Abstracts

• Fats and Oils

SUPEROXIDE AND SINGLET OXYGEN IN MILK LIPID PEROXIDATION. L.W. Aurand, N.H. Boone and G.G. Giddings (Dept. of Food Sci., North Carolina State Univ., Raleigh, N.C. 27607) *J. Dairy Sci.* **60**, 363–9 (1977). The extent to which the excited state molecular oxygen species, singlet oxygen $^1O_2$, participates in milk lipid oxidation was considered. Light, copper, and enzyme catalyzed oxidative reactions were included in the study. The working hypothesis was that superoxide anion was produced in the copper and xanthine oxidase systems and that the anion once produced underwent chemical dismutation to singlet oxygen; light-catalyzed reactions produced singlet oxygen directly through mediation of a triplet sensitizer. In the presence of a singlet oxygen trapper (1,3-diphenylisobenzofuran) or a single oxygen quencher (1,4-diazabycyclo-[2-2-2] octane), lipid oxidation was inhibited. Too, superoxide dismutase acted as a protective agent against lipid oxidation by catalyzing superoxide dismutation to ground state oxygen, thus circumventing singlet-generating spontaneous dismutation. Singlet oxygen is the immediate source of the hydroperoxides that initiate lipid oxidation catalyzed by the three agents.

A NEPHELOMETRIC PROCEDURE FOR DETERMINATION OF FAT AND PROTEIN IN MILK. D.C. Beitz, M.E. Phillips and S.H. Eklund (Dept. of Animal Sci., Iowa State Univ., Ames, Ia.) *J. Dairy Sci.* **60**, 701–5 (1977). A nephelometric procedure for determination of protein and fat content of milk has been developed. Protein is aggregated into a colloidal suspension by the addition of an anhydrous reagent consisting of 15. 75% acetic anhydride, 83.75% glacial acetic acid, and .5% p-toluenesulfonic acid. Protein then is redissolved, and the fat is aggregated into an emulsion by addition of an aqueous surfactant solution consisting of 5% IGEPAL DM-970 in water. Correlation of the nephelometric method with the standard Kjeldahl protein determination gives a coefficient of .977; that of the nephelometric and Babcock fat determination gives .985.

THE POLAR LIPIDS OF GROUP B STREPTOCOCCI I. GLUCOSYLATED DIPHOSPHATIDYGLYCEROL, A NOVEL GLYCOPHOSPHOLIPID. W. Fischer (Inst. of Physiological Chem., Univ. of Erlangen-Nurnberg, D-8520 Erlangen, Wasserturmstr. 5 [G.F.R.]) *Biochim. Biophys. Acta* **487**, 74–88 (1977). 1. From group B *Streptococci* a novel glycophospholipid was isolated, which contained D-glucose, glycerol, acyl groups and phosphorus in a molar ratio of approx. 1:3:4:2. It was established to be 2'O-α-D-glucopyranosyl-, 1',3'-bis(1,2-diacyl-*sn*-glycer-3-phospho-)-glycerol. 2. The structure of the deacylated core was accomplished by analyses of the breakdown products obtained on (i) strong alkaline hydrolysis, (ii) Smith-degradation, and (iii) periodate oxidation with subsequent hydrazinolysis. The four acyl groups were located by sequential degradation of the native lipid with phospholipase A_2 and 98% acetic acid. 2. In group B *Streptococci* approximately 25% of diphosphatidylglycerol occurs in the form of its glucosylated derivative which accounts for 18% of the lipid phosphorus. The glucosylated phosphatidylglycerol analogue could not be detected. A phosphoglucolipid, however, was present, which was tentatively identified as glycerophosphodiglucosyldiacylglycerol.

ETUDE DES STEROLS D'ALGUES BRUNES DU GENRE CYSTOSEIRA. IDENTIFICATION PAR CHROMATOGRAPHIE GAZ-LIQUIDE COUPLEE A LA SPECTROMETRIE DE MASSE. C. Francisco, G. Combaut, J. Teste and B.F. Maume (Laboratoire de Chimie des Substances Naturelles Marines, Centre Universitaire de Perpignan et Laboratoire de Biochimie des Interactions Cellulaires, Universite de Dijon, France) *Biochim. Biophys. Acta* **487**, 115–21 (1977). Study of sterols from brown seaweeds of the genus *Cytoseira*. Identification by gas-liquid chromatography coupled with mass spectrometry. All the samples of brown seaweeds (Cystoseira) that we have studied present the same Δ^5 sterols, fucosterol, cholesterol, 22-*trans*-dehydrocholesterol, brassicasterol, 24-methylene cholesterol as well as cystosterol, a new C_{27} sterol. This sterol has been submitted to gas-liquid chromatographic-mass spectrometric analysis.

CONJUGATED POLYENE FATTY ACIDS ON FLUORESCENT PROBES: SPECTROSCOPIC CHARACTERIZATION. L.A. Sklar, B.S. Hudson, M. Petersen, and J. Diamond (Dept. of Chem., Stanford Univ., Stanford, Calif. 94305) *Biochemistry* **16**, 813–8 (1977). This paper is the first in a series which extends introductory studies of parinaric acid and its phospholipid derivatives as membrane probes. Parinaric acid has a conjugated tetraene chromophore and exhibits many spectroscopic properties common to linear polyenes. Its absorption spectrum is characterized by a strong near-ultraviolet transition with vibronic structure, which is strongly affected by solvent polarizability. The fluorescence emission occurs at considerably lower energy than the absorption and the wavelength of the emission is nearly independent of the solvent. The fluorescence quantum yield and lifetime are strongly affected by temperature and solvent. These spectral features are interpreted in terms of an excited electronic-state order such that a weak transition occurs at longer wavelengths than the strongly allowed transition which dominates the absorption.

CONJUGATED POLYENE FATTY ACIDS AS FLUORESCENT PROBES: SYNTHETIC PHOSPHOLIPID MEMBRANE STUDIES. L.A. Sklar, B.S. Hudson, and R.D. Simoni (Depts. of Chem. and Biol. Sci., Stanford Univ., Stanford, Calif. 94305) *Biochemistry* **16**, 819–28 (1977). The preparation of polyene fatty acid membrane probes *cis-* and *trans*-parinaric acid and parinaroylphosphatidylcholines and their use in studies of several one- and two-component lipid systems are described. The fluorescence quantum yield of *trans*-parinaric acid in dipalmitoylphosphatidylcholine at 20°C is approximately 0.3; the quantum yield in aqueous solution is negligibly small. Thermal-phase transitions in single-component phospholipid dispersions are monitored with absorption and fluorescence excitation peak position, fluorescence intensity, lifetime, and polarization. The transition temperatures observed are consistent with previous determinations. The phase diagram of the binary lipid mixture dipalmitoylphosphatidylcholine-dipalmitoylphosphatidylethanolamine has also been examined and found to be essentially identical to the one constructed using a nitroxide probe.

PHYSICAL PROPERTIES OF THE DIMYRISTOYLPHOSPHATIDYLCHOLINE VESICLE AND OF COMPLEXES FORMED BY ITS INTERACTION WITH APOLIPOPROTEIN C-III. K.C. Aune, J.G. Gallagher, A.M. Gotto, Jr. and J.D. Morrisett (Dept. of Med. and Biochem., Baylor Coll. of Med., Methodist Hosp., and the Dept. of Chem. Engr., Rice Univ., Houston, Texas). *Biochemistry* **16**, 2151–6 (1977). The structure of a single bilayer vesicle of dimyristoylphosphatidylcholine has been characterized by sedimentation, densimetry, and light-scattering measurements. The molecular weight, partial specific volume, Strokes radius, and degree of hydration were found to be 2.68×10^6, 0.972 cm^3/g, 125 Å, and 0.86 g/g, respectively. From these quantities, a spherically symmetrical model has been derived that features a phospholipid bilayer 35.5 Å thick and a hydration shell 9.3 Å thick. This particle was shown to bind apolipoprotein C-III (apoC-III) up to 0.08 g/g without loss of its original vesicular structure. At protein-lipid ratios in excess of 0.08 g/g, sedimentation, gel chromatography, and light-scattering measurements indicated a dramatic decrease in Stokes radius and molecular weight. The sedimentation data showed these parameters to become constant at protein-lipid ratios in excess of 0.25 g/g. In this region, the Stokes radius and molecular weight were found to be ~80 Å and 442,000 respectively. Within the constraints of these values and other data, several models for this complex are discussed.

THE USE OF ^{13}C SPIN RELAXATION TO INVESTIGATE MOLECULAR MOTION IN LIQUID TRISTEARIN. P.T. Callaghan (Dept. of Chem., Biochem., and Biophys., Massey Univ., Palmerston North, New Zealand). *Chem. Phys. Lipids* **19**, 56–73 (1977). Longitudinal and transverse ^{13}C spin relaxation times have been used to investigate the molecular motion of tristearin over a range of temperatures in the melt. Overall molecular rotational diffusion rates have been obtained as well as the diffusion rates about successive bonds in the stearoyl chains. The data can be explained using an anisotropic rotor model in which

the fast and slow molecular diffusion rates are $\geqslant 8 \times 10^{9}$ sec^{-1} and $0.018(2) \times 10^{9}$ sec^{-1} respectively. The alignment of the fast diffusion axis is close to the long chain axis of the 'tuning fork' model and the existence of such a configuration in the melt is supported by the observation of different relaxation times for the two chemically equivalent primary glyceryl carbons. The low flexibility gradient and high end group mobility of the acyl chain found at low temperatures in the melt is similar to that observed in lipid vesicle studies and suggest that the chains are aligned parallel. A break down of this short range order is apparent above 150°C.

STEAM DISTILLATION-SOLVENT EXTRACTION RECOVERY OF VOLATILES FROM FATS AND OILS. R. Teranishi, E.L. Murphy and T.R. Mon (Western Regional Res. Lab., Agr. Res. Service, U.S. Dept. of Agr., Berkeley, CA) *J. Agric. Food Chem.* **25**, 464–6 (1977). This system combines steam distillation and liquid-liquid extraction to recover volatiles from fats and oils. Oil is pumped in at the top of a spinning-band distillation column, in which the oil is heated to 100°C and spread to a thin film. As the oil film drops down to the pot, steam, which is introduced at the bottom, travels upward to strip the volatiles from the oil. The steam distillate is extracted in a liquid-liquid extractor incorporated in the system, and the extracted water is recycled as steam. Stripped oil in the pot serves as a liquid seal to force steam up the column. The level of the oil in the pot is maintained automatically by an overflow system. Many liters of oil can be pumped through this system to be stripped of volatiles by steam. The volatiles can be isolated easily from the small amount of solvent recycled in the liquid-liquid extractor.

QUANTITATIVE ANALYSIS OF PLASMA NEUTRAL GLYCOSPHINGOLIPIDS BY HIGH PERFORMANCE LIQUID CHROMATOGRAPHY OF THEIR PERBENZOYL DERIVATIVES. M.D. Ullman and R.H. McCluer (Eunice Kennedy Shriver Center for Mental Retardation, Inc., W.E. Fernald State Schl., Walttham, Ma.). *J. Lipid Res.* **18**, 371–8 (1977). A quantitative high performance liquid chromatography method for the analysis of neutral glycosylceramides as their perbenzoyl derivatives has been devised. Samples containing more than 2.5 nmol each of mono-, di-, tri-, and tetra-glycosylceramide are benzoylated with 10% benzoyl chloride in pyridine at 37°C for 16 hr. The products are separated from excess reagents by solvent distribution and injected onto a pellicular silica gel (Zipax) column (2.1 mm × 50 cm). The derivatives are eluted with a 10 min linear gradient of 2–17% ethyl acetate in hexane at 2 ml/min and absorbance at 280 nm is recorded. The detector response was proportional to the weight of sample used (2–30 nmol) and the lower limit of detection was about 70 pmol. The procedure has been applied to the quantitative analysis of erythrocyte and plasma glycolipids. As little as 0.5 ml of plasma can be used for analysis. The relative standard deviation of repetitive analyses ranged between 2.0% for glucosylceramide to 5.4% for galactosyllactosylceramide.

RAMAN SPECTRA AND CONFORMATION OF THE GLYCEROPHOSPHORYLCHOLINE HEADGROUP. Y. Koyama, S. Toda and Y. Kyogoku (Faculty of Sci., Kwansei Gakuin Univ., Uegahara, Nishinomiya 662, Japan). *Chem. Phys. Lipids* **19**, 74–92 (1977). The Raman and infrared spectra of L-α-glycerophosphorylcholine (GPC) and its cadmium complex (GPC · $CdCl_2$) have been measured for both the anhydrous and hydrate crystals. The Raman spectra of GPC and GPC · $CdCl_2$ dissolved in water are also reported. All the spectra fall into three distinctive patterns, demonstrating the existence of at least three different kinds of rotational isomers; the first is stable in the anhydrous GPC crystal, the second is stable in the GPC hydrate and GPC · $CdCl_2$ hydrate crystals and the third is stable in the anhydrous GPC · $CdCl_2$ crystal and also in aqueous solutions of GPC and GPC · $CdCl_2$. The latter form is reported for the first time. Raman spectra of dimyristoyl-L-α-phosphatidylcholine (L-DMPC) and dipalmitoyl-(L and DL)-α-phosphatidylcholine (L and DL-DPPC)hydrates have been measured below, around and above the transition temperature T_t. The spectra below T_t indicate that the conformation of the glycerophosphorylcholine (GPC) headgroup in the L-DMPC crystal is similar to that in the L-DPPC crystal. The headgroup conformation in the DL-DPPC crystal is quite different from that in the L-DPPC crystal, although they became the same after a crystalline transformation of DL-DPPC. The changes of the Raman spectra through the T_t transitions for the above crystals show that the headgroup conformation remains unchanged at the beginning of the transition.

UV-SYNTHESIS OF AMPHIPHILIC MOLECULES FROM N-ALKANES AND ITS BIOLOGICAL SIGNIFICANCE. S.A. Seleznev, L.M. Ferodov, S.I. Kuzina and A.I. Mikhailov *Rev. Fr. Corps Gras* **24**(4), 191–3 (1977). This work has been undertaken in an attempt to follow one of possible ways of amphiphilic molecule formation—such as lipids or fatty acids—under simulated prebiotic conditions. The raw material employed is a commercial petroleum hydrocarbon (C12–C14), applied on an aqueous salt solution (typical of sea water). The solution is irradiated with a diffused UV-light. The synthesis of amphiphilic molecules seems reliable to the well-known mechanism of polymorphous phase transition in liotropic systems.

NEW METHOD FOR THE DETERMINATION OF THE GLYCERIC DISTRIBUTION. J.P. Wathelet, M. Severin, J. Ledieu and C. Deroanne *Rev Fr. Corps Gras,* **24**(4), 195–202 (1977). The method for calculating the glyceridic distribution described by Deroanne is improved by adjusting, with successive iterations, the Van der Wal distribution, not only with the coefficients obtained after determination of the separated triglycerides by thin-layer –AgNO3 chromatography, but also with the calculated coefficients after separation of triglycerides by gas-liquid chromatography.

REFINING AND SEGREGATION OF PALM OIL. A. Athanassiadis *Oleagioneux* **32**(4), 173–8 (1977). The recent and spectacular extension of the cultures of the palm tree in the world has made nowadays of the palm oil one of the most profitable vegetable oils to process. This oil is indeed specially easy to physically refine and it is well known that this process gives a much higher yield in refined oil than the classical caustic refining. A complete process, of which all the details are given, has been developed, which enables now to obtain refined oils of the same quality and the same stability as those obtained by the classical caustic refining. Everybody knows that physical refining gives much higher yields than caustic refining. (Refining ratio 1.1 instead of 1.8 to 1.7). In addition it does not produce any pollution and gives automatically distilled fatty acids of commercial quality. In addition segregation in two phases obtained by winterization gives an easy way to obtain more than 50 p. 100 of a fluid fraction of palm oil which has a much higher commercial value than the original oil. The article gives a description of the different successive phases and the required equipment of this process giving the highest yield of a liquid fraction of neutral, bleached and deodorized palm oil.

CYCLIC MONOMERS EVALUATION IN VEGETABLE OILS. M. Gente and R. Guillaumin *Rev Fr. Corps Gras* **24**(4), 211–8 (1977). A new method of evaluation for cyclic monomers in the vegetable oils uses a gas-liquid chromatography on polar column (7% BDS) of hydrogenated methylic esters. Under these conditions, model components obtained by alkaline isomerisation of linoleic acid are found on the chromatogram between the C18 and C20 esters. The percentage of cyclic monomers is evaluated by planimetry compared with an internal standard. This method is fast; its repeatability and reproductibility are good. The detection limit is about 0.1%. Refined unheated oils contain less than 0.2% cyclic monomers. Those are not practically formed under usual frying conditions (t. max; 220°C) for contents of linoleic acid below 20% in the oil. The use of glass capillary column (Carbowax 20 M-30 m) improves the method and time-saves for the evaluation.

THE ANALYSIS OF WAXES AND THEIR DETERMINATION IN INDUSTRIAL MIXTURES BY HIGH TEMPERATURE GL-CHROMATOGRAPHY. G. Valmalle and A. Karleskind *Rev. Fr. Corps Gras* **24**(4), 203–9 (1977). The field of application for the gas-liquid chromatography is extended by the use of some stationary phases at high temperatures. For instance, the waxes are analysed without pretreatment. The components of waxes or their ratio are specific for every kind of wax. The composition of several usual waxes has been determined by gas-liquid chromatography. These compositions are suitable to control the purity of waxes, to detect and determinate these waxes in industrial blends. This method is easier, faster and more precise than the other usual methods.

FATTY ACID AND STEROL CONTENT OF DIOSPYROS MAONTANA ROXB SEEDS. M.P. Goutam and R.M. Purohit. *Seifen, Ole, Fette, Wachse* **103**(4), 99–101 (1977). Diospyros maontana ROXB seeds yielded 1.5% of fixed oil. Saponification of the oil gave mixed fatty acids 82.5% and unsaponifiable matter 1.04%. Mixed fatty acids in the oil were identified by paper and thin layer chromatography and quantitatively estimated by column chromatographic separation of their methyl esters. Saponifiable portion of the oil was found to contain palmitic

27.91%, stearic 11.09%, oleic 37.08% and linoleic 23.92%, whereas the unsaponifiable matter could be established as lupeol, B-sitosterol and stigmasterol.

PARSUANCE OF THE GLYCEROLYTIC PROCESS OF LINSEED OIL BY MEANS OF GEL PERMEATION CHROMATOGRAPHY. A. Mandik, A. Tutalkova and J. Makes. *Farbe + Lack* **83**(3), 186–92 (1977). In order to follow the glycerolytic process of linseed oil a method based on gel permeation chromatography was developed for the determination of the concentrations of mono-, di- and triglycerides. For calibration purposes a mixture of glycerides of known composition was used. The glycerol content was determined by titration and stoichiometrically calculated. As the results show, the gel permeation chromatography permits a sensitive and reliable analysis of the glycerolysis products.

HYDROGENATION OF OILS WHICH CONTAIN LINOLENIC ACID. L.S. Golodova et al. *Maslo-zhir. Prom-st.* **1976**(6), 15–16. The results obtained by hydrogenation of soybean oil with 11% of linolenic acid are presented. Hydrogenation of linseed oil which contains 25% linolenic acid is also done. Hydrogenation is done in solution of ethanol at 65C and under atmospheric pressure. As catalysts, Pd/Al_2O_3 and nickel on support are used. The results show that the hydrogenation with nickel catalyst is less selective than with palladium. To obtain a maximum of monosaturated fatty acids, the quantity of saturated acids increases by 5 and 2%, respectively. (Rev. Fr. Corps Gras)

GENERAL MATHEMATICAL MODEL OF CATALYST HYDROGENATION OF FATS AND OILS. K.A. Zhubanov et al. *Maslo-zhir. Prom-st.* **1976**(6), 17–9. A method for the calculation of rate constants of the individual stages of hydrogenation of the fats and oils for unsaturated compounds is described. The hydrogenation was done with cottonseed oil with palladium as a catalyst (prepared by the Frampton method) at 200C and with a pressure of 1 kg/cm^2. The rate constants of the particular stages of hydrogenation were determined on the electronic calculator "Minsk-22" by the method of the least squares. The results show high trans isomerization power of the palladium catalyst. The described method of calculation allows one to characterize differentially and quantitatively the isomerization power of the hydrogenation catalysts. (Rev. Fr. Corps Gras)

INFLUENCE OF BLEACHING IN THE MISCELLA ON THE PROCESS OF FILTRATION. U.V. Klioutchkine et al. *Maslo-zhir. Prom-st.* **1976**(7), 18–20. The use of miscella instead of the oil in the bleaching process allows an increase in the efficiency of the filtration process since the yield of the filtrate is increased and the time of filtration is decreased. The theoretical calculation is confirmed experimentally. This can also be used in the calculation of the adsorption process through the layer of sorbent. In this paper, some rules for the filtration applied for the separation of the used adsorbent are examined. The experiments have been done with the miscella of soybean oil before distillation. (Rev. Fr. Corps Gras)

PREPARATION OF THE PURE PALMITIC ACID. N.D. Klotchko et al. *Maslo-zhir. Prom-st.* **1976**(6), 25–8. A method was elaborated to obtain palmitic acid from a fraction of the fatty acids separated from highly hydrogenated cottonseed oil. In this method, rectification is done on an installation with two columns. Pure palmitic acid is distilled from the second column. The method assures a high economical efficiency of the production and allows a decrease in the price of this product by about 18 times. (Rev. Fr. Corps Gras)

THE REFINING PROCESS OF VEGETABLE OILS. USSR patent no. 486038 "Inventions" 1975 no. 36. *Maslo-zhir. Prom-st.* **1976**(8), 43. The oil is treated with a mineral acid before hydration which is done with 0.3–0.5% (in relation to the mass of the oil) of a solution of 0.5–1.0% of polyacrylamide. The temperature of hydration is maintained between 60 and 90C. This method simplifies the refining of vegetable oils and allows the obtainment of a higher yield of refining oil by 4 to 6% (depending on the acid value of the oil). (Rev. Fr. Corps Gras)

• Drying Oils and Paints

THE PART PLAYED BY THE STRUCTURE OF CARBOXYLIC ACIDS IN MAKING SOLIDS HYDROPHOBIC. A. Serrano, D. Bochnia and H. Schubert. *Tenside Deterg.* **14**(2), 67–73 (1977). The structure of ionic surfactants plays an important role for their adsorption on solid surfaces in aqueous solutions and thus for their technological activity. For the examinations the structure of alcane carboxylic acids was varied by substitution with –Br and –COOH (1-Br-alcane carboxylic acids, 1,1- and 1,2 alcane dicarb. a.). The changes in the adsorption behavior are explained by dimerization, dissociation, association within the films and interactions between the polar groups and the solid surfaces (corundum, cassiterite).

WATER DILUTABLE, DISPERSIBLE, AND EMULSIFIABLE EPOXY RESINS. E.G. Bozzi and R.C. Nelson (CIBA-Geigy Corp.), *J. Coatings Technol.* **49**(628), 61–5 (1977). Three procedures have been developed for introducing epoxy resins into waterborne ecologically acceptable systems: (1) Epoxy resins modified with hydrophilic moieties that make the polymer water dilutable; (2) Resin/surfactant combinations that can be dispersed in water; and (3) Resin/surfactant/solvent mixtures that can subsequently be emulsified. Each procedure results in systems that have specific advantages and disadvantages. Recently, formulations containing a water dilutable epoxy resin in the aqueous phase and either an emulsified or dispersed resin in the particulate phase were prepared. Preliminary data indicate that these systems possess the processing and handling properties of standard solvent systems and the performance of epoxy resins.

ELECTRO-CAPILLARY BEHAVIOR OF HOMOLOGOUS DODECANOL POLYGLYCOL ETHERS. E. Müller and H.-D. Dörfler. *Tenside Deterg.* **14**(2), 75–9 (1977). The electro-capillary behavior of homologous dodecanol polyglycol ethers, $C_{12}H_{25}$–O–$(CH–CH_2O)_xH$ with x = 4.5 or 8 was studied using the droplet time method. Determinations of concentration and potential dependence of surface tension showed that this is systematically lowered with increasing concentration. Characteristically, the preferred surface tension reduction at potential values is more negative than that of the electro-capillary maximum. The evaluation of the electro-capillary curves produces very high interfacial concentrations γ ($= 18 \times 10^{10}$ mol/cm^2) which are not compatible with monolayer absorption. Presumably in the positive region of the electro-capillary curve it is the alkyl groups that are preferably adsorbed on the mercury surface, whilst in the negative region it is the ethylene oxide units. The critical micelle formation concentration determined from the Gibbs isotherms and the Esin-Markow graph leads to almost identical values. As far as the liquid/gas interface is concerned however, differences in the cmc values are observed and it is quite possible that these differences are connected with the specific molecular orientation and polylayer formation.

TECHNOLOGY OF EPOXIDE RESINS. W. Brushwell. *Farbe + Lack* **83**(3), 194–200 (1977). Detailed description of the state of developments of epoxide coatings. A second part cites contributions from the USSR. Finally a survey of patents of the last years is given.

• Biochemistry and Nutrition

DIRECT DESATURATION OF EICOSATRIENOYL LECITHIN TO ARACHIDONOYL LECITHIN BY RAT LIVER MICROSOMES. E.L. Puch and M. Kates (Dept. of Biochem., Univ. of Ottawa, Ottawa, Canada K1N 6N5) *J. Biol. Chem.* **252**, 68–73 (1977). A microsomal enzyme system from rat liver was shown to catalyze desaturation, in presence of reduced pyridine nucleotides and oxygen, of 1-acyl-2-[^{14}C]eicosatrienoyl-*sn*-glycero-3-phosphorylcholine to 1-acyl-2-[^{14}C]arachidonoyl-*sn*-glycerophosphorylcholine. This desaturation was linear with time and proportional to microsomal protein concentration, and proceeded with no significant breakdown of the lecithin substrate. The microsomal enzyme system will also desaturate 1,2-di-[^{14}C]eicosatrienoyl-*sn*-glycero-3-phosphorylcholine and [^{14}C]eicosatrienoyl-CoA, but not free [1-^{14}C] eicosatrienoic acid in the absence of ATP, Mg^{2+}, and CoA. Desaturation of 1-acyl-2-[^{14}C]eicosatrienoyl-glycerophosphorylcholine as well as [^{14}C]eicosatrienoyl-CoA was dependent on oxygen and either NADH or NADPH, and was inhibited by cyanide but not by carbon monoxide, indicating the involvement of cytochrome b_5 and not P_{450}. The activity of both eicosatrienoyl-glycerophosphorylcholine desaturase and the eicosatrienoyl-CoA desaturase was increased in rats that had been starved for 48 h and refed a fat-free diet. These data indicate the existence of a new route to synthesis of arachidonate, namely, by desaturation of eicosatrienoyl lecithin to arachidonoyl lecithin.

ENZYMIC SYNTHESIS OF ETHER TYPES OF CHOLINE AND ETHANOLAMINE PHOSPHOGLYCERIDES BY MICROSOMAL FRACTIONS FROM RAT BRAIN AND LIVER. A. Radominska-Pyrek, J.

Strosznajder, Z. Dabrowiecki, G. Goracci, T. Chojnacki, and L.A. Horrocks (Polish Academy of Sciences, Warsaw, Poland) *J. Lipid Res.* 18, 53–8 (1977). The formation of product by ethanolaminephosphotransferases (EC 2.7.8.1) and cholinephosphotransferases (EC 2.7.8.2) in microsomal fractions from brains and liver of mature rats is increased several fold by 1,2-diacyl-*sn*-glycerols. With the addition of 1-alkyl-2-acyl-*sn*-glycerols, we have found an 11-fold increase with brain microsomes and a 20-fold increase with liver microsomes in the synthesis of choline ether lipids (1-alkyl-2-acyl- and 1-alk-1′-enyl-2-acyl-*sn*-glycero-3-phosphorylcholines). For the synthesis of ethanolamine ether lipids (1-alkyl-2-acyl-and 1-alk-1′-enyl-2-acyl-*sn*-glycero-3-phosphorylethanolamines), the stimulation of alkylacylglycerols was 7-fold for brain microsomes and 18-fold for liver microsomes. The alkylacyl glycerols (8 mM) also inhibited the synthesis of diacyl phosphoglycerides by 44 to 65%, indicating that the same ethanolaminephosphotransferases and cholinephosphotransferases are utilized for the synthesis of alkylacyl phosphoglycerides and diacyl phosphoglycerides.

EFFECT OF WHEAT FLOUR AND ITS STARCH AND GLUTEN COMPONENTS OF LIPID METABOLISM IN CHOLESTEROL-FED RATS. G.S. Ranhotra, R.J. Loewe and L.V. Puyat (Nutr. Lab., American Inst. of Baking, Chicago, Ill. 60611) *J. Food Sci.* 42, 79–82 (1977). In short-term studies, substitution of dietary sucrose (50%) with wheat flour or its starch component caused, in hypercholesteremic young male rats, and appreciable decrease in fatsing serum cholesterol levels; patent flour showed a more pronounced such effect than high-extraction flour. Liver cholesterol levels were also greatly reduced in flour-fed rats. Substitution of dietary sucrose with wheat gluten substantially reduced the liver accumulation of cholesterol, triglycerides and phospholipids but lowering effect on serum lipids was mostly observed under the condition of partial, rather than complete, substitution of sucrose with gluten.

LECITHIN INHIBITS FATTY ACID AND BILE SALT ABSORPTION FROM RAT SMALL INTESTINE IN VIVO. D.R. Saunders and J. Sillery (Dept. of Med., Univ. of Washington, Seattle, Wash. 98195) *Lipids* 11, 830–2 (1976). During digestion of a fatty meal, long chain free fatty acids (FFA) and lecithin are among the lipids solubilized in intestinal contents as mixed micelles with bile salts. We hypothesized that if lecithin were not hydrolyzed, the mixed micelles would be abnormal, and absorption of FFA and bile salts would be depressed. To test this hypothesis, isolated segments of rat small intestine were infused in vivo with micellar solutions of 2 mMolar linoleic acid and 10 mMolar taurocholate to which was added 3 mMolar 1-palmitoyl, 2-oleoyl lecithin (a common lecithin in bile and food), or 1-palmitoyl lysolecithin (the hydrolytic product of lecithin). Absorption of FFA and bile salt was measured under steady state conditions using a single-pass technique. Lecithin depressed the rate of FFA absorption by 40% ($p < 0.025$) in jejunal and ileal segments whereas lysolecithin was associated with normal rates of FFA absorption. Lecithin also reduced taurocholate absorption from the ileum by 30% ($p < 0.05$). These data support the idea that lecithin may depress FFA and bile salt absorption from the small intestine in pancreatic insufficiency.

RESPONSE TO THE HYPOBETALIPOPROTEINEMIC AGENT ADAMANTYLOXYPHENYL PIPERIDINE IN HYPERLIPEMIC RATS. P.E. Schurr and C.E. Day (Diabetes and Athero. Res., The Upjohn Co., Kalamazoo, Mich. 49001) *Lipids* 12, 22–8 (1977). The hypobetalipoproteinemic activity of U-41,792 (1-[p-(1-adamantyloxy)-phenyl]-piperidine) is a marked and selective reduction of heparin precipitating lipoproteins (low density plus very low density lipoproteins) in cholesterol-cholic acid induced hypercholesterolemic rats. This activity consists of both a reduction in heparin precipitating lipoproteins (HPL) and an increase in high density lipoproteins that are not precipitated by heparin. The increase in high density lipoproteins is routinely noted by decreases in HPL/cholesterol ratios. The pattern of response following single 100 mg/kg doses of U-41,792 was determined. After an I.V. dose was administered in a cottonseed oil emulsion, serum cholesterol levels were reduced, beginning at 8 hr. after administration and persisting for 96 hr. Similar results, though delayed somewhat, were obtained after a single oral dose. Activity was accompanied by increases in weight and cholesterol content of livers. After multiple, daily, oral doses, liver weights, total lipids, and cholesterol contents were reduced. Hypobetalipoproteinemic activity was enhanced by prolonged treatments as demonstrated by analyses of serum obtained weekly throughout 7 wk.

SERUM 25-HYDROXYCALCIFEROL IN MYOCARDIAL INFARCTION. H. Schmidt-Gayk, J. Goossen, F. Lendle and D. Seidel (Lab. of Clin. Chem., Med. Univ. Hosp., D-6900 Heidelberg, W. Germany) *Atherosclerosis* 26, 55–8 (1977). Oral intake of calciferol (vitamin D) was higher in patients with myocardial infarction than in controls. Having established an assay for 25-hydroxycalciferol (25-OH-D), the principal circulating form of the vitamin, we measured this compound in control subjects and patients with myocardial infarction. In controls, 25-OH-D varied with the season: levels were high in summer and low in winter. Furthermore, levels were low in control subjects above 60 years of age. In patients with myocardial infarction, normal and low values for 25-OH-D were found. It is concluded that in this region patients with myocardial infarction do not consume greater amounts of vitamin D.

STUDIES ON THE METABOLISM OF β-CAROTENE AND APO-β-CAROTENOIDS IN RATS AND CHICKENS. R.V. Sharma, S.N. Mathur, A.A. Dmitrovskii, R.C. Das and J. Ganguly (Dept. of Biochem., Indian Inst. of Sci., Bangalore-560 012, India) *Biochim. Biophys. Acta* 486, 183–94 (1977). The relative abilities of the various cell fractions of rat and chicken liver to oxidize and reduce retinal and 8′- and 12′-apo-β-carotenal were investigated and it has been shown that, while retinal is exclusively oxidized by the soluble fraction, the apocarotenals are mostly oxidized by the particulate fractions of the homogenate. In the light of these observations it is suggested that during conversion to vitamin A, the β-carotene molecule is simultaneously attacked by the dioxygenase at several double bonds, the primary attack being at the central double bond and a tentative scheme for the mechanism of conversion is proposed.

METFORMIN: AN ANTIATHEROSCLEROTIC AGENT MODIFYING VERY LOW DENSITY LIPOPROTEINS IN RABBITS. C.R. Sirtori, A. Catapano, G.C. Ghiselli, A.L. Innocenti and J. Rodriguez (Ctr. E. Grossi Paoletti for the Study of Metab. Diseases and Hyperlipidemias, Univ. of Milan, 20129 Milan, Italy) *Atherosclerosis* 26, 79–89 (1977). The composition of very low density lipoproteins (VLDL: $d < 1.019$) of New Zealand male rabbits receiving cholesterol (2 g/day) and metformin (135 mg/kg/day) is investigated. These rabbits, while showing only a slight reduction of plasma cholesterol levels, as compared to cholesterol-fed (h.c.) animals, show a marked decrease of the aortic cholesterol esters and atheromatous process. VLDL from the cholesterol + metformin group (h.c. + met), as compared to the h.c. animals, are homogenous in size and not separable into VLDL-1 and VLDL-2 subfractions by Sepharose 4B chromatography. These findings are confirmed by electron microscopy, which shows homogeneity of particle size, as well as a decreased tendency of h.c. + met VLDL to aggregate. Apoprotein pattern of h.c. + met VLDL in polyacrylamide gels shows a relative increase of peptides with C *mobility* and a decrease of proteins corresponding to the arg-rich peptides. These findings exemplify a case of altered lipoprotein composition and decreased atheromatosis, in the presence of marked hypercholesteremia.

HYPOLIPIDEMIC ACTIVITY OF (−)-HYDROXYCITRATE. A.C. Sullivan, J. Triscari, and J.G. Hamilton (Dept. of Biochem. Nutr., Roche Res. Cen., Hoffmann-LaRoche Inc., Nutley, N.J. 07110) *Lipids* 12, 1–9 (1977). The influence of (−)-hydroxycitrate, a potent competitive inhibitor of adenosine triphosphate (ATP) citrate lyase, on serum triglyceride and cholesterol levels, and in vitro and in vivo rates of hepatic fatty acid and cholesterol synthesis was investigated in normal and hyperlipidemic rat model systems. (−)-Hydroxycitrate reduced equivalently the biosynthesis of triglycerides, phospholipids, cholesterol, diglycerides, cholesteryl esters, and free fatty acids in isolated liver cells. In vivo hepatic rates of fatty acid and cholesterol synthesis determined in meal-fed normolipidemic rats were suppressed significantly by the oral administration of (−)-hydroxycitrate for 6 hr, when control animals exhibited maximal rates of lipid synthesis; serum triglyceride and cholesterol levels were significantly reduced by (−)-hydroxycitrate. In two hypertriglyceridemic models-the genetically obese Zucker rat and the fructose-treated rat-elevated triglyceride levels were due, in part, to enhanced hepatic rates of fatty acid synthesis. (−)-Hydroxycitrate significantly reduced the hypertriglyceridemia and hyperlipogenesis in both models. The marked hypertriglyceridemia exhibited by the triton-treated rat was only minimally due to increased hepatic lipogenesis; (−)-hydroxycitrate significantly inhibited both serum triglyceride levels and lipogenesis in this model.

RATE OF METABOLISM OF RETINOL IN RETINOIC ACID-MAINTAINED RATS AFTER A SINGLE DOSE OF RADIOACTIVE RETINOL. P.R. Sundaresan (Lipids Lab., Res. Inst., St. Joseph Hosp., Lancaster, Penn. 17604) *J. Nutr.* 107, 70–8 (1977). The half-life and metabolism of vitamin A were determined in a group of vitamin A-deficient retinoic acid supplemented rats after a single dose of 340 μg of [6,7-$^{14}C_2$]-retinol. The total daily urinary radioactivity, plotted semilogarithmically as a function of days after injection, revealed three pools for retinol and/or metabolites in the rat:a rapidly declining pool with a half-life of 0.75 day; a slowly declining pool with a constant rate of decrease and a pool with a half-life of 13 days which begins at approximately 6 weeks after dose. The total daily fecal radioactivity also indicated three pools with half-lives of 2, 28.5 and 11.5 days. The effect of retinoic acid feeding was observed on the fifth day after supplementation, as indicated by a decrease in the total daily urinary radioactivity. Thus, retionic acid is probably in the metabolic pathway of retinol. The half-life and metabolism time of liver vitamin A in these rats were determined as 7 and 10 days, respectively. The specific activities of liver retinyl esters and retinol determined at different intervals after dose indicated continuous mixing of radioactive retinol with a pool of endogenous retinol. Blood retinol levels indicated normal values at 1 week after dose. However, they decreased at 2 weeks after dose and remained constant until the sixth week. The specific activity of blood retinol did not change indicating rapid equilibration after initial mixing and no further dilution from endogenous source.

METABOLISM OF CHENODEOXYCHOLIC ACID IN HAMSTERS. T. Tateyama and K. Katayama (Dept. of Drug Metabolism, Sec. of Ex. Therapeutics Res., Res. and Develop. Div., Eisai Co., Ltd., Koishikawa, Bunkyo-ku, Tokyo 112, Japan) *Lipids* 11, 845–7 (1976). The study on the metabolism after oral administration of chenodeoxycholic acid-24-^{14}C was performed by analysis of radioactivity that had appeared in bile and feces of male hamsters. The radioactive bile acids were analyzed by thin layer chromatography and identified by the isotope dilution method. In the bile of the hamsters with bile fistula, radioactivity was originated from unchanged chenodeoxycholic acid for the most part, and 7-ketolithocholic acid, lithocholic acid, and β-muricholic acid for the remainder. In the feces lithocholic acid, dehydrolithocholic acid, isolithocholic acid, and unchanged form were identified. After the multiple dosing of chenodeoxycholic acid-24-^{14}C for 6 days, β-muricholic acid was also identified in the feces.

EFFECT OF PHOSPHOLIPIDS ON CHOLESTEROL-INDUCED MODIFICATIONS IN MOUSE BRAIN. G. Toffano, A. Leon, D. Benvegnu and F. Cerrito (FIDIA Res. Lab., Abano Terme, Italy) *Atherosclerosis* 26, 59–66 (1977). Mice on an atherogenic diet for 40 days show a decrease in brain content of catecholamines, cyclic AMP and in dopamine degradation, and modification of the glycolytic pathway. The metabolic changes are paralleled by changes in behaviour, i.e. decrease in spontaneous motor activity and in conditioning avoidance response. The decrease in dopamine degradation and in behaviour parameters is partly due to the propylthiouracil present in the diet. Endovenous treatment with sonicated dispersions of bovine brain phospholipids induces a modification in the parameters of behaviour and metabolism. The possibility is discussed that some of the defects arising during the atherogenic diet are related with the establishment of a hypoxic state.

DIETARY INFLUENCES ON GASTRIC EMPTYING OF CARBOHYDRATE VERSUS FAT IN THE RAT. D.L. Trout, E.S. Conway and J.D. Putney (Nutr. Inst., Agr. Res. Service, U.S. Dept. of Agr., Beltsville, Md. 20705) *J. Nutr.* 107, 104–11 (1977). The degree to which the rat stomach empties carbohydrate in preference to fat was studied in rats fed a diet or various test meals providing carbohydrate and fat in a 3:1 (w/w) ratio. When rats were ad libitum fed a glucose-containing diet, the glucose:fat ratio in gastric contents was consistently lower than in the diet and was 10% as great at noon as at midnight. When starved rats were fed a single meal of the same diet, the average fractional emptying rate for carbohydrate exceeded that for fat; and the ratio of these rates ("the gastric emptying ratio") was essentially the same when calculated from gastric contents observed 1,2,4, or 6 hours after the test meal. The gastric emptying ratio was also not changed when test meals were made with hard, soft or liquid fat or with no or extra protein (lactalbumin). Use of finely divided glucose monohydrate, dried crystalline glucose or of cornstarch resulted, respectively in high intermediate and low gastric emptying ratios. The kind of form of carbohydrate in the meals and the ease of its extraction with water appear to be important factors governing the degree to which carbohydrates is preferentially emptied from the stomach.

HYPERTRIGLYCERIDEMIA IN THE DIABETIC RAT. DEFECTIVE REMOVAL OF SERUM VERY LOW DENSITY LIPOPROTEINS. A. Van Tol (Dept. of Biochem. I, Erasmus Univ. Rotterdam, Rotterdam, Netherlands) *Atherosclerosis* 26, 117–28 (1977). Streptozotocin-induced diabetic rats show a marked fasting hypertriglyceridemia. It appears that only the very low density lipoprotein (VLDL) fraction is increased. VLDL from either normal or diabetic rats was labelled in vivo in the triglyceride moiety with [^{3}H]palmitate and isolated. Both preparations, if injected intravenously into recipient rats, are removed more slowly from the circulation of diabetic rats as compared to normal rats, resulting in a reduction of the fractional catabolic rate (F.C.R.) by 70%. However, the absolute catabolic rate (turnover) of VLDL triglycerides was not changed in diabetes. It is concluded that the hypertriglyceridemia of the diabetic rat is caused by a defective removal mechanism of VLDL triglycerides. The F.C.R. of ^{125}I-labelled low density lipoproteins and high density lipoproteins is only 1–3% of the value for VLDL triglycerides and unchanged in diabetic rats.

NEURAMINIC ACID-SPECIFIC MODIFICATION AND TRITIUM LABELLING OF GANGLIOSIDES. R.W. Veh, A.P. Corfield, M. Sander and R. Schauer (Inst. for Physiol. Chem., Arbeitsgruppe fur Zellchem., Ruhr-Univ. Bochum, Postfach 102148, D-4630, Bochum 1, G.F.R.) *Biochim. Biophys. Acta* 486, 145–60 (1977). A crude ganglioside mixture and pure G_{M1} and G_{D1a} from bovine brain grey matter were prepared on a large scale. The C_7- and C_8-analogues of NeuNAc were prepared from *Collocalia* mucoid and their structures established by gas-liquid chromatography and mass spectrometry. Using model compounds in addition to various gangliosides, the conditions for the periodate oxidation and subsequent borohydride reduction of gangliosides were investigated with regard to the yield of C_7- and C_8-analogues of NeuNAc and the integrity of other monosaccharides in the oligosaccharide chain. These conditions were optimised to yield maximum C_8-NeuNAc production and low C_7-NeuNAc formation. Thus products were obtained which closely resemble the native gangliosides. Using boro [^{3}H]hydride, ganglioside derivatives with high specific radioactivity were prepared for the first time, containing either NeuNAc and labelled C_8-NeuNAc or mainly labelled C_7-NeuNAc depending on the prevailing conditions.

LECITHIN:CHOLESTEROL ACYL TRANSFER RATE IN PLASMA AND ITS RELATION TO LIPID AND LIPOPROTEIN CONCENTRATIONS IN PRIMARY HYPERLIPIDEMIA. L. Wallentin (Dept. of Internal Med. and Clin. Res. Ctr., Linkoping Univ., Med. School, S-581 85 Linkoping, Sweden) *Atherosclerosis* 26, 233–48 (1977). Plasma lecithin:cholesterol acyl transfer (LCAT) rate and concentrations of lipids in plasma and lipoproteins were studied in 107 hyperlipidemic subjects. In all types of hyperlipoproteinemia LCAT rates were higher than in a normolipidemic reference group. LCAT rates were highest in type IV and V. There was a considerable overlap of LCAT rates between type IIa, IIb and reference subjects. The LCAT rate correlated positively with very low density lipoprotein concentration, body mass and excess body mass. Low density lipoprotein concentration correlated positively with the LCAT rate only in the reference group The high density lipoprotein concentration correlated negatively with the LCAT rate. Analysing the relations between the LCAT rate and the concentrations of lipids in plasma by multiple regression indicated hypothetically deficiencies of lipoprotein removal from plasma in half of type IIa and one third of type IIb subjects.

FATTY ACID SYNTHESIS IN TESTES OF FAT-DEFICIENT AND FAT-SUPPLEMENTED RATS. A.R. Whorton and J.G. Coniglio (Dept. of Biochem., Vanderbilt Univ., Nashville, Tenn. 37232) *J. Nutr.* 107, 79–86 (1977). Fatty acid synthesis was studied in testes of rats fed a fat-free or fat-supplemented diet. Testes of fat-deficient rats incorporated nearly twice as much intratesticularly injected [1-^{14}C]acetate into total fatty acids (primarily into palmitic acid) as did supplemented rats. To determine the mechanism for the increased synthesis, the activities of the following enzymes were determined in the cytoplasmic fraction of testicular homogenates: fatty acid synthetase, acetyl CoA carboxylase [EC 6.4.1.2], citrate-cleavage [EC 4.1.3.8], malic [EC 1.1.1.38], and the glucose-6-phosphate dehydrogenase [EC 1.1.1.49]: 6-phosphogluconate dehydrogenase pair [EC 1.1.1.44]. Although the activity of fatty acid synthetase did increase in livers from rat-deficient

rats, no change was observed in corresponding testes. No difference between the two groups could be demonstrated in testicular activity of citrate-cleavage enzyme, malic enzyme, or the glucose-6-phosphate dehydrogenase:6-phosphogluconate dehydrogenase pair.

FATTY ACID SYNTHESIS IN AORTA. ISOLATION OF FATTY ACID SYNTHETASE FROM CHICKEN AORTA. A.C. Wilson, M. Murtadha and S.J. Wakil (Marrs McLean Dept. of Biochem., Baylor Col. of Med., Houston, Texas 77030) *Atherosclerosis* 26, 103–15 (1977). Fatty acid synthesis by subcellular fractions of human aorta was studied by measuring the incorporation of either radioactive acetyl-CoA or malonyl-CoA into long chain fatty acids. The high speed supernatant fraction contained fatty acid synthetase and was capable of *de novo* fatty acid synthesis. The fatty acid synthetase from chicken aorta was purified 800-fold from the high speed supernatant and was judged to be 10% pure at this level. Its molecular weight was estimated to be 450,000 on the basis of agarose gel filtration chromatography, while under dissociating conditions a molecular weight of 220,000 was obtained on sodium dodecyl sulphate disc gel electrophoresis. Fatty acid synthesis was dependent on acetyl-CoA, malonyl-CoA and NADPH. The major product was free palmitic acid. In enzymatic and physical characteristics the chicken aorta fatty acid synthetase strongly resembles the synthetase isolated from chicken liver. The two enzymes cross-react immunochemically and this homology provides the possibility of studying the synthesis and degradation of the aorta synthetase during the development of atherosclerosis.

QUANTIFICATION OF INFARCTION IN CROSS SECTIONS OF CANINE MYOCARDIUM IN VIVO WITH POSITRON EMISSION TRANSAXIAL TOMOGRAPHY AND ^{11}C-PALMITATE. E.S. Weiss, S.A. Ahmed, M.J. Welch, J.R. Williamson, M.M. Ter-Pogossian and B.E. Sobel (Washington Univ. School of Med., St. Louis, Mo.) *Cir. Res.* 55, 66–73 (1977). To assess myocardial infarction quantitatively in 15 mm thick transverse sections of the canine heart *in vivo* we utilized a new technique, positron emission transaxial tomography (PETT) and cyclotron-produced ^{11}C-palmitate (^{11}C-P) injected intravenously. Results were compared to regional myocardial creatine phosphokinase (CPK) depletion, diminished ^{11}C-palmitate accumulation in tissue extracts, and infarction estimated morphometrically 48 hours after coronary occlusion. CPK activity and ^{11}C-P content declined in parallel in transmural biopsies (N = 44) from normal and ischemic zones (r = .92) in six dogs; and infarct in 10 mm thick cross sections of the entire left ventricle estimated morphometrically (N = 26) in six other animals correlated with CPK depletion in contiguous 2.5 mm thick slices (r = .92). When the percentage of infarction in 15 mm thick cross sections was assessed tomographically in six other dogs 48 hours after coronary occlusion with ^{11}C-P injected intravenously, results correlated with infarction in corresponding cross sections from the same hearts estimated morphometrically (r = .97, N = 9) and by analysis of CPK depletion (r = .93, N = 9). Thus, PETT permits estimation of infarction in cross sections of the left ventricle *in vivo* after intravenous injection of ^{11}C-palmitate.

A COMPARATIVE ANALYSIS OF THE BINDING OF DIFFERENT LONG CHAIN FREE FATTY ACIDS BY HUMAN SERUM ALBUMIN. W.D. Wosilait and C. Soler-Argilaga (Dept. of Pharmacol., School of Med., Univ. of Missouri, Columbia, Mo. 65201) *FEBS Letters* 73, 72–6 (1977). The computed level of unbound FFA increased rapidly in a non-linear fashion, with quantitative differences due to the structures, as the concentration of FFA and the molar ratio of FFA to albumin increased. In all cases the unbound form amounted to only a small fraction of the total (less than 0.03%), which is in accord with experimental studies.

REGULATION OF PHOSPHOLIPID METABOLISM IN DIFFERENTIATING CELLS FROM RAT BRAIN CEREBRAL HEMISPHERES IN CULTURE. SERINE INCORPORATION INTO SERINE PHOSPHOGLYCERIDES: BASE EXCHANGE AND DECARBOXYLATION PATTERNS. E. Yavin and B.P. Zeigler (Dept. of Neurobiol. and Dept. of Applied Math., The Weizmann Inst. of Sci., Rehovot, Israel) *J. Biol. Chem.* 252, 260–7 (1977). The patterns of serine metabolism into phospholipids of cultured brain cells was examined. Labeled serine was incorporated predominantly into serine- and ethanolamine-containing phospholipids and sphingolipids. The highest rates of labeling were observed in the (1)acyl-(2)acyl- and (1)alkyl-(2)-acyl-serine phosphoglyceride fractions. Serine incorporation into both compounds appears to proceed via a base exchange mechanism. A decrease in the rate of serine phosphoglycerides labeling and a depletion of the ATP levels were observed when oligomycin or the calcium ionophore A23187 was added to the incubation medium. The inhibition of serine incorporation by A23187 could be partially reversed following addition of 10 mM $CaCl_2$. Based on these findings it is suggested that in addition to demonstrating the energy-independent calcium-stimulated pathway, there may also be an energy related pathway.

HIGH PLASMA CHOLESTEROL IN MINK (MUSTELA VISON) WITHOUT ATHEROSCLEROSIS. D.B. Zilversmit, T.B. Clarkson and L.B. Hughes (Div. of Nutr. Sci.; and Section of Biochem., Molecular and Cell Biol., Div. of Biol. Sci., Cornell Univ., Ithaca, N.Y. 14853) *Atherosclerosis* 26, 97–102 (1977). Mink fed a commercial ration moderately high in cholesterol or fed a cholesterol-free semipurified diet have plasma cholesterol similar to that found in human beings living in industrialized countries. In contrast with human beings, 80% of the plasma cholesterol in mink is carried in the high density lipoprotein fraction. Aortas and coronary arteries from animals up to 8 yr. old were found to be free of fatty streaks and atherosclerotic plaques, both grossly and microscopically.

METHOD FOR ADSORPTION AND ELUTION OF LIPID HYDROLYZING ENZYMES. Y. Kosugi, H. Suzuki, and A. Kamibayashi (Agency of Industrial Science and Technology, Tokyo). *U.S. 4,013,512.* A method for separating a lipid hydrolyzing enzyme whose molecules have outwardly exposed hydrophobic residues from a solution containing the enzyme comprises bringing the crude enzyme solution into contact with an insoluble carrier having an attached hydrophobic organic compound thereby causing the enzyme to be adsorbed on the carrier. Then the adsorbed enzyme is contacted with a liquid containing a surface active agent, a liquid containing a protein denaturing agent and an organic solvent miscible with water for thereby eluting the enzyme into the liquid.

STUDIES ON THE RELATIONSHIP BETWEEN THE NUTRITIVE VALUE AND THE STRUCTURE OF POLYMERIZED OILS. XI. MECHANISM OF TOXICITY OF HEAT-POLYMETIZED OILS. M. Saito and T. Kaneda (Dept. of Food Chemistry, Tohoku University, Sendai), *Yukagaku* 25(12), 28–34 (1976). To study toxic effects of cyclic fatty acids, rats were given a diet containing 2.2–4.9% cyclic fatty acids for three weeks and the inhibition of body weight gain, hypertrophy and/or hyperplasy of liver, and a little decrease of digestibility were observed. Analyses of liver lipids revealed the symptom of fatty liver accompanied by increase of neutral lipids and phospholipids. Histopathological changes were also observed in nuclei, cytoplasms and intercellular substances of liver cells by histological examination. To investigate the absorption of cyclic fatty acids which caused pathological changes, component fatty acids of plasma and liver lipids were determined by gas chromatography-mass spectrometry (GC-MS). The presence of these cyclic fatty acids was observed in both plasma and liver. This result indicated the possibility of absorption of these cyclic fatty acids through small intestine. Furthermore, cyclic fatty acids were also found in neutral lipids, phospholipids and cholesteryl esters fractions of liver lipids. To know the relationship between the skeletal structure of cyclic fatty acids and toxicity, cyclic fatty acids were reduced and administered orally to mice. The hydrogenated cyclic fatty acids showed less toxicity than the original acids. From these results, they concluded that cyclic fatty acids are absorbed through small intestine and cause fatty liver and the other histophathological changes. Thus one of the reasons for the toxic effect of cyclic fatty acids is attributable to the metabolic injury in liver.

THE ERYTHROCYTE MEMBRANE SITE FOR THE EFFECT OF TEMPERATURE ON OSMOTIC FRAGILITY. B. Aloni, A. Eitan and A. Livne (Dept. of Biol