOGENIC AND LIPOGENIC ENZYMES IN BOVINE RUMEN MUCOSA, LIVER AND ADIPOSE TISSUE. J. W. Young, S. L. Thorp and J. Z. DeLumen (Dept. of Animal Sci., Iowa State Univ., Ames, Iowa 50010). *Biochem. J.* 114, 83-88 (1969). The activities of phosphoenolpyruvate carboxykinase, 'malic enzyme,' citrate-cleavage enzyme and glucose 6-phosphate dehydrogenase were assayed in homogenates of rumen mucosa, liver and adipose tissue of cattle. Rumen mucosa cytoplasm contained activities of 'malic enzyme' approximately sevenfold those of phosphoenolpyruvate carboxykinase, suggesting that the conversion of propionate into lactate by rumen mucosa involves 'malic enzyme.' Neither starvation for 8 days nor feeding with a concentrated diet for at least 3 months before slaughter produced enzyme patterns in the tissues different from those in cattle given only hay, except that the all-concentrate diet caused increased activities of glucose 6-phosphate dehydrogenase and 'malic enzyme' in adipose tissues. Rumen mucosa, liver and adipose tissue contained phosphoenolpyruvate carboxykinase activity. 'Malic enzyme' was absent in liver. Citrate-cleavage enzyme activity was present in liver and adipose tissue but was quite low in rumen mucosa. Liver contained much less glucose 6-phosphate dehydrogenase activity than rumen mucosa or adipose tissue.

• Fatty Acid Derivatives

THE BASE-CATALYSED REARRANGEMENT OF VERNOLIC AND OTHER EPOXY ESTERS: THE PARTIAL SYNTHESIS OF METHYL CORIOLATE, METHYL DIMORPHECOLATE, AND SOME CONJUGATED POLYENOIC ESTERS BY A POSSIBLE BIOSYNTHETIC ROUTE. H. B. S. Conacher and R. D. Gunstone (Dept. of Chem., Univ. of St. Andrews, Purdie Bldg., St. Andrews, Scotland). *Chem. Phys. Lipids* 3, 191-202 (1969). Methyl vernolate is rearranged to methyl coriolate by lithium diethylamide at 0°C in about 60% yield. Dehydration of this hydroxy ester yields a mixture of octadecatrienoates. By similar reactions linoleic ester has been converted to racemic coriolic ester and to racemic dimorphecolic ester, and linolenic ester to parinaric ester.

THE REARRANGEMENT OF METHYL 12,13-EPOXYOLEATE BY BORON TRIFLUORIDE WITH FORMATION OF CYCLOPROPANE ESTERS. *Ibid.*, 203-220. Treated with boron trifluoride etherate in dioxan solution, methyl 12,13-epoxyoleate gives the 12- and 13-oxo-oleates as major products. These are accompanied by cyclopropane compounds (8-11%) which are mainly the *cis* and *trans* isomers of methyl 9,10-methylene-12-oxo-heptadecanoate. In benzene solution the cyclopropanes are formed in higher yield (38%). The structure of long-chain cyclopropane esters can be determined by oxidation with chromic acid.

THE MODIFIED SYNTHESIS OF PHOSPHATIDYL ETHANOLAMINE. I. Barzilay and Y. Lapidot (Dept. Biol. Chem., Hebrew Univ., Jerusalem). *Chem. Phys. Lipids* 3, 280-285 (1969). The synthesis of phosphatidylethanolamine is described. The method involves a condensation of dipalmitoylglycerolphosphate with N-tertiary butyloxycarbonylethanolamine in the presence of dicyclohexylcarbodiimide. After removal of the N-blocking group, the final product is purified by Sephadex LH-20 column chromatography.

THE CONDENSATION OF AZIRIDINE WITH PHOSPHATIDIC ACID; SYNTHESIS OF O-(1,2-DIACYL-SN-GLYCERO-3-PHOSPHORYL)-ETHANOLAMINE. R. Aneja, J. S. Chadha, A. P. Davies and C. A. Rose (Unilever Lab., Frythe, Welwyn, England). *Chem. Phys. Lipids* 3, 286-291 (1969). The condensation of N-tritylaziridine with phosphotidic acid is facile and gives a good yield of N-trityl phosphotidylethanolamine which is detriptylated to a phosphatidylethanolamine. This novel route is illustrated by the synthesis of O-(1,2-dioctadecanoyl-sn-glycero-3-phosphoryl)-ethanolamine.

METHOD FOR DETERMINING THE CONTRIBUTION OF METHYL KETONES TO FLAVORS OF STERILE CONCENTRATED MILKS. C. Allen and O. W. Parks (Dairy Products Lab., USDA, Washington, D.C. 20250). *J. Dairy Sci.* 52, 1547-1551 (1969). A method for determining the concentrations of the C₅ through C₁₅ odd-carbon-numbered methyl ketones in fluid milks, based on the free C₁₃ methyl ketone content and the methyl ketone potential remaining in the fat phase of the product, is presented. Application of the procedure to samples of commercial evaporated milk led to the conclusion that the role of methyl ketones in the off-flavor of this product is dependent on total methyl ketone potential of the milk fat, composition of the methyl ketone potential (especially the heptanone-2 potential), and degree of hydrolysis and decarboxylation of β-keto acids as determined by heat treatment and storage conditions.

SYNTHESIS OF WAXES BY THE GUERBET REACTION. W. Strassberger (Farbwerke Hoechst AG, Frankfurt/M.-Hoechst, W. Ger.). *Fette Seifen Anstrichmittel* 71, 215-217 (1969). Branched chain waxes possess better liquefying properties and superior solubility in organic solvents than straight chain waxes. Branched chain waxes having both of the above properties were therefore synthesized by a new principle, namely the Guerbet reaction. Guerbet alcohols, based on technical octadecyl alcohol were esterified with various mono and dicarboxylic acids. Strongly branched and relatively low melting waxes were thus obtained which in spite of their greater solubility in organic solvents, exhibit insignificant liquefying effect in wax pastes. As against these, when the Guerbet alcohols from stearyl alcohol were oxidized, branched chain wax alcohols are obtained which in native as well as saponified state show liquefying action on wax pastes. The Guerbet reaction is explained taking the example of a mixture of two primary alcohols.

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• Drying Oils and Paints

THE EXTENT OF SUBSTITUTION OF LINSEED OIL BY DEHYDRATED CASTOR OIL AND THE PROBLEM OF LOST ACIDITY IN THE MALEINIZATION REACTION. N. A. Ghanem and A. M. M. Nasser (Lab. of Polymers and Pigments, Nat. Res. Center, Dokki, Cairo, Egypt). *Farbe Lack* 75, 419-430 (1969). The extent to which dehydrated castor oil (DCO) produced by a new dehydration method, can be introduced in a reaction mixture of linseed oil and maleic anhydride is examined. By careful control of the reaction temperature and duration, a mixture of linseed oil and DCO containing up to 50% by volume of the latter gives fairly viscous, water-soluble product when reacted with 20% of the total weight of maleic anhydride and neutralized with ammonia. Any further elevation of the DCO ratio causes gel formation. The material has considerably better properties than the straight linseed maleinized product regarding the color, stability of the adduct, drying time and the water resistance. Its use in surface coatings of various types is satisfactory. Viscosity reduction curves using ethyl alcohol show optimum solid contents and consistency at 15-25% alcohol content. The maleinization reaction is followed by the determination of the product's acid value and the amount of CO₂ evolved, and the sum of the two values in mg KOH is compared with the theoretical acid value. A loss in acidity is noted. The work was conducted on model compounds; pure fatty acids of different saturations and their simple esters. The theoretical acidity of the reaction product is equal to the experimental acidity plus twice the amount of CO₂ evolved. This finding is explained in terms of the different reactivities of the fatty chains.

DIFFERENTIAL THERMAL ANALYSIS OF ESTERS OF SATURATED FATTY ACIDS WITH PENTAERYTHRITOL. M.-T. Richert (I.R.H.O., Paris) and C. Paquot (Cent. Nat. de la Rech. Scientifique, Thiais, France). *Oléagineux* 24, 413-416 (1969). The different crystalline forms of the mono-, di-, and tetraesters of lauric and palmitic acids with pentaerythritol were investigated. Using differential thermal analysis equipment of their own design, melting points of the different crystalline forms of the esters were determined and thermal behavior described during transitions from one form to another. The esters were prepared non-catalytically in the presence of phenol, and the resulting mixture was separated on a silicic acid column. Pure esters were thus obtained.

IDENTIFICATION AND SEMI-QUANTITATIVE ANALYSIS OF VARNISH OILS BY PAPER CHROMATOGRAPHY. I. Prazák. *Chem. Průmysl* 18(43), 487-91 (1968). A reversed-phase paper chromatography was used for separation of oils. The best results were obtained by impregnating paper with a high-boiling fraction of petroleum ether or paraffin oil. The mobile phase was acetic acid with various amounts of water, aqueous alcohols and acetonitrile. The oils were saponified and fatty acids were used dissolved in benzene. Iodine vapours or alkaline solution of KMnO₄ were used for detection. Photometric methods were used for quantitative estimates. (World Surface Coat. Abs. No. 325)

ALKYD RESINS. I—VEGETABLE OILS FOR ALKYDS. J. Staněk, K. Hájek, J. Huml and J. Hires. *Plaste u. Kautschuk* 15, 372-5 (1968). The world supply position of the different oils for alkyds is examined. (World Surface Coat. Abs. No. 325)

VINYLACTION OF FATTY SUBSTANCES. D. D. Taft (Ashland Oil & Refining Co.). *U.S.* 3,453,224. A process is described for producing vinylated fatty substances such as styrenated alkyd resins, by contacting at 320-450F an unsaturated fatty substance free of terminal unsaturation having more than 8 C atoms with a vinyl monomer such as styrene. A number of examples are given.

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CATALYTIC ISOMERIZATION OF SAFFLOWER OIL. D. Swaramaiah and B. S. Kulkarni (Dept. Chem. Technol., Osmania Univ., Hyderabad, India). *Paintindia* 19, No. 6, 19-25 (1969). The isomerization of safflower oil to increase conjugation was achieved to the extent of 27.4% using an anthraquinone catalyst at a level of 5% and at 250C. The characteristics of the isomerized product were comparable to dehydrated castor oil. Using IR, one sample was found to have a 28.2% transformation of *cis* to *trans* isomer. Bodying rates and varnish production were comparable to dehydrated castor oil.

• Detergents

A NEW REDEPOSITION TEST METHOD. S. Shimauchi, R. Tsuzuki and H. Mizushima (Teijin Ltd., Osaka, Japan). *Am. Dyestuff Rept.* 58(16), 15-9 (1969). The development of a standard soil redeposition test method having good correlation with actual laundering results is described. The conclusion was reached that the redeposition of artificial soils based on carbon black is far different from that observed with natural soils. The correlation is considerably improved, however, by the inclusion of albumin in the artificial soil, intended to simulate the protein content normally encountered in natural soils.

NON-CAKING SODIUM TRIPOLYPHOSPHATE. R. J. Fuchs (FMC Corp.). *U.S.* 3,446,580. A compacted sodium tripolyphosphate having a specified density and particle size and having distributed on it 2.5 to 7% of water is provided. The presence of the specified amount of water on this kind of sodium tripolyphosphate renders it capable of dissolving readily in water without caking.

STRAIGHT-CHAIN ALKYL ARYL SULFONATE DETERGENT COMPOSITIONS. D. M. Marquis (Chevron Res. Co.). *U.S.* 3,446,743. A process for suppressing the caking tendencies of straight-chain sodium alkyl benzene sulfonate detergents comprises dispersing through the detergent an effective amount of a sodium or potassium salt of a phthalic acid such as ortho-, iso- or terephthalic. The amount of anticaking agent used

(Continued on page 704A)

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ABSTRACTS: DETERGENTS

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is in the range of 2-25%, by wt., based on the detergent. These anticaking properties are particularly effective in built particulate detergent compositions in which an inorganic sodium salt detergent builder is combined with the straight-chain sodium alkyl benzene sulfonate.

HYDROTROPIC SUBSTANCES FOR THE PREPARATION OF LIQUID WASHING AGENTS. H. Stache (Huls Chem. Plant, Marl-Huls, Ger.). *Fette Seifen Anstrichmittel* 71, 381-386 (1969). The hydrotropic action of short chain alkyl benzenesulfonates was investigated on anion active and non-ionic raw materials for detergents as well as on their combinations. In the combinations studied, it was found that alkali salts of cumol sulfonate were the best hydrotropic agents. The solubility of the raw materials for washing agents could be improved such that concentrated charges with a clear-melting point could be prepared. The salting-out effect of inorganic salts on aqueous solutions of raw materials for detergents is prevented by alkali-cumol sulfonate.

INVESTIGATIONS ON THE EFFICACY OF PRESERVING AGENTS IN ANION ACTIVE SURFACTANTS. II. G. Shuster and H. Modde (Grunau Res. Lab., Illertissen, Ger.). *Fette Seifen Anstrichmittel* 71, 394-399 (1969). The preserving action of p-

chloro-m-cresol and p-hydroxybenzoic acid methyl/ethyl ester combination (1+1) in solutions of anion active surfactants was investigated. The preservatives were employed at two different concentrations. The concentration of the surfactants (lauryl sulfate and protein-fatty acid condensate as sodium, potassium and triethanolamine salts) was varied from 0.1 to 10.0g/100g. The pH of the solutions was 7. The deactivation of the preservatives investigated here by the anion active surfactant occurs only after a certain ratio of surfactant to preservative is reached. Under this ratio, the preservatives are fully effective; above this, however, the preservatives are deactivated.

INVESTIGATIONS ON THE ACTIVITY OF PROTEOLYTIC ENZYMES ESPECIALLY UNDER THE CONDITIONS EMPLOYED IN WASHING TECHNIQUE. H. R. Joag, Jr. (Soap. Res. Lab., Schnyder & Cie., Biel, Switzerland). *Fette Seifen Anstrichmittel* 71, 404-406 (1969). Number of methods have been suggested for determining the activity of proteases and amylases. A few of these methods are currently employed for enzymes used in washing agents. The results thus obtained do not necessarily permit an evaluation of practical value since very specific conditions prevail in the actual washing technique. A method of testing is reported here in which the conditions employed in the washing operation are especially taken into account. Furthermore, the determination of enzymes in biologically active washing agents and the tolerance of proteases toward perborate oxygen are reported.