• Fats and Oils

THE AUTOCATALYTIC FAT-SPLITTING PROCESS. F. Crespo (Inst. Nacional de Tecnol. Indust., Buenos Aires, Argentina). *Seifen-Öle-Fette-Wachse* 99(3), 55-57 (1973). The author discusses the theory of the fat-splitting process and reports trials with beef tallow. Cited is the formation of glycerin, diglycerides and monoglycerides.

CHROMATOGRAPHIC ANALYSIS OF ROSIN PART IN CANDELILLA WAX. J.C. Morales and G. Torres E. (Mexican Inst. of Technol. Invest., A.C., Anal. Central Lab., Calzada Legaria No. 694, Mexico (10) D.F.). *Seifen-Öle-Fette-Wachse* 99(1), 17-20 (1973). Qualitative and quantitative analyses of the resinous part of candelilla wax by means of thin-layer chromatography demonstrate that 15–18% of the wax is resinous. It is composed of esterified triterpenes and tetra- and penta-cyclenes.

HEATS OF HYDROGENATION. IX. CYCLIC ACETYLENES AND SOME MISCELLANEOUS OLEFINS. R.B. Turner, A.D. Jarrett, P. Goebel and B.J. Mallon (Dept. of Chem., Rice Univ., Houston, Tx. 77001). *J. Amer. Chem. Soc.* 95, 790–2 (1973). The heats of hydrogenation in acetic acid solution at 25C of octyne-4, cyclooctyne, cyclononyne, cyclodecyne, cycloundecyne, cyclododecyne, and cyclotetradecadiyne-1,8 have been determined. Results for 16 miscellaneous olefins are also reported.

PREPARATION AND FRACTIONATION OF THE HIGH MELTING GLYCERIDE FRACTION OF MILK FAT. J.W. Sherbon and R.M. Dolby (New Zealand Dairy Res. Inst., Palmerston North, New Zealand). *J. Dairy Sci.* 56, 52–60 (1973). High melting glyceride (HMG) was prepared from anhydrous milk fat and from a commercially prepared hard fraction of a different milk fat. The latter glyceride was further fractionated by progressive crystallization from acetone. The first precipitation of HMG reduced the softening point of the supernatant fat to about 18C, primarily be eliminating fats melting above 20C. The melting properties of the supernatant to the recrystallization of HMG depended upon the source of fat. That from anhydrous milk fat had a limited melting range whereas that from the hard fraction had an extended melting range.

SOME UNUSUAL VOLATILE CARBONYL COMPONENTS OF POTATO CHIPS. R.G. Buttery (Western Reg. Res. Lab., ARS, USDA, Berkeley, Cal. 94710). *J. Agr. Food Chem.* 21, 31–33 (1973). Combined capillary gas chromatography-mass spectrometry analysis of the carbonyl fraction of the volatile oil of potato chips detected a number of unusual aldehydes which were shown to be 4-methylpent-2-enal, 4-methylhex-2-enal, 2-isopropylbut-2-enal, 2-methylmercaptomethylbut-2-enal, 2-methylmercaptomethyl-4-methylpent-2-enal, 2-phenylbut-2-enal, 2-phenyl-4-methylpent-2-enal and 2-phenyl-5-methylhex-2-enal. These compounds are probably formed in the potato chips during the frying by aldol-type condensations. Other compounds also characterized included 2-methylhexa-4,5-dione, acetophenone, hepta-trans,-trans-2,4-dienal, and octa-trans,trans-2,4-dienal.

CHARACTERIZATION OF LIPIDS FROM SEEDS OF THE ROSACEA FAMILY. T. Gutfinger, S. Romano and A. Letan (Dept. of Food Eng. Biotech., Technion-Israel Inst. of Tech., Haifa, Israel). *J. Food Sci.* 37, 938–40 (1972). Apricot and peach kernels are by-products in the canning and fruit drying industry. The oil from almond kernel is used in various cosmetic products and pharmaceuticals; also oils extracted from kernels of apricot and peach are sometimes used for the same purposes. Data are available on the fatty acid composition of the different Prunus species, but no data are available on the fatty acid composition of the Prunus species cultivated in Israel. Sterol and tocopherol composition was reported only for almond oil; no data are available on sterol and tocopherol composition of the apricot and peach oils. Recently the canning of apricots and peaches in this country increased very appreciably. It was of interest therefore to find possible uses for oils from kernels of apricots and peaches (also substitutes for almond oil). An investigation of compositions of all three oils was thus undertaken. It is also worth mentioning that the prices of apricot and peach seeds (kernels) are much lower than those of almond seeds.

SURFACE AREAS OF NATURALLY OCCURRING LIPID CLASSES AND THE QUANTITATIVE MICRODETERMINATION OF LIPIDS. L.I. Burke, Gajanan S. Patil, R.V. Panganamala, J.C. Geer and D.G. Cornwell (Depts. of Physiol. Chem. and Pathol., Ohio State Univ., Columbus, Ohio 43210). *J. Lipid Res.* 14, 9–15 (1973). The surface area (A) of a lipid was directly proportional to the amount of lipid in a surface film ($A = k$ μmoles) measured at constant surface pressure, temperature and subphase composition. A surface area of 2300 $cm^2/\mu mole$ was obtained for cholesterol isolated from human adrenal and aorta and for cholesterol from hydrolysates of cholesteryl esters isolated from the same tissues. Unsaturated methyl esters that contained from one to four cis double bonds had the same surface area, 39.4 $Å^2$/molecule. As a consequence, naturally occurring triglyceride mixtures which had similar saturated-unsaturated fatty acid ratios had the same surface area, 6090 $cm^2/\mu mole$. Naturally occurring phospholipid mixtures had the same surface area, 4590 $cm^2/\mu mole$, and it appeared that the composition of these mixtures was regulated to control the physical properties of the mixtures. Surface area was much more sensitive than colorimetric procedures for the estimation of cholesterol and triglycerides. The surface area/molecule was a criterion of purity and an expanded surface area/molecule was an indication of autoxidation. Thus, surface area measurements were valuable for both the microdetermination and the characterization of lipid classes.

KINETICS OF THE PROCESSES OF DESORPTION FROM FATTY ACID MONOLAYERS. G.S. Patil, R.H. Matthews and D.G. Cornwell (Dept. of Physiol. Chem., Ohio State Univ., Columbus, Ohio 43210). *J. Lipid Res.* 14, 26–31 (1973). The surface area, A, of contracting fatty acid monolayers was measured as a function of time, t, at constant surface pressure. In the initial temporal phase, ln A was linear with $t^{1/2}$. In a subsequent steady-state phase, ln A was linear with t. The initial de-

sorption coefficient for sodium palmitate, K_i, and the steady-state desorption coefficient, K_s, varied directly with surface pressure and subphase pH, and these desorption coefficients also varied with the composition of the subphase buffer. However, the K_s/K_i ratio was independent of these variables. The diffusion coefficient, D_{25}, for sodium palmitate calculated from desorption coefficient ratios was $4.8 \pm 0.6 \times 10^{-6}$ cm²/sec. This value was in reasonable agreement with D_{25} for sodium palmitate measured by time-lag diffusion, $3.7 \pm 0.6 \times 10^{-6}$ cm²/sec. D_{25} values obtained for a series of fatty acids suggested that higher members of the series diffused as small aggregates averaging two to four molecules in size. Kinetic and diffusion data both supported a model for the desorption process described by Ter Minassian-Saraga.

IDENTIFICATION AND QUANTITATION OF LIPID-BOUND, SHORT-CHAIN DIOLS. E. Schupp and W.J. Baumann (Univ. of Minn., The Hormel Inst., Austin, Minn. 55912). *J. Lipid Res.* 14, 121–4 (1973). Procedures are described for the hydrolysis of neutral lipid fractions containing long-chain esters and alk-1-enyl ethers of short-chain diols, and for the identification and quantitation of the constituent diols as long-chain cyclic acetals using gas-liquid chromatography in combination with mass spectrometry.

CHOLESTEROL, FAT AND PROTEIN IN DAIRY PRODUCTS. D.E. Lacroix, W.A. Mattingly, N.P. Wong and J.A. Alford (Nutr. Inst., A.R.S., U.S.D.A., Beltsville, Maryland). *J. Am. Dietetic Assoc.* 62, 275–9 (1973). Twenty-seven kinds of dairy products were analyzed for fat, cholesterol and protein content. The data showed a direct relationship between fat and cholesterol content. The correlation was better for products having a fat content greater than whole milk than it was for low fat products. Fluid whole milk was found to contain about 4 mg cholesterol per gram fat whereas higher fat dairy products contained about 3 mg per gram fat. Based on regression analysis, a chart is presented and an equation given which can be used to calculate cholesterol values in dairy products in which only the fat content is known. No relationship was observed between cholesterol and protein.

PREPARATION OF METAL SALTS OF TALL OIL. J.N. Stone (Continental Oil Co.). *U.S. 3,719,647.* Tall oil soap is reacted with an acid metal salt in an aqueous medium, and the tallate is extracted from the aqueous phase with a water immiscible organic solvent in which the metal tallate is soluble.

METHOD FOR DEFATTING SOYBEAN MEAL. K.H. Steinkraus (Cornell Res. Found., Inc.). *U.S. 3,721,569.* Organoleptically bland soybean meal is prepared by extracting ground, unheated soybeans with concentrated ethyl alcohol followed by a mixture of ethyl alcohol and chloroform. The meal, in addition to being defatted and debittered, is free of undesirable mouth-coating factor.

MILK-FREE MARGARINE. L.L. Linteris (Lever Bros.). *U.S. 3,721,570.* Milk-free margarines have little tendency to discolor when used for frying but have the undesirable attribute of a reduced salt sensation. In this disclosure, the reduction if counteracted by the presence of very small amounts of sodium caseinate.

PROCESS FOR EXTRACTING OIL FROM PALM FRUITS AND OLIVES. R.A. Couche. *U.S. 3,723,487.* The fruit is disintegrated in the presence of the extraction solvent, and the resulting slurry is passed through a multi-stage, countercurrent extraction process. The solvent used is either acetone or a mixture of ethyl alcohol, ethyl acetate and acetone in the ratio of 1:1:1 by volume, or ethyl alcohol, ethyl acetate and isopropyl ether in the ratio of 4:2:1 by volume. The temperature is maintained at 50–55C throughout the extraction process, and the water: solvent ratio is adjusted to 1:1–1:2 by weight in the first stage. The recovery of oil from the solvent occurs principally from the outgoing liquor of the first stage.

• Biochemistry and Nutrition

THE ROLE OF PHOSPHOLIPID IN SUCCINATE CYTOCHROME C REDUCTASE. Linda Yu, Chang-an Yu and T. E. King. (Dept. of Chem., State Univ. of New York at Albany, Albany, N.Y. 12222). *Biochemistry* 12, 540–6 (1973). A lipid-depleted succinate cytochrome c reductase was prepared by fractionation of succinate cytochrome c reductase in the presence of 0.5% cholate and 40% ammonium sulfate. The lipid-depleted preparation possessed the same content of cytochromes b and c_1 as the intact reductase, but showed only 40% of the succinate phenazine methosulfate activity and no succinate cytochrome c reductase activity. The depleted preparation in solution form was stable for a few hours in contrast to the intact reductase which retained full activity for at least 1 week. Upon the addition of coenzyme Q and phospholipids derived from the 40% supernatant fraction, the succinate cytochrome c activity of the depleted reductase was restored.

FEEDBACK REGULATION OF CHOLESTEROL BIOSYNTHESIS: STUDIES WITH CHOLESTYRAMINE. L.W. White (Depts. of Pharm. and Med., Case Western Reserve Univ. Schl. of Med., Cleveland, Ohio 44106). *Circulation Res.* 31, 899–907 (1972). Regulation of hepatic cholesterol biosynthesis is thought to occur at the step involving formation of mevalonate from 3-hydroxy-3-methylglutaryl-coenzyme A (HMG-CoA); this reaction is catalyzed by HMG-CoA reductase. Oral administration of cholestyramine, an agent that interferes with cholesterol reabsorption and is associated with a compensatory increase in hepatic cholesterol synthesis, was used to evaluate feedback regulation of cholesterol biosynthesis. Changes in incorporation of [^{14}C]-acetate into HMG, mevalonate and cholesterol corresponded to changes in product synthesis.

THE CATABOLISM OF TAY-SACHS GANGLIOSIDE IN RAT BRAIN LYSOSOMES. J.F. Tallman and R.O. Brady (Developmental and Metabolic Neurology Branch, Nat'l. Inst. of Neur. Diseases and Stroke, Nat'l. Inst. of Health, Bethesda, Md. 20014). *J. Biol. Chem.* 247, 7570–5 (1972). The metabolism of Tay-Sachs ganglioside, Cer-Glc-Gal-(NeuAc)-GalNAc (G_{M2}) was investigated by using G_{M2} specifically labeled with ^{3}H in the NeuAc moiety or with ^{14}C in GalNAc. There are two possible pathways for the catabolism of G_{M2} in brain, initiated via G_{M2}-sialidase or G_{M2}-hexosaminidase. The products of the sialidase reaction were identified and are Cer-Glc-Gal-GalNAc and NeuAC; the hexosaminidase yields Cer-Glc-Gal-NeuAc and GalNAc. The pH optimum, apparent K_m and V_{max} of these enzymes were determined. These findings both support the presence of two alternate pathways in mammalian brain for the catabolism of Tay-Sachs ganglioside and form a starting point for the eventual clarification of the molecular pathogenesis of Tay-Sachs disease.

EFFECT OF DIETARY STARCHES ON THE SERUM, AORTA AND HEPATIC LIPID LEVELS IN HIGH-FAT CHOLESTEROL-FED RATS. PART 2. NATURE OF THE STARCH AND HYPOLIPIDAEMIC ACTIVITY. P. Vijayagopalan and P.A. Kurup (Dept. of Biochem., Univ. of Kerala, Trivandrum-1, India). *Atherosclerosis* 16, 247–56 (1972). The effect of different purified starches on the lipid levels of the serum, liver and aorta of high fat-cholesterol-fed rats has been studied and compared with that of glucose and sucrose. Purified ragi starch (*Eleusine coracana*) showed the lowest cholesterol and phospholipid levels in these tissues and the values obtained were comparable to those of the glucose-fed group. No correlation could be found between the effect on lipid levels and the amylose content of the different starches. Similarly, the amount of N in the different starches also had no relation to their lipid-lowering effect. Ragi and tapioca starch which showed maximum lipid-lowering effect were the least digested with pancreatic α-amylase. The other starches in order of increasing digestibility were bajra, rice, wheat and jowar. The ease of acid hydrolysis of the different starches also showed variation, but no correlation between this and the lipid-lowering effect could be found.

EFFECTS OF TRAINING AND DETRAINING ON THE DISTRIBUTION OF CHOLESTEROL, TRIGLYCERIDE AND NITROGEN IN TISSUES OF ALBINO RATS. E.W. Watt, M.L. Foss and W.D. Block (Phys. Performance Res. Lab. and the Nutr. and Biochem. Res. Lab., Univ. of Mich., Ann Arbor, Mich. 48104). *Circulation Res.* 31, 908–14 (1972). The effects of standardized exercise training and detraining on the content of cholesterol, triglyceride and nitrogen in blood, myocardium, skeletal muscle and epididymal fat of rats were studied. Forty-eight 90-day-old male rats were randomly assigned to four groups (two control groups, one trained group, and one trained and detrained group). Trained rats were subjected to 8 weeks of moderate running in motor-driven wheels; detraining was effected by discontinuing the running program for 8 weeks. Rats were provided a standard diet and water ad libitum. Heart cholesterol content was lowered. The results indicate that the changes in lipid content associated with training and detraining are tissue specific: reduction occurred in some tissues (serum and adipose tissue), although the lipid content of others (skeletal and heart muscle) was independent of training status, body weight or circulating lipid levels.

MECHANISM OF ACTION OF VITAMIN K: DEMONSTRATION OF A LIVER PRECURSOR OF PROTHROMBIN. J.W. Suttie (Dept. of Biochem., College of Agr. and Life Sci., Univ. of Wisconsin, Madison, Wis. 53706). *Science* 179, 192–4 (1973). Extracts of sonicated liver microsomes that are prepared from rats deficient in vitamin K or from rats given vitamin K antagonists contain a factor that liberates a thrombin-like activity when it is incubated with venom from *Echis carinatus*. The amount of this factor is low in control rats and in hypoprothrombinemic rats given vitamin K 1 hour before they were killed. These data indicate that this factor is a protein precursor of prothrombin, which is synthesized in the liver.

OXIDATION OF 20α-HYDROXYCHOLESTEROL BY ADRENAL CORTEX MITOCHONDRIA. L.D. Wilson and B.W. Harding (Depts. of Med. and Biol. Chem., Univ. of Cal., Davis, Cal. 95616). *J. Biol. Chem.* 248, 9–14 (1973). The oxidative conversion of 20α-hydroxycholesterol to pregnenolone by adrenal cortex mitochondria has been studied and correlated with the spectral change produced by this substrate in these particles. The difference spectrum produced by 20α-hydroxycholesterol in the absence of suitable electron donors is a characteristic type II spectrum. Pregnenolone and aminoglutethimide both inhibit side chain cleavage of 20α-hydroxycholesterol at moderately high concentrations. Like metyrapone, these substances interact with cytochrome P-450 in this tissue producing type II difference spectra with estimated dissociation constants smaller than the concentrations required to inhibit the oxidative side chain cleavage of 20α-hydroxycholesterol.

STUDIES ON THE MECHANISM OF ACTIVATION OF ADIPOSE TISSUE PYRUVATE DEHYDROGENASE BY INSULIN. S.I. Taylor, C. Mukherjee and R.L. Jungas (Dept. of Biol. Chem., Harvard Med. Schl., Boston, Mass. 02115). *J. Biol. Chem.* 248, 73–81 (1973). Incubation of rat epididymal adipose tissue fragments with insulin led to increases of up to 2.4-fold in the activity of pyruvate dehydrogenase subsequently assayed in tissue homogenates. The activation of pyruvate dehydrogenase by insulin was greatest when tissue was incubated in the presence of bicarbonate ions and when 2 mg of glucose or fructose per ml was added to the incubation medium. Several other agents known to inhibit lipolysis and to decrease cyclic AMP levels in fat cells, including niacin, 5-methylpyrazole-3-carboxylic acid and prostaglandin E_1 were also effective in activating pyruvate dehydrogenase. Severe depletion of tissue ATP levels caused by the addition of oligomycin or dinitrophenol, by anaerobic incubation or by prolonged incubation with epinephrine in the absence of albumin also activated pyruvate dehydrogenase. Incubation of adipose tissue with 1 mM oleate, 1.5 mM octanoate, 1.5 mM heptanoate, 1 mM butyrate or 3 mM DL-β-hydroxybutyrate decreased pyruvate dehydrogenase activity 20 to 70%. The addition of 5 mM acetate, 5 mM propionate or 3 mM 4-pentenoate led to 70 to 160% activation of pyruvate dehydrogenase, whereas 1.5 mM pentanoate had no effect. High concentrations of glucose (20 mg per ml) or of pyruvate-lactate (5 and 30 mM,

respectively) also increased tissue pyruvate dehydrogenase activity.

THE SYNTHESIS OF MEDIUM-CHAIN FATTY ACIDS BY LACTATING-RABBIT MAMMARY GLAND STUDIED IN VITRO. C.R. Strong, E.M. Carey and R. Dils (Dept. of Biochem., Med. Schl., Univ. of Nottingham, Nottingham NG7 2RD, U.K.). *Biochem. J.* 132, 121–3 (1973). The proportion of 8:0 and 10:0 fatty acids synthesized by the microsomal plus particle-free supernatant fraction from lactating rabbit mammary gland is enhanced at high protein concentrations. This fraction appears to contain a soluble high-molecular-weight factor that modifies the specificity of the fatty acid synthetase complex for termination of the growing acyl chain.

UTILIZATION OF EXOGENOUS FATTY ACIDS FOR COMPLEX LIPID BIOSYNTHESIS AND ITS EFFECT ON DE NOVO FATTY ACID FORMATION IN ESCHERICHIA COLI K-12. D.F. Silbert, T.M. Ulbright and J.L. Honegger (Dept. of Biol. Chem., Wash. Univ. Schl. of Med., St. Louis, Missouri). *Biochemistry* 12, 164–71 (1973). The utilization of exogenous fatty acids by wild-type *Escherichia coli* K-12 has been studied using [^{14}C]-acetate incorporation to monitor fatty acid synthesis and unusual fatty acid analogs as a nonradioactive supplement to distinguish acyl groups in the phospholipid derived from the exogenous source. Certain strains are found to regulate synthesis of saturated and unsaturated fatty acids in response to exogenous fatty acid supplements. This effect is associated with incorporation of the exogenous supplement into phospholipid. Even and odd chain-length saturated, trans and cis unsaturated fatty acids were included among the fatty acid supplements examined. Although a principal result of the incorporation of the exogenous fatty acid is a replacement of structurally related acyl groups, the effect is pleotropic in many instances: for example, 16:0 replaces 18:1 and vice versa. These observations can be rationalized in terms of the positional distribution in the phospholipid of the acyl residues derived from synthesis and the pattern of incorporation of the exogenous fatty acids. β-OH-14:0 and -12:0, added to cultures as supplements, are not detectably incorporated into complex lipid (would be predominantly lipid A) and show little or no influence on fatty acid synthesis.

HETEROGENEITY OF HUMAN PLASMA HIGH DENSITY LIPOPROTEIN. G.S. Sundaram, H.S. Sodhi, and S.L. MacKenzie (Dept. of Med., Univ. of Saskatchewan and Prairie Regional Lab., Nat'l. Res. Council, Saskatoon, Saskatchewan, Canada). *Proc. Soc. Exp. Biol. Med.* 141, 842–5 (1972). It is suggested that high density lipoprotein as isolated in the preparative ultracentrifuge is a heterogeneous mixture of several lipoprotein subspecies.

INTERMEDIATES IN FATTY ACID OXIDATION. H.B. Stewart, P.K. Tubbs and K.K. Stanley (Dept. of Biochem., Univ. of Cambridge, Tennis Court Road, Cambridge CB2 1QW, U.K.). *Biochem. J.* 132, 61–76 (1973). Aqueous extracts of acetone-dried liver and kidney mitochondria, supplemented with NAD^+, CoA and phenazine methosulphate, efficiently convert fatty-acyl-CoA compounds into acetyl-CoA; the process was followed with an O_2 electrode. Label from [1-^{14}C]-octanoyl-CoA appears in acetyl-CoA more rapidly than that from [8-^{14}C]-octanoyl-CoA. Oxidation of [8-^{14}C]-octanoyl-CoA was terminated by addition of neutral ethanolic hydroxylamine and the resulting hydroxamates were separated chromatographically. Hydroxamate derivatives of 3-hydroxyoctanoyl-, hexanoyl-, butyryl-, and acetyl-CoA were obtained. These and other observations suggest that oxidation of octanoyl-CoA by extracts involves participation of free intermediates rather than uninterrupted complete degradation of individual molecules to acetyl-CoA by a multi-enzyme complex. Intact liver mitochondria studied by the hydroxamate technique were also shown to form intermediates during oxidation of labelled octanoates. In addition to octanoylhydroxamate, [8-^{14}C]-octanoate gave rise to small amounts of hexanoyl- butyryl- and 3-hydroxyoctanoyl-hydroxamate. In contrast with extracts, however, where the quantity of intermediates found was a significant fraction of the precursors, mitochondria oxidizing octanoate contained much larger quantities of octanoyl-CoA than of any other intermediate.

EFFECT OF CHLOROPHENOXYISOBUTYRIC ACID (CPIB) ON FAT-MOBILIZING LIPOLYSIS AND CYCLIC AMP LEVELS IN RAT EPIDIDYMAL FAT. L.A. Carlson, G. Walldius and R.W. Butcher (Dept. of Geriatrics, Univ. of Uppsala, Uppsala, Sweden). *Atherosclerosis* 16, 349–57 (1972). High concentrations of chlorophenoxyisobutyric acid (CPIB) reduced basal glycerol release from rat epididymal fat pads in vitro and antagonized the lipolytic effects of noradrenaline. Furthermore, very high concentrations of CPIB significantly antagonized the effects of noradrenaline or ACTH on cyclic AMP accumulation by isolated rat adipocytes. These data are not incompatible with the hypothesis that a primary mechanism in the hypolipidemic action of CPIB is to lower the levels of cyclic AMP in adipose tissue, resulting in decreased hormone-sensitive lipase activity and/or increased lipoprotein lipase activity.

CHOLINE ACETYLTRANSFERASE IN THE NETTLE URTICA DIOICA L. R.B. Barlow and R.O.D. Dixon (Dept. of Pharm., Univ. of Edinburgh, 1 George Sq., Edinburgh EH8 9JZ, U.K.). *Biochem. J.* 132, 15–18 (1973). Extracts of acetone-dried powders prepared from nettle leaves were shown to catalyze the synthesis of acetylcholine. The specific activity of the enzyme in these extracts is of the same order as that of extracts from mammalian sources, such as ox brain, and the effects of temperature and pH are similar to those reported for mammalian choline acetyltransferase. Synthesis is not restricted to the younger leaves but appears to be continuous up to senescence.

LACK OF MAMMALIAN REDUCTION OR ALKYLATION OF 24-METHYLENECHOLESTEROL. W.R. Nes, J.W. Cannon, N.S. Thampi and P.A.G. Malya (Dept. of Biol. Sci., Drexel Univ., Phila., Penn. 19104). *J. Biol. Chem.* 248, 484–7 (1973). The $\Delta^{24(28)}$-bond of 24-methylenecholesterol was not detectably reduced by rats either in vivo or in a liver enzyme preparation. Since the $\Delta^{24(25)}$-bond of other sterols is reduced under the same conditions, mammalian $\Delta^{24(25)}$-reductase must discriminate between the two positions of the pi-electronic system. It also follows that rats do not possess a $\Delta^{24(28)}$-reductase and are therefore incapable of biosynthesizing 24-methylcholesterol as plants do through reduction of a 24-methylene intermediate. 24-Methylenecholesterol also failed in the rat to undergo further addition of a carbon atom to yield a 24-ethylidene- or 24-ethyl derivative as occurs in plants.

ADIPOSE AND LIVER TISSUE ENZYME PROFILES IN OBESE HYPERGLYCEMIC MICE. R.J. Martin, R.F. Welton and B.R. Baumgardt (Dept. of Animal Sci., Penn. State Univ., Univ. Park, Penn. 16802). *Proc. Soc. Exp. Biol. Med.* 142, 241–5 (1973). In order to determine metabolic abnormalities associated with obesity in the obese hyperglycemic mouse, enzyme levels in adipose and liver tissue were measured in three groups of mice. One lean group and one obese group were fed ad libitum. The third group consisted of obese mice subjected to weight gain control by dietary restriction and exercise. Levels of adipose tissue enzymes associated with lipogenesis were higher in obese mice than the lean mice. The data also indicated that obese mouse adipose tissue enzymes were not appreciably affected by diet restriction and exercise. However, liver tissue enzymes associated with lipogenesis and gluconeogenesis, which are normally elevated in obese mice, were restored to normal levels when the obese mice were on a weight gain control schedule. These data indicate that some differences in enzyme profiles observed in a genetically obese mouse are secondary adaptations caused by changes in either dietary intake or spontaneous activity.

EFFECT OF COLESTIPOL (U-26,597A) ON EXPERIMENTAL ATHEROSCLEROSIS IN RABBITS. D. Kritchevsky, Hong K. Kim and S.A. Tepper (Wistar Inst. of Anatomy and Biol., Phila., Penn. 19104). *Proc. Soc. Exp. Biol. Med.* 142, 185–8 (1973). Colestipol, a high molecular-weight polymer which binds bile acids, exerts a hypocholesteremic effect in several species. The addition of 1% colestipol to an atherogenic regimen (2% cholesterol, 6% corn oil) did not significantly affect serum or liver cholesterol levels of rabbits but did cause a significant reduction ($p < 0.05$) in atheromata in both the aortic arch and thoracic aorta. An appreciable number of rabbits fed 1% colestipol were free of atheromata.

LIPID CHANGES IN THE PLASMA LIPOPROTEINS OF BABOONS GIVEN AN ATHEROGENIC DIET. PART 3. A COMPARISON BETWEEN LIPID CHANGES IN THE PLASMA OF THE BABOON AND CHIMPANZEE GIVEN ATHEROGENIC DIETS AND THOSE IN HUMAN PLASMA LIPOPROTEINS OF TYPE II HYPERLIPOPROTEINAEMIA. A.N. Howard, V. Blaton, D. Vandamme, N. Van Landschoot and H. Peeters (Simon Stevin Inst., Bruges, Belgium). *Atherosclerosis* 16, 257–72 (1972). Plasma from normal human subjects and those with type II hyperlipoproteinaemia were separated into α- and β-lipoproteins by electrochromatography; the lipoprotein fractions were analyzed for lipid and fatty acid composition. These data were then compared with those obtained previously in baboons and chimpanzees fed control and atherogenic diets. In type II hyperlipoproteinaemia the increases in the free and esterified cholesterol and cholesterol/phospholipid (C/PL) ratio were chiefly in the β-lipoproteins, but changes in the same direction also occurred in the α-lipoproteins. Increases in the concentration of phosphatidyl choline (PC), lysophosphatidyl choline (OH-PC), and sphingomyelin (S) were confined to the β fraction only. An increase in the cholesterol/phosphatidylcholine ratio occurred in both lipoproteins. It is concluded that both the chimpanzee and baboon fed atherogenic diets provide experimental models for the type II hyperlipoproteinaemia that occurs in man, and that there is a distinct similarity in the change of the lipid composition of the lipoproteins among the three species.

SODIUM-POTASSIUM-ACTIVATED ADENOSINE TRIPHOSPHATASE. IX. THE ROLE OF PHOSPHOLIPIDS. S.S. Goldman and R.W. Albers (Lab. of Neurochem., Nat'l. Inst. of Neur. Diseases and Stroke, Nat'l. Inst. of Health, Bethesda, Md. 20014). *J. Biol. Chem.* 248, 867–74 (1973). The $(Na^+ + K^+)$-ATPase from *electrophorus* electroplax was shown to require phospholipids. Treatment of the enzyme with phospholipase A from *Naja naja* venom removed almost all phosphatides with concomitant loss of enzymatic activity and of the ability to form phosphoryl enzyme. On the other hand digestion with phospholipase C from *Clostridium perfringens* removed nearly all of the lecithin, two-thirds of the phosphatidylethanolamine and none of the phosphatidylserine. ATPase activity was partly inhibited by phospholipase C whereas the steady state level of the phosphoryl enzyme and the rate of the Na^+-dependent nucleotide transphosphorylation were not impaired. Phospholipase C treatment produced an increased proportion of the E_2 conformation at a given level of Mg^{++}; inhibition of the ATPase and p-nitrophenylphosphatase by high Mg^{++} were more pronounced with the phospholipase C-treated enzyme. The reduced turnover of the phospholipase C-treated ATPase probably arises from this same effect. It was concluded that phosphatidylserine is a requirement for the formation of phosphoryl enzyme as well as its dephosphorylation, and phosphatidylethanolamine acts as a modifier which influences the affinity of the enzyme for Mg^{++}.

INDUCTION OF CHOLINE PHOSPHOTRANSFERASE AND LECITHIN SYNTHESIS IN THE FETAL LUNG BY CORTICOSTEROIDS. P.M. Farrell and R.D. Zachman (Pediatric Metabolism Branch, Nat'l. Inst. of Arthritis, Metabolism, and Digestive Diseases, Bethesda, Md. 20014). *Science* 179, 297–8 (1973). Rabbit fetuses 23 to 24 days of gestation were injected with either 9-fluoroprednisolone acetate or saline. Three days later the lungs of steroid-treated animals showed a significant increase in lecithin concentration and cholinephosphotransferase activity. In addition, lung slices from these animals incorporated more [^{14}C]-choline into lecithin. The rise in enzyme activity and [^{14}C]-choline incorporation was blocked by prior treatment of fetuses with cycloheximide but not by treatment with actinomycin D. It is proposed that the corticosteroids induce de novo synthesis of the lung enzyme, which in turn leads to increased synthesis of lecithin through the choline incorporation pathway. Furthermore, it appears that the site of regulation involves translation of messenger RNA.

THE ASSEMBLY OF A STRUCTURAL LIPOPROTEIN IN THE ENVELOPE OF ESCHERICHIA COLI. Masayori Inouye, Jiin Shaw and C. Shen (Dept. of Biochem., State Univ. of N.Y. at Stony Brook, Stony Brook, N.Y. 11790). *J. Biol. Chem.* 247, 8154–9 (1972). A major envelope protein of *Escherichia coli* was found to exist in two different forms in the envelope. One-third of the protein is covalently linked to the peptidoglycan (bound form) as found by Braun and his co-workers. The remaining two-thirds of the protein are in the envelope but are not covalently linked to the peptidoglycan (free form). In contrast to the free form, the bound form can only be solubilized by 1% sodium dodecyl sulfate (SDS) after lysozyme treatment of the envelope. The bound form thus solubilized shows a slightly higher molecular weight than the free form on SDS acrylamide gel electrophoresis because it has an attached peptidoglycan fragment.

ACETYL COENZYME A CARBOXYLASE. MOLECULAR FORMS AND SUBUNIT COMPOSITION OF BIOTIN CARBOXYL CARRIER PROTEIN. R.R. Fall and P.R. Vagelos (Dept. of Biol. Chem., Wash. Univ. Schl. of Med., St. Louis, Missouri 63110). *J. Biol. Chem.* 247, 8005–15 (1972). Milligram quantities of the apparent native form of biotin carboxyl carrier protein (BCCP), a component of *Escherichia coli* acetyl-CoA carboxylase, have been purified to apparent homogeneity using a procedure which largely avoided dissociation or proteolysis. The isolated BCCP served as an effective carboxyl acceptor and donor in the biotin carboxylase and transcarboxylase half-reactions of acetyl-CoA carboxylase at concentrations which were 50- to 100-fold lower than those needed with either BCCP (9100) or BCCP (10400) (previously isolated smaller forms of BCCP with molecular weights of 9,100 and 10,400, respectively). The evidence to date suggests that the apparent native form of BCCP is best represented by a dimeric structure (molecular weight 45,000) which is composed of 2 subunits (molecular weight 22,500), each containing one biotin prosthetic group.

ISOLATION AND CHARACTERIZATION OF LIPOPROTEIN B FROM HIGH-DENSITY HUMAN SERUM LIPOPROTEINS. G. Kostner (Inst. of Med. Biochem., Univ. of Graz, 8010 Graz, Austria). *Biochem. J.* 130, 913–7 (1972). Lipoprotein B from female Lp(a)-lipoprotein-negative serum was isolated from the fraction of density 1.073–1.125 by using immunoadsorbent; 2.5 mg of freeze-dried material was obtained from 100 ml of serum from a fasting patient. The hydrated density of this lipoprotein was found to be 1.084 g/cm.^{3} A flotation rate $F_{1.200}$ of 9.4 and lipid/protein ratio 1.40 were found, similar to that of high-density (d 1.073–1.125) lipoprotein preparations. From immunochemical and electrophoretic studies of the intact and totally delipidized lipoprotein B it was concluded that this lipoprotein represents a separate family within

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• Four Corners. . .

(Continued from page 232A)

tons of total margarine, ghee and bakery fats produced in Turkey.

At present there are five main margarine factories in Turkey—two in Istanbul and two others in Turkey's third major city, Izmir. The fifth is in the south at Adana, in the center of Cukurova, known as the cottonbarn of Turkey.

In recent years the main raw material for margarine in Turkey has been cottonseed oil. Increasing popularity and production of sunflower seed oil today covers almost 65% of yearly oil consumption for edible production. When there is shortage of local oils, generally the third component is imported soybean oil as use of animal fats is prohibited. These three vegetable oils constitute both the liquid and hydrogenated parts of the total margarine, ghee and bakery fats production.

Turkish regulations allow the usage of 16% water phase in margarine and application of β-carotene as a coloring agent. Vitaminization is done by addition 2000 IU vitamin A and 100 IU vitamin D per 100 kg margarine.

Finally, the total vegetable fats market of 1972 with 50% vanaspati, 40% margarine and 10% bakery fats produced by its five main factories is expected to grow to a capacity of more than 200,000 tons in the present year. ■

the high-density range of human serum lipoproteins. The possibility that the isolated lipoprotein B is an artifact created by the isolation procedure is discussed.

FATTY ACID CHAIN ELONGATION IN RAT BRAIN SYNAPTOSOMES. A.H. Koeppen, K.D. Barron and E.J. Mitzen (Res. Service (Neurology), Vet. Adm. Hosp., and Depts. of Neur. and Biochem., Albany Med. College of Union Univ., Albany, N.Y. 12208). *Biochemistry* 12, 276–81 (1973). Rat brain synaptosomes were incubated with ^{14}C-labeled acetate, malonate and their respective coenzyme A(CoA) esters. Only malonyl-CoA was found to be a suitable precursor for fatty acids. Acetyl-CoA was not effectively taken up by synaptosomes. Biosynthesis of synaptosomal fatty acids was stimulated by increased added concentration of ATP or by inclusion of an ATP-generating system (D-glucose). A reduced pyridine nucleotide was indispensable. Docosatetraenoic acid (22:4ω6) contained most of the fatty acid radioactivity after incubation. No de novo biosynthesis of fatty acids occurred. After incubation, the labeled fatty acids were recovered mainly from the synaptosomal mitochondria (80%). Synaptic membranes contained 20% of the label; synaptic vesicles were not radioactive. Synaptosomes had an active malonyl-CoA decarboxylase.

PARTIAL RESOLUTION OF THE ENZYMES CATALYZING OXIDATIVE PHOSPHORYLATION. XXVI. SPECIFICITY OF PHOSPHOLIPIDS REQUIRED FOR ENERGY TRANSFER REACTIONS. Y. Kagawa, A. Kandrach and E. Racker (Section of Biochem. and Molecular Biol., Cornell Univ., Ithaca, N.Y. 14850). *J. Biol. Chem.* 248, 676–84 (1973). Vesicles catalyzing $^{32}P_i$-ATP exchange and ATP-driven proton translocation were reconstituted with chemically defined phospholipids and mitochondrial membrane proteins which were virtually free of electron transport carriers. Phosphatidylcholine and phosphatidylethanolamine were both required for the reconstitution of vesicles with high exchange activity.

EFFECT OF PURIFICATION AND CELLULASE TREATMENT ON THE HYPOCHOLESTEROLEMIC ACTIVITY OF CRUDE KONJAC MANNAN. Shuhachi Kiriyama, Yuriko Ichihara, Akiko Enishi and Akira Yoshida (Dept. of Nutr., Schl. of Med., Tokushima Univ., 3 Kuramoto, Tokushima, Japan). *J. Nutr.* 102, 1689–98 (1972). This paper presents a series of experiments to identify the hypocholesterolemic principle in crude konjac mannan (CKM) prepared from tubers of *Amorphophallus konjac* K. Koch (Araceae). Extensively purified konjac mannan (HPKM) showed hypocholesterolemic activity comparable to or rather higher than that of CKM or partially purified konjac mannan (PPKM). Therefore, various low molecular contaminants do not appear to contribute to the activity of CKM. Hydrolysis with fungal cellulase completely canceled the activity of CKM, PPKM or HPKM even after a short 30-minute hydrolysis. Also, concurrent addition of intact cellulase and konjac mannan preparations could not depress the plasma cholesterol level which is elevated by dietary cholesterol plus bile salts. It was concluded that konjac mannan itself is an active principle and that both the essential properties in konjac mannan itself (macromolecular and water-soluble) must be simultaneously satisfied for the actualization of hypocholesterolemic activity as suggested previously.

CHOLINE METABOLISM IN THE CEREBRAL CORTEX OF GUINEA PIGS. PHOSPHORYLCHOLINE AND LIPID CHOLINE. M.J. Dowdall, L.A. Barker and V.P. Whittaker (Dept. of Neurochem., Inst. for Basic Res. in Mental Retardation, Staten Island, N.Y. 10314). *Biochem. J.* 130, 1081–94 (1972). The labelling of phosphorylcholine and choline-containing phospholipids in the subcellular fractions of guinea-pig cerebral cortex after the intraventricular injection of [N-Me-^3H]choline into conscious animals has been studied. Special emphasis was placed upon the synaptosome fraction and early time-periods after administration. The labelling of phosphorylcholine was rapid compared with that of phospholipid and was confined to two distinct subcellular fractions: the soluble cytoplasmic fraction and the synaptosome fraction. Most of the labelled phosphorylcholine of the synaptosome fraction was readily released by osmotic rupture indicating location in the nerve-ending cytoplasm. The two pools of phosphoryl-choline had similar specific radioactivities at all observed times. ^3H-labelled phospholipid was found in all membranous fractions. The labelling was confined to choline-containing phospholipids, notably phosphatidylcholine. The labelling of the different membranous fractions was similar. The half-life of the choline-containing phospholipids in the synaptic vesicle fraction was very much greater than the acetylcholine in this fraction. Evidence is presented that synthesis de novo of phosphatidylcholine at nerve terminals occurs in vivo.

THE PHOSPHOLIPID-CHOLESTEROL INTERACTION. KINETICS OF WATER PERMEABILITY IN LIPOSOMES. R. Bittman and L. Blau (Dept. of Chem., Queens College of the City Univ. of New York, Flushing, N.Y. 11367). *Biochemistry* 11, 4831–9 (1972). Stopped-flow light-scattering measurements were made on liposomes exposed to hypo- and hyperosmolar KCl solutions. The rapid recordings of changes in light transmission accompanying rapid changes in osmolarity represent liposomal water permeability. Equilibrium and kinetic data indicate that liposomes prepared with KCl in the osmotically active spaces obey the Boyle-van't Hoff law and behave as ideal osmometers. Increasing unsaturation of the phospholipid species caused an increase in liposomal water permeability. The initial rate of water permeability of liposomes derived from saturated and unsaturated lecithins above their crystalline to liquid-crystalline transition temperatures decreased with increasing cholesterol concentration. On the other hand, cholesterol enhanced the rate of water permeability of liposomes derived from saturated lecithins below their transition temperatures. The results are consistent with the hypothesis that the permeability of the bilayer is governed by the fluidity of the hydrocarbon chains and support the solubility-diffusion mechanism of water permeation.

A CASE OF MASSIVE HYPERTRIGLYCERIDEMIA CORRECTED BY NICOTINIC ACID OR NICOTINAMIDE THERAPY. L.A. Carlson, S. Froberg and L. Oro (Dept. of Internal Med., Karolinska Hosp., Stockholm, Sweden). *Atherosclerosis* 16, 359–68 (1972). A case of massive hypertriglyceridemia with fasting plasma triglycerides around 100 mmoles/l is described. Large amounts of chylomicra were present in fasting plasma and the amounts of low-density and high-density lipoproteins were very low. Postheparin plasma lipolytic activity was normal and intravenous heparin rapidly cleared the patient's abnormally prolonged alimentary lipemia with a concomitant rise in plasma free fatty acid levels. Nicotinic acid or nictotinamide given in doses of 3 g or more daily reduced plasma triglyceride levels to about 2–3 mmoles/l and raised the reduced levels of low- and high-density lipoproteins. The mode of onset of this therapeutic effect was slow and the effect persisted for several weeks after withdrawal of either nicotinic acid or nicotinamide. The pathogenesis of the hypertriglyceridemia as well as the mode of action of nicotinic acid and nicotinamide is discussed.

PANCREATECTOMY IN THE EEL: EFFECTS ON SERUM GLUCOSE AND CHOLESTEROL. T.L. Lewis and A. Effle (Daniel Baugh Inst. of Anatomy, Jefferson Med. College of Thomas Jefferson Univ., Philadelphia, Pa. 19107). *Science* 178, 1286–8 (1972). Pancreatectomy in the eel causes a slight tendency toward hyperglycemia; total and partial pancreatectomies cause a drop of total serum cholesterol. Thus, complete removal of the islet tissue in this teleost is not followed by diabetes mellitus, and also the endocrine control of the cholesterol-containing serum components seems to differ from that in mammals.

THE EFFECT OF ALFATOXIN B_1 ON NORMAL AND CORTISOL-STIMULATED RAT LIVER RIBONUCLEIC ACID SYNTHESIS. G.E. Neal (Med. Res. Council, Toxicology Unit, Med. Res. Council Labs., Woodmansterne Rd., Carshalton, Surrey, U.K.). *Biochem. J.* 130, 619–29 (1972). Aflatoxin B_1, administered in vivo, inhibits the incorporation of [^{14}C]orotic acid in vivo into rat liver nuclei, and also inhibits both Mg^{2+} and Mn^{2+} dependent RNA polymerase activities in nuclei assayed in vitro. Aflatoxin B_1 inhibits the cortisol-induced increase in incorporation of [^{14}C]leucine in vivo, but does not affect the control value of this activity. Aflatoxin B_1 administered in vivo inhibits the increase in nuclear Mg^{2+} dependent RNA polymerase activity, assayed in vitro, which results from the treatment with cortisol. It is suggested that aflatoxin B_1 exerts its effect on RNA polymerase by decreasing the template capacity of the chromatin and that the aflatoxin 'target' area of the chromatin includes that region which is stimulated by cortisol. This process, however, does not involve inhibiting the movement of cortisol from the outside of the hepatic cell to the nuclear chromatin.

A MICROASSAY FOR MITOCHONDRIAL α-GLYCEROPHOSPHATE DEHYDROGENASE. R.S. Gardner and D.M. Kipnis (Metabolism Div., Washington Univ., Schl. of Med., St. Louis, Mo. 63110). *Proc. Soc. Exp. Biol. Med.* **141**, 400–2 (1972). A simple reproducible assay for mitochondrial α-GPD has been described. It takes advantage of the ability of INT to accept directly electrons from the dehydrogenase. The assay as described measures accurately enzyme activity using 20 μg of mitochondrial protein, but it could be scaled down for smaller quantities of protein by using smaller reaction volumes and microcuvettes.

RETINOL GLYCOLOPIDS. L. DeLuca, N. Maestri, G. Rosso and G. Wolf (Lung Cancer Unit, Nat'l. Cancer Inst., Nat'l. Inst. of Health, Bethesda, Md. 20014). *J. Biol. Chem.* **248**, 641–8 (1973). In vivo and in vitro experiments demonstrated that rat liver cells are able to synthesize a double-labeled mannolipid from [^{3}H]-retinol and GDP-[^{14}C]-mannose. The double-labeled mannolipid was purified by DEAE-cellulose, silicic acid and thin-layer chromatography, and shown to contain retinol and mannose in a molar ratio in the range of 1:1.

THE ACTIVITIES OF LIPASES AND CARNITINE PALMITOYLTRANSFERASE IN MUSCLES FROM VERTEBRATES AND INVERTEBRATES. B. Crabtree and E.A. Newsholme (Agr. Res. Council Unit of Insect Physiol., Dept. of Zoology, Univ. of Oxford, South Parks Road, Oxford OX1 3PS, U.K.). *Biochem. J.* **130**, 697–705 (1972). The activities of tri-, di- and mono-glyceride lipase and carnitine palmitoyltransferase were measured in homogenates of a variety of muscles. These activities were used to estimate the rate of utilization of glycerides and fatty acids by muscle. In muscles whose estimated rates of fat utilization can be compared with rates calculated for the intact muscle from such information as O_2 uptake, there is reasonable agreement between the estimated and calculated rates. In all muscles investigated the maximum rates of hydrolysis of glycerides increase in the order triglyceride, diglyceride, monoglyceride. The activity of diglyceride lipase is highest in the flight muscles of insects such as the locust, waterbug and some moths and is lowest in the flight muscles of flies, bees and the wasp. These results are consistent with the utilization of diglyceride as a fuel for some insect flight muscles. In many muscles from both vertebrates and invertebrates the activity of glycerol kinase is similar to that of lipase. It is concluded that in these muscles the metabolic role of glycerol kinase is the removal of glycerol produced during lipolysis. However, in some insect flight muscles the activity of glycerol kinase is much greater than that of lipase, which suggests a different role for glycerol kinase in these muscles.

FAT METABOLISM IN HIGHER PLANTS. β-HYDROXYLATION OF FATTY ACIDS BY A SOLUBLE PREPARATION FROM MATURING AVOCADO MESOCARP. J.L. Harwood, A. Sodja and P.K. Stumpf (Dept. of Biochem. and Biophysics, Univ. of Cal., Davis, Cal. 95616). *Biochem. J.* **130**, 1013–8 (1972). An avocado supernatant fraction converted fatty acids of medium chain length (C_8–C_{12}) into a polar product. The product was identified as the β-hydroxy derivative of the substrate by GLC and TLC analysis. For hydroxylation of the fatty acids, CoA, ATP and molecular oxygen were required. Acyl carrier protein gave some stimulation. The reaction took place with oxygen alone if acyl-CoA was the substrate. Hydroxylation was maximal with decanoic acid but dodecanoic acid and octanoic acid were also very active. Acids of shorter or longer chain lengths were not hydroxylated. NAD^+ concentration caused complete inhibition at 0.5 mM and may be an important control mechanism for the reaction in vivo. The reaction was inhibited by iodoacetamide and by bipyridyl and carbon monoxide, indicating involvement of thiol and heavy metal groups.

PHYSICOCHEMICAL PROPERTIES OF BOVINE SERUM HIGH DENSITY LIPOPROTEIN. A. Jonas (Dept. of Biochem., Schl. of Chem. Sci., Univ. of Illinois, Urbana, Ill. 61801). *J. Biol. Chem.* **247**, 7767–72 (1972). Bovine high density lipoprotein, floating between densities of 1.063 g per ml and 1.125 g per ml, constitutes about 80% of all bovine serum lipoproteins. Preparations of this lipoprotein appear homogeneous by several criteria: electrophoresis, sedimentation velocity, sedimentation equilibrium and NH_2-terminal amino acid analysis. Bovine high density lipoprotein is a spherical protein-lipid aggregate, containing 32% protein and 68% lipid by weight; its molecular weight is 376,000. The protein component appears to consist of 4 sub-units of molecular weight 28,100. Tryptophan fluorescence spectra of the native lipoprotein and of its delipidated form have maximum fluorescence wave lengths at 328 and 338 nm, respectively, an indication that on delipidation the environment of tryptophan residues becomes more polar.

MEDIUM CHAIN FATTY ACID BINDING TO HUMAN PLASMA ALBUMIN. J.D. Ashbrook, A.A. Spector and J.E. Fletcher (Lab. of Applied Studies, Div. of Computer Res. and Tech., Natl. Insts. of Health, Bethesda, Md. 20014). *J. Biol. Chem.* **247**, 7039–42 (1972). The binding of hexanoic, octanoic and decanoic acids to defatted human plasma albumin was measured by equilibrium dialysis at 37C in a calcium-free Krebs-Ringer phosphate buffer, pH 7.4. The results were analyzed in terms of multiple stepwise equilibria. For each of the albumin binding sites, the magnitude of the equilibrium constants increased as the chain length of the acid increased: decanoate > octanoate > hexanoate. Octanoate binding was relatively insensitive to pH changes over the range of 6.0 to 8.2. Decanoate binding also was similar at pH 6.5 and 7.4. A decrease in octanoate binding occurred when the albumin was acetylated or when the medium contained 6 M urea. Octanoate binding also was decreased when either palmitate or oleate was added to albumin, suggesting that medium chain fatty acid transport may be influenced by changes in the plasma long chain free fatty acid concentration.

SIGNIFICANCE OF SKIN AS A SITE OF FATTY ACID AND CHOLESTEROL SYNTHESIS IN THE CHICK. Shu-Jen Chang Yeh and G.A. Leveille (Lab. of Nutr. Biochem., Dept. of Animal Sci., Univ. of Ill. at Urbana-Champaign, Urbana, Ill. 61801). *Proc. Soc. Exp. Biol. Med.* **142**, 115–9 (1973). The significance of skin as a site of fatty acid and cholesterol synthesis has been studied in the chick. Both in vitro and in vivo experiments indicate that chick skin has the capacity to synthesize fatty acid and cholesterol. The addition of glucose to the incubation medium enhanced fatty acid synthesis but not cholesterol synthesis from acetate-1-^{14}C in chick skin. The rate of in vitro lipogenesis in skin is markedly lower than that in liver and slightly lower than that in adipose tissue. The estimated contribution of skin to total fatty acid synthesis in the intact chick was about 7%, while skin accounted for approximately 6% of total cholesterol synthesis.

MODE OF ACTION OF VITAMIN K. CALCIUM BINDING PROPERTIES OF BOVINE PROTHROMBIN. G.L. Nelsestuen and J.W. Suttie (Dept. of Biochem., College of Agr. and Life Sci., Univ. of Wis., Madison, Wis. 53706). *Biochemistry* **11**, 4961–4 (1972). Due to recent observations which suggest that the calcium binding property of prothrombin results from the action of vitamin K, the binding of calcium to prothrombin has been examined more carefully. These observations indicate that the abnormal prothrombin differs from the normal protein in some way other than its disulfide-bond arrangement or tertiary structure.

EFFECT OF METHYLPREDNISOLONE IN EPINEPHRINE-THYROXINE INDUCED ARTERIOSCLEROSIS. R.O. Langner and G.C. Fuller (Univ. of Conn., School of Pharmacy, Storrs, Conn. 06268). *Proc. Soc. Exp. Biol. Med.* **141**, 999–1003 (1972). Rabbits treated with methylprednisolone had decreased levels of aortic proline hydroxylase activity, indicating that glucocorticoids decrease collagen content by affecting the rate of synthesis. The inhibitory effects of methylprednisolone on enzyme activity were reversed by treatment of rabbits with epinephrine-thyroxine. Methylprednisolone which is reported to inhibit aortic sclerosis in cholesterol fed rabbits also did not protect epinephrine-thyroxine treated rabbits from aortic sclerosis.

THE IN VITRO EFFECT OF STEROIDS ON POLYMORPHONUCLEAR LEUKOCYTE METABOLISM. M.R. Cooper, L.R. DeChatelet and C.E. McCall (Depts. of Med. and Biochem., Bowman Gray School of Med., Winston-Salem, N.C. 27103). *Proc. Soc. Exp. Biol. Med.* **141**, 986–90 (1972). Steroids added in vitro to human neutrophils have numerous effects on cellular metabolism, including inhibition of hexose monophosphate shunt activity, particle uptake and iodination of zymosan particles. The magnitude of these effects varies considerably with the steroid employed.

Obituaries

Robert Starr, who joined AOCS in 1952, died March 18, in Minneapolis, Minn. Starr was employed for 20 years as chief chemist for Honeymead Products and was a consultant for the American Soybean Association and National Soybean Processors.

Word has also been received of the death of John D. Zech, who was employed by ICI America, Inc., Wilmington, Del., and became a member of the Society in 1963. ■

TIME COURSE OF THE ACUTE ALCOHOLIC FATTY LIVER AND CONCOMITANT MITOCHONDRIAL FUNCTION IN FASTED RATS. E.S. Higgins and W.H. Friend (Dept. of Biochem., Med. College of Va., Health Sci. Div. of Va. Commonwealth Univ., Richmond, Va. 23219). *Proc. Soc. Exp. Biol. Med.* 141, 944–7 (1972). Pyrazole, an inhibitor of liver alcohol dehydrogenase and ethanol metabolism, was used to distinguish between acute biochemical responses produced directly by alcohol and secondarily by its metabolic effects. Over the 40 hr. period studied, accumulation of hepatic triglycerides was dependent upon an initial uptake of fatty acids from adipose stores and continued oxidation of ethanol by the liver. The increased efficiency of mitochondrial function, as monitored by the magnitude of respiratory control ratios, followed a time course isochronous with the blood alcohol curve and was dependent upon in situ exposure of the organelle to the alcohol molecule, not its metabolic sequelae. These relatively short-term responses are not sustained in well nourished animals receiving ethanol chronically.

BODY FAT CONTENT AND METABOLIC RATE OF RODENTS: DESERT AND MOUNTAIN. I.M. Scott, M.K. Yousef and W.G. Bradley (Dept. of Biol. Sci., Univ. of Nevada, Las Vegas, Nevada 89109). *Proc. Soc. Exp. Biol. Med.* 141, 818–21 (1972). Fifteen species of rodents representing three families and a wide range of ecologic distribution were used to investigate relationships between body fat and metabolic rates (V_{O_2}). There was no apparent relationship between fat content and V_{O_2}. The correlation between V_{O_2}, body weight, body water and fat free body weight was essentially the same. Ecologic distribution appears to be of more importance than fat content in modification of V_{O_2}.

THE FORM OF ABSORPTION OF LIPIDS IN THE CHICKEN, GALLUS DOMESTICUS. A. Bensadoun and A. Rothfeld (Dept. of Poultry Sci., Cornell Univ., Ithaca, N.Y. 14850). *Proc. Soc. Exp. Biol. Med.* 141, 814–7 (1972). It has been demonstrated with the use of a functionally hepatectomized preparation and in the presence of a lipoprotein lipase inhibitor, Triton WR 1339, that in the chicken, long chain fatty acids are absorbed in the form of triglycerides as the major component of a lipoprotein of density less than 1.006.

REGULATION OF THE ACTIVITY OF ACETYL COENZYME A CARBOXYLASE BY PALMITOYL COENZYME A AND CITRATE. A.G. Goodridge (Banting and Best Dept. of Med. Res., Univ. of Toronto, Toronto 101, Ontario, Canada). *J. Biol. Chem.* 247, 6946–52 (1972). Palmitoyl-CoA (100 μM), in the presence of albumin (24 mg per ml), inhibited the incorporation of [^{14}C]citrate into fatty acids in a cytosol fraction of chick liver and inhibited the activity of acetyl-CoA carboxylase purified from chick liver. Under similar incubation conditions, palmitoyl-CoA (200 μM) did not inhibit fatty acid synthetase, ATP-citrate lyase, malic enzyme, NADP-linked isocitrate dehydrogenase, pyruvate kinase or glutamate dehydrogenase. It is concluded that palmitoyl-CoA inhibition of acetyl-CoA carboxylase is reversible and competitive with citrate and, therefore, may play an important role in the regulation of fatty acid synthesis in vivo.

EFFECTS OF ADENOSINE NUCLEOSIDES ON ADENYLATE CYCLASE, PHOSPHODIESTERASE, CYCLIC ADENOSINE MONOPHOSPHATE ACCUMULATION AND LIPOLYSIS IN FAT CELLS. J.N. Fain, R.H. Pointer and W.F. Ward (Div. of Biol. and Med. Sci., Brown Univ., Providence, R.I. 02912). *J. Biol. Chem.* 247, 6866–72 (1972). Adenosine and 2′-deoxyadenosine at concentrations in the range of 5 to 50 μM inhibited the activation of adenylate cyclase activity in fat cell "ghosts" by norepinephrine. 2′,5′-Dideoxyadenosine was more potent than adenosine while arabinosyl adenosine, 3′-deoxyadenosine and 3-fluoroadenosine were equi-potent to adenosine as inhibitors of adenylate cyclase. The large accumulation of cyclic adenosine 3′,5′-monophosphate (cyclic AMP) seen in fat cells incubated for 5 min with catecholamines plus theophylline was markedly inhibited by 2′,5′-dideoxyadenosine, adenosine and 2-fluoroadenosine. The effect of adenosine and related nucleosides on cyclic AMP accumulation suggest that they may play a physiological role as feedback regulators of fat cell adenylate cyclase.

LYSOPHOSPHOLIPASES OF RAT BRAIN. Z. Leibovitz-BenGershon, I. Kobiler and S. Gatt (Dept. of Biochem., Hebrew Univ.-Hadassah Med. Schl., Jerusalem, Israel). *J. Biol. Chem.* 247, 6840–7 (1972). The properties of rat brain lysophospholipase were investigated. Three subcellular fractions were employed: a particulate preparation which sedimented at 26,000 × g, a microsomal preparation which sedimented at 100,000 × g, and the supernatant of the above. When reactions rates were plotted as a function of enzyme concentration, straight lines were obtained with the soluble enzyme. With the particulate or microsomal enzymes, these curves were straight lines only at lysolecithin concentrations below 0.05 to 0.1 mM. Above these concentrations, the curves were parabolic. Albumin increased the reaction rates catalyzed by the particulate or microsomal enzymes at all substrate concentrations.

LONG CHAIN BASE-ACETYL COENZYME A ACETYLTRANSFERASE FROM THE MICROSOMES OF HANSENULA CIFERRI. II. KINETIC PROPERJIES. Y. Barenholz and S. Gatt (Dept. of Biochem., Hebrew Univ.-Hadassah Med. Schl., Jerusalem, Israel). *J. Biol. Chem.* 247, 6834-9 (1972). The microsomal long chain base-acetyl coenzyme A acetyltransferase catalyzes a bisubstrate reaction in which the acetyl group of acetyl-CoA is transferred to free or N-acetylated sphingosine bases and to long chain primary amines. In this reaction the acetyl-CoA is present in a true, molecular solution while the lipid substrates are present as molecular solutions only at low concentrations; at higher concentrations they form micellar aggregates.

CHOLESTEROL: VITAMIN C CONTROLS ITS TRANSFORMATION IN BILE ACIDS. E. Ginter (Inst. of Human Nutr. Res., Bratislava, Czech.). *Science* 179, 702–4 (1973). Cholesterol accumulates in the blood serum and in the liver of guinea pigs with chronic latent vitamin C deficiency. The reason for this is the decreased rate of transformation of cholesterol to bile acids in the liver of animals deficient in vitamin C. A significant direct correlation exists between the vitamin C concentration in the liver and the rate of cholesterol transformation to bile acids.

FETAL GROWTH AND PLACENTAL PERMEABILITY IN RABBITS FED CHOLESTEROL. D.B. Zilversmit, M. Remington and L.B. Hughes (Graduate Schl. of Nutr. and Section of Biochem. and Molecular Biol., Div. of Biol. Sci., Cornell Univ., Ithaca, N.Y. 14850). *J. Nutr.* 102, 1681–8 (1972). Rabbits fed cholesterol before and during pregnancy showed a high fetal mortality. Decreasing the dose and duration of cholesterol feeding decreased this mortality. Successful insemination in cholesterol-fed does was less frequent than in control animals. Fetuses from cholesterol-fed does weighed significantly less than those from control does, but newborn young did not show a significant difference in birth weight. Placentas from cholesterol-fed does showed greatly increased concentrations of cholesterol, particularly in the esterified form. Placental transfer of ^{131}I measured in vivo did not show significant differences as a result of cholesterol feeding.

BIOSYNTHESIS OF UNSATURATED SPHINGOLIPID BASES BY MICROSOMAL PREPARATIONS FROM OYSTERS. R.K. Hammond and C.C. Sweeley (Dept. of Biochem., Michigan State Univ., East Lansing, Mich. 48823). *J. Biol. Chem.* 248, 632-40 (1973). A cell-free particulate preparation from oyster viscera incorporated [^{14}C]palmitate and [^{14}C]-serine into the long chain base sphingadienine. Both double bonds of the product were shown to have the trans configuration. trans-2-Hexadecenoic and trans-6-hexadecenoic acids were not effective in decreasing the incorporation of the radioactive label from palmitate, nor was label from palmitate incorporated into these two monounsaturated fatty acids. Excess sphinganine (dihydrosphingosine) and sphing-4-enine (sphingosine) added to incubation mixtures also had little effect on the level of incorporation of palmitate into the dienic base, whereas about 30% reduction was observed when 3-keto-sphinganine was added. Mono- and diunsaturated 3-keto bases were produced in vitro in the absence of NADPH, and 3-keto[^{14}C]sphinganine and 3-keto[^{14}C]sphing-4-enine were converted in high yields to labeled 3-ketosphingadienine under these conditions. These data are consistent with a mechanism involving condensation of palmitoyl coenzyme A with serine and subsequent desaturations of the 3-keto-sphinganine to form mono- and diunsaturated intermediates, followed by reduction to sphingadienine.

HETEROGENEITY OF HUMAN PLASMA VERY LOW DENSITY LIPOPROTEINS. SEPARATION OF SPECIES DIFFERING IN PROTEIN COMPONENTS. V.G. Shore and B. Shore (Biomed. Div., Lawrence Livermore Lab., Univ. of Cal., Livermore, Cal. 94550). *Biochemistry* 12, 502–7 (1973). The heterogeneity of human plasma very low density lipoproteins (VLDL) with respect to the proteins associated with the lipoprotein particles was demonstrated by fractionation of VLDL by affinity chromatography. The proteins associated with the VLDL fractions were determined by disc electrophoresis in polyacrylamide gels containing 8 M urea and by immunodiffusion assay. The identities of the various components in the electrophoretic patterns were established by amino acid composition of the corresponding proteins isolated by ion exchange chromatography. These studies suggest that most VLDL species contain more than one kind of protein and that some species are deficient in one or more of the proteins found in unfractionated VLDL. No absolute deficiences in any of the major VLDL proteins were seen among a number of fasting individuals, both normal and hyperlipemic, but the proportions of the proteins varied, with some hyperlipemic VLDL proteins showing a relative deficiency or excess of one or other of the proteins. It is suggested that differences among VLDL species may reflect a dynamic or changing composition of the plasma VLDL. Individual differences in the proportions of the VLDL proteins, and presumably the VLDL species, may reflect differences in the homeostasis of lipid metabolism.

THE PATHOGENESIS OF NEUROGENIC HYPERCHOLESTEROLEMIA. IV. ABNORMAL METABOLISM OF CHYLOMICRONOUS CHOLESTEROL. M. Friedman, S.O. Byers and S. Elek (Harold Brunn Inst., Mt. Zion Hosp. and Med. Center, San Francisco, Cal. 94115). *Proc. Soc. Exp. Biol. Med.* 142, 359–64 (1973). Intestinal lymph was collected from both normal and hypothalamus-injured rats given ^{3}H-cholesterol or ^{3}H-tripalmitin. It was found that chylomicronous cholesterol obtained from the hypothalamus-injured rat was removed from the blood and accumulated in the liver of the normal rat at the same rate as chylomicronous cholesterol collected from normal rats. However, the liver of the hypothalamus-lesioned rat removed normal chylomicronous cholesterol from plasma at a much slower rate than the liver of the normal rat. Chylomicronous triglyceride, however, was removed from plasma at a normal rate by the lesioned rat. The heparin-induced lipoprotein-lipase activity of the plasma of hypothalamus-injured rats was significantly greater than that of the plasma of normal rats.

EFFECTS OF ETHANOL ON THE METABOLISM OF FREE FATTY ACIDS IN ISOLATED LIVER CELLS. J.A. Ontko (Cardiovascular Res. Program., Oklahoma Med. Res. Found., and Dept. of Biochem. and Molecular Biol., College of Med., Univ. of Oklahoma Health Sciences Center, Oklahoma City, Okla. 73104). *J. Lipid Res.* 14, 78–86 (1973). Ethanol inhibited the oxidation and enhanced the esterification of albumin-bound [1-^{14}C]palmitate incubated with isolated rat liver cells. Ethanol decreased the conversion of [1-^{14}C]palmitate to $^{14}CO_2$ and ^{14}C-labeled ketone bodies and enhanced the incorporation of [1-^{14}C]palmitate into glycerolipids, especially triglyceride; cholesteryl ester synthesis was unaffected. The half-maximal effective ethanol concentration for each of these processes was 6–10 μg/ml and a maximum effect was produced by about 50 μg/ml. Ethanol oxidation was required for each of these alterations, since the effects were completely abolished by pyrazole. The energy obtainable from ethanol oxidation was in excess of the energy deficit from decreased fatty acid oxidation. However, ethanol did not affect O_2 consumption, indicating that ethanol oxidation replaced the oxidation of both fatty acids and other substrates. Ethanol inhibited the citric acid cycle in the intact liver cells by 20–30%. The major site of inhibition was α-ketoglutarate oxidation. Results suggest that ethanol inhibited α-ketoglutarate dehydrogenase in the mitochondria of hepatocytes by elevating the mitochondrial NADH:NAD ratio.

LIPOGENIC ACTIVATION AFTER NIBBLING AND GORGING IN MICE. N. Baker and R.J. Huebotter (Radioisotope Res., Vet. Admin., Wadsworth Hosp. Center, Los Angeles, Cal. 90073). *J. Lipid Res.* 14, 87–94 (1973). Lipogenic activation was studied in mice that had been restricted to a single large meal once a day rather than being allowed to eat at frequent intervals throughout the night. Mice were injected intravenously with [U-^{14}C]glucose, and the flux of glucose C to total lipid fatty acids (TLFA) and to all "end products" was estimated from serial plasma glucose specific activities and measurements of incorporation of ^{14}C into TLFA of hepatic and extrahepatic tissues. Tracer studies were carried out in mice fasted for 1 day and at various times after the mice ate one or two small test meals or a single large test meal. Test meals consisted of a fat-free, 58% glucose diet. The flux of glucose C to TLFA increased by an order of magnitude within an hour after mice nibbled a test meal for several minutes. After ingestion of two small test meals or a single large test meal, the flux of glucose C to TLFA increased from a fasting rate of 0.5 to 35 and 87 μg of glucose C/min/20 g body wt, respectively. Although trained meal eaters are thought to have abnormally increased lipogenesis, their lipogenic response to a single test meal was the same as that previously reported for untrained nibbling mice.

SPECIFIC ROLE OF GLUCOSE IN RAPID LIPOGENIC ACTIVATION IN VIVO. *Ibid.*, 95–101. Lipogenesis from glucose C was previously found to be rapidly activated as soon as mice nibbled a fat-free, glucose-rich diet. We have studied here whether such rapid activation is a specific effect of dietary glucose. The flux of endogenous glucose C to total lipid fatty acids (TLFA) in mice fasted for 1 day was compared with the minimal average flux of exogenous dietary glucose to TLFA during a 40-min period after the ingestion of various glucose-rich test meals by previously fasted mice. Only 0.6 to 0.8 μg of glucose C/min/20 g body wt was converted to TLFA, whereas 208 ± 16 μg glucose C/min/20 g body wt was converted to all "end products" in the fasted animals.

LUNG SURFACE-ACTIVE FRACTION AS A MODEL SYSTEM FOR MACROMOLECULAR ULTRASTRUCTURAL STUDIES WITH CROTALUS ATROX VENOM. M.F. Frosolono, R. Pawlowski, B.L. Charms, C. Corbusier, M. Abrams and J. Jones, III (Pulmonary Res. Lab., Mt. Sinai Hosp. of Cleveland, Univ. Circle, Cleveland, Ohio 44106). *J. Lipid Res.* 14, 110–20 (1973). The dog lung surface-active fraction and phosphatidylcholine constituents were subjected to hydrolysis by *Crotalus atrox* phospholipase A_2. Relative rates of hydrolysis were: dipalmitoyl glycerophosphorylcholine > phosphatidylcholine isolated from the surface-active fraction > phosphatidylcholine as an integral component of the intact surface-active macromolecular structure. Cholesterol markedly inhibited, whereas tripalmitin increased, the rate of hydrolysis with both pure phosphatidylcholine substrates. The effect of temperature on the velocity indicated the enzyme was most active when the substrates were in the gel state. These kinetic results, in conjunction with surface chemistry studies, can be interpreted to indicate that the phosphatidylcholine in the intact surface-active macromolecular particle is liquid crystalline due to molecular interactions with other constituents. Gas-liquid chromatographic analysis of the 2-lysophosphatidylcholines and fatty acids produced from the enzymatic hydrolysis of the intact surface-active fraction indicated that palmitoyl residues were more accessible to the enzyme, perhaps because they occupied positions near the surface of the particle.

INTRAMUSCULAR ENERGY SOURCES IN DOGS DURING PHYSICAL

(Continued on page 247A)

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WORK. D.G. Therriault, G.A. Beller, J.A. Smoake and L.H. Hartley (Biochem.-Pharmacology and Physiology Labs., U.S. Army Res. Inst. of Environmental Med., Natick, Mass. 01760). *J. Lipid Res.* **14**, 54–60 (1973). Three groups of dogs were run under different experimental conditions characterized by varying the work load or the running time. Lipid and glycogen analyses were carried out on biopsy specimens from the biceps femoris muscle before and after exercise. In addition, arterial and venous triglycerides and free fatty acids were determined on plasma samples from one group of dogs that had been previously catheterized. Under the conditions of these experiments, results revealed: plasma triglycerides did not contribute significantly to the energy supply for muscle contraction; plasma free fatty acid efflux into muscle was increased during mild exercise but significantly lowered during heavy exercise; exercise did not affect the phospholipid level or its composition in the muscle; and muscle triglyceride levels may increase, decrease or remain unchanged, depending upon the work load imposed by the exercise.

ACTIVATION OF BRANCHED AND OTHER LONG-CHAIN FATTY ACIDS BY RAT LIVER MICROSOMES. K. Lippel (Nutr. Inst., Agr. Res. Service, U.S. Dept. of Agr., Beltsville, Md. 20705). *J. Lipid Res.* **14**, 102–9 (1973). Acyl coenzyme A synthetase (EC 6.2.1.3) of rat liver microsomes activates iso- and anteiso-branched long-chain fatty acids containing 12 to 20 carbon atoms. Fatty acid chain length appears to be the major determinant of the maximum rate of acyl CoA biosynthesis of branched, or saturated, or cis monounsaturated long-chain fatty acids. Based on activation studies conducted at 22–45 C, it is concluded that the rate of activation is a function of long-chain fatty acid solubility. The shape of the in vitro activation curve with respect to fatty acid concentration appears to be determined by fatty acid melting point as well as by the presence and position of double bonds. Differently shaped activation curves were observed for cis or trans Δ^6 to Δ^{12} central positional isomers of octadecenoic acid and for Δ^3, Δ^4, Δ^{13} to Δ^{15} terminal isomers of octadecenoic acid. The relationships between fatty acid structure, melting point, solubility and shape of the activation curve observed during in vitro measurement of acyl CoA formation are discussed.

VALIDATION OF A "CLEARING" ASSAY FOR MILK LIPOPROTEIN LIPASE IN AGAROSE GEL. D.E. Wilson, C.M. Flowers and J.C. Reading (Depts. of Med. and Community and Family Med., The Univ. of Utah, College of Med., Salt Lake City, Utah 84112). *J. Lipid Res.* **14**, 124–8 (1973). We have developed a simplified method for the quantitative assay of lipoprotein lipase in cow's milk based on the hydrolysis of a glyceride emulsion in semisolid agarose gel. The area of clearing produced thereby is a function of enzyme concentration. Absolute molar rates for unknown samples may be calculated if standards of known activity are used concurrently. The precision of the simplified assay compared favorably with a method based on titrimetric determination of the rate of free fatty acid release. A modified assay has been used to assess the potency of lipoproteins in lipoprotein lipase activation.

SPHINGOLIPID COMPOSITION OF HUMAN PLATELETS. R.V.P. Tao, C.C. Sweeley and G.A. Jamieson (Dept. of Biochem., Mich. State Univ., East Lansing, Mich. 48823). *J. Lipid Res.* **14**, 16–25 (1973). Total lipid extracts from washed trypsinized human platelets were fractionated into neutral lipids, glycosphingolipids and phospholipids by silicic acid chromatography. The concentrations and chemical structures of the neutral and acidic glycosphingolipids were then studied in detail. On the basis of sugar molar ratios, studies of permethylation products, and the action of stereospecific glycosidases on the lipids, identifications were made of four neutral glycosphingolipids. Lactosylceramide was the most abundant type and accounted for 64% of the total neutral glycolipid mixture. The major fatty acids of the lactosylceramide were 20:0, 22:0, 24:0 and 24:1; the major long-chain base was 4-sphingenine. The platelets were surprisingly rich in a ceramide fraction, which represented 1.3% of the total platelet lipids. It had a different fatty acid composition than the neutral glycosphingolipid and ganglioside fractions. Hematoside was also isolated from the total lipid fraction of platelets; the neuraminic acid component was N-acetylneuraminic acid.

EARLY CHANGES IN PLASMA LIPOPROTEIN STRUCTURE AND BIOSYNTHESIS IN CHOLESTEROL-FED RABBITS. G. Camejo, V. Bosch, C. Arreaza and H.C. Mendez (Centro de Biofis. y Bioquim., Inst. Venezolano de Invest. Cientificas, and Catedras de Bioquim. y Fisiopatol., Inst. de Med. Exp., Univ. Central de Venezuela, Caracas, Venezuela). *J. Lipid Res.* **14**, 61–68 (1973). Plasma lipoproteins of $d < 1.063$ g/ml from rabbits fed a diet containing 1% cholesterol for 4 days showed changes in concentration and rates of flotation as determined by analytical ultracentrifugation. A marked increase in cholesteryl ester content of lipoprotein with $d < 1.019$ g/ml was the most prominent change in rabbits fed the diet for 21 days. Gel electrophoresis and immunochemical procedures demonstrated that in control and hypercholesterolemic rabbits there were some common apolipoproteins found in all lipoproteins with density < 1.063 g/ml. In control rabbits, there were also apolipoproteins specific to the lipoprotein fraction with $d < 1.019$ and to the fraction with d 1.019–1.063 g/ml. However, in rabbits fed the hypercholesterolemic diet for 21 days, the apolipoproteins characteristic of fraction 1.019–1.063 were the most abundant in the fraction with $d < 1.019$ g/ml. Liver slices from rabbits fed the high cholesterol diet for 7 and 21 days incorporated more L-[^{14}C]leucine into very low density and low density lipoproteins than controls.

BRANCHED-CHAIN AND ODD-NUMBERED FATTY ACIDS AND ALDEHYDES IN THE NERVOUS SYSTEM OF A PATIENT WITH DERANGED VITAMIN B_{12} METABOLISM. Yasuo Kishimoto, M. Williams, H.W. Moser, C. Hignite and K. Biemann (E.K. Shriver Center for Mental Retardation, Waltham, Mass. 02154). *J. Lipid Res.* **14**, 69–77 (1973). A mixture of isomers of methylhexadecanoic acid was isolated from glycerolipids of brain, spinal cord and sciatic nerve of a patient who died from methylmalonic aciduria, a disease in which vitamin B_{12} is not converted to deoxyadenosyl B_{12}. The isomers were identified by gas-liquid chromatographic-mass spectrometric analyses, and the data indicated that the points of methyl branching are located predominantly on the even-numbered carbon atoms. The concentration of these branched-chain acids among the glycerolipid fatty acids in the patient's nervous system was at least 0.3–0.9%, while the control tissues contained no more than a trace amount, if any, of these acids. In the spinal cord, these branched acids were distributed among all phosphatides and were in highest concentration on the β position of phosphatidylcholine. On the other hand, most extraneural tissues contained these acids in much lower concentrations; there were only trace amounts in liver, kidney, muscle and skin, and 0.2, 0.2 and 0.5% in total ester-linked fatty acids in spleen, duodenum and lung, respectively. A second abnormality was the 6-13-fold increase in 15:0 and 17:0 fatty acids in all of the glycerolipids in the nervous system of the patient.

25-HYDROXYCHOLECALCIFEROL-1-HYDROXYLASE. SUBCELLULAR LOCATION AND PROPERTIES. R.W. Gray, J.L. Omdahl, J.G. Ghazarian and H.F. DeLuca (Dept. of Biochem., College of Agr. and Life Sci., Univ. of Wis., Madison, Wis. 53706). *J. Biol. Chem.* **247**, 7528–32 (1972). The subcellular distribution and properties of an enzyme system responsible for hydroxylation of 25-hydroxycholecalciferol (25-OHD$_3$) to 1,25-dihydroxycholecalciferol (1,25-(OH)$_2$D$_3$) in chicken kidney have been described. The enzyme, which is highly specific for 25-OHD$_3$ since it does not hydroxylate cholecalciferol or dihydrotachysterol, is located in the mitochondria and has a pH optimum of 7.4 and an apparent K_m of 2.2×10^{-6}M. At 8.34×10^{-7}M 25-OHD$_3$, 1,25-(OH)$_2$D$_3$ production is linear with mitochondrial protein concentrations of up to 8 mg per ml. Enzyme activity is maximal in Tris-acetate buffer and requires Mg^{2+}, oxygen and a source of reduced pyridine nucleotide, which in intact mitochondria is provided by malate or succinate. The succinate-supported hydroxylation is dependent upon electron transport and oxidative phosphorylation. The 25-OHD$_3$-1-hydroxylase is inhibited by carbon monoxide, metapyrone, *p*-chloromercuribenzoate and diphenyl-*p*-phenylenediamine. The enzyme is also inhibited by its product, 1,25-(OH)$_2$D$_3$. It is, however, much more strongly inhibited competitively by 25-hydroxydihydrotachysterol, a close analog of 1,25-(OH)$_2$D$_3$.

ROLE OF 1,25-DIHYDROXYCHOLECALCIFEROL IN CALCIFICATION OF BONE AND MAINTENANCE OF SERUM CALCIUM CONCENTRATION IN THE RAT. Yoko Tanaka, H. Frank and H.F. DeLuca (Dept. of Biochem., College of Agr. and Life Sci., Univ. of Wis., Madison, Wis. 53706). *J. Nutr.* **102**, 1569–78 (1972). 1,25-Dihydroxycholecalciferol, the metabolite of vitamin D believed to be the metabolically active form in the stimulation of intestinal calcium absorption and in the mobilization of calcium from bone, is much less effective administered orally and chronically than either cholecalciferol or 25-hydroxycholecalciferol in the calcification of bone and in the elevation of serum calcium concentration of rats. However, when it is administered either intravenously or intraperitoneally, it is

at least as effective and possibly more effective than 25-hydroxycholecalciferol in the elevation of plasma calcium and phosphorus and in the calcification of bone. These results are consistent with the concept that 1,25-dihydroxycholecalciferol represents the hormonal form of vitamin D responsible for the maintenance of serum calcium at the expense of either bone or diet.

CHOLESTEROL METABOLISM IN RABBITS WITH OLEIC ACID-INDUCED CHOLELITHIASIS. P.A. Kyd and I.A.D. Bouchier (Med. Unit, the Royal Free Hosp., London WCIX 8LF, U.K.). *Proc. Soc. Exp. Biol. Med.* 141, 846–9 (1972). Cholesterol metabolism has been studied in rabbits with bile acid gallstones induced by feeding a high oleic acid diet. Lithogenic rabbits showed hypercholesterolemia, compared with control rabbits, which was accompanied by a striking increase in the liver cholesterol concentration. The lithogenic diet contained no supplementary cholesterol and hepatic cholesterol synthesis was markedly inhibited. Since intestinal cholesterol synthesis was unchanged in lithogenic rabbits, it is suggested that the increase in plasma and liver cholesterol concentrations was due to increased absorption of cholesterol by the presence of 15% oleic acid in the lithogenic diet. The significance of the raised plasma and liver cholesterol concentrations in relation to the 2–3 fold increase in cholestanol concentration in the livers of lithogenic rabbits remains to be elucidated.

• Drying Oils and Paints

EPOXY COMPOSITION BASED ON EPOXY RESIN, GOSSYPOL RESIN AND CROSS-LINKING AGENT. A.M. Zamyshlyaeva and R.L. Malinkevich. *U.S.S.R. 313,838.* The gossypol resin is the by-product from alkali-refining of cottonseed oil and contains phenols, aldehydes, acids and other active groups. Polyethylene polyamine is used as curing agent. Typical compositions comprise (in %) 70–90 epoxy resin, 10–30 gossypol resin and 7–9 polyethylene polyamine (I). For example, gossypol resin is slowly added with stirring to a homogeneous blend of epoxy resin and (I) at 20–40C. and the mixture is stirred until homogeneous to give a composition, with a useful pot-life under 3 hr, which may be used for lacquers and paints. This composition is superior to epoxy resin alone. Impact resistance is increased from 20 to 50 kg./cm. and chemical resistance to 10% H_2SO_4 is increased by 35%. (World Surface Coatings Abs. No. 366)

INVESTIGATIONS OF SURFACTANTS IN PAINTS. H. Haagen (Technical Application Div., Res. Inst. for Pigments & Varnishes, e. V., (Stuttgart). *Farbe u. Lack* 78(12), 1156–62 (1972). After the drying of a coating material the surfactants remain in the dry paint film. They may have an effect not only on the preparation and storage stability but also on additional properties of technological importance. The investigation shows that the selection of the surfactants should be influenced not only by the effects desired but by those side-effects on coating material properties which are important in the field of application.

EFFECTS OF THE STOVING CONDITIONS ON THE FILM PROPERTIES OF ALKYD MELAMINE RESIN COMBINATIONS. J.F.A. Hazenberg and M. Hoeflaak (Verfinstituut TNO, Delft, Netherlands). *Farbe u. Lack* 78(12), 1151–6 (1972). The effects of the stoving conditions on the film properties of some alkyd melamine resin combinations were investigated with a standard laboratory oven and with an improved gradient oven. The investigation has shown that only exactly defined conditions result in good reproducibility. The improved gradient oven permits investigation of the whole range of stoving conditions in one run.

SPREADING AND WETTING OF AN ELECTRODEPOSITION COATING. C. Hansen and J. Knudtson (PPG Ind. Inc., Springdale, Pa.). *Farbe u. Lack* 79(2), 115–7 (1973). The wettability of an electrodeposition coating was systematically improved to the point where water would not bead up after it was applied as a film. Spreading and wetting measurements were made on a number of modifications of the original coating and support a solubility parameter interpretation of surface wetting. Roughness improved the wetting significantly; soaking the rough panels improved it to product acceptability.

WETTING AS AN AID TO PIGMENT DISPERSION. P.R. Buechler, G.L. Brown, V.P. Parikh and H.J. Salmon (Mobil Chem. Corp.). *J. Paint Technol.* 45(577), 60–64 (1973). The bottle neck in most coating manufacturing operations is pigment dispersion—particularly with hard-to-wet organic pigments. The critical surface tension was determined for several of these troublesome pigments. Using this information and also by reducing the forces opposing wetting, it is possible to make a stable pigment dispersion more readily. An example is given of the stable dispersion in water of an untreated phthalocyanine blue in rapid kinetic dispersing equipment.

• Detergents

SOAP TABLET PRODUCTION. E.H. Evans (Lever Bros.). *U.S. 3,723,329.* A process for the manufacture of soap tablets incorporating minor amounts of alkaline earth metal alkyl aryl sulfonates is described. In the process, the tablets are superfatted by the addition to a soap base of a minor amount of free alkyl aryl sulfonic acid. Preferably, the alkaline earth metal alkyl aryl sulfonates are formed in situ in a soap base by the addition to the base of an anhydrous alkaline earth metal oxide-sulfonic acid slurry, the tablets being formed subsequently from the soap base.

DETERGENT COMPOSITION. B.W. Sheflin. *U.S. 3,723,330.* A general purpose aqueous alkaline detergent composition particularly effective against oily soil consists of a homogeneous dispersion of soap or synthetic detergent, a volatile alkaline detergent builder, water and volatile, water insoluble, organic solvents of the halogenated hydrocarbon or dialkyl ether types. The composition is formulated as a concentrated emulsion, and at the time of use is diluted with water to obtain a working concentration.

SYNERGISTIC EMULSIFIER. A. Cahn, J.A. Aekilli and F.E. Carroll (Lever Bros.). *U.S. 3,723,356.* Emulsifying agents characterized by unusual mildness toward the skin are disclosed. They consist of a synergistic combination of a water soluble taurine salt of the general formula: $R_1 - CH(OH) - CH_2 - N(CH_3) - CH_2 - CH_2 - SO_3M$. R_1 is an alkyl radical having 12–18 carbon atoms or an oxyalkyl radical and M is a water solubilizing cation. The other parts of the combination are a surface active organic sulfate or sulfonate detergent which may be amidomethanesulfonate, and acyl isothionate, or an N-(acyloxyethyl) sulfoacetamide. The taurine salt and the detergent are present in a specified weight ratio to obtain maximum synergism.

LIQUID DETERGENT COMPOSITION. K.R. Hansen (Colgate-Palmolive). *U.S. 3,723,357.* The composition consists of N(2-hydroxy C_{10}–C_{18} alkyl) derivatives of N-methyl taurine or sarcosine or diethanolamine in combination with an anionic sulfate, zwitterionic or amphoteric detergent in an aqueous medium. The addition of the derivatives results in increased foam-drainage times and better foaming properties.

NEW POLYMERS AND DETERGENTS CONTAINING THEM. F.E. Hardy, P. Robson and P.R.H. Speakman (Procter & Gamble). *U.S. 3,719,647.* New copolymers of acrylic and methyl acrylic acid (I) with acrylic and methacrylic acid-ethylene oxide condensates (II) are disclosed. The components have formulas: (I) $CH_2 = C(R)COOM$; (II) $CH_2 = C(R)COO(C_2H_4O)_nH$, in which R is H or CH_3; M is H, alkali metal, ammonium or amine; and n is at least 1. The preferred ratio of (I) to (II) is about 2:1 and n is preferably 20–100. These compounds are mixed with surface active agents to form built detergents with improved whitening properties. ■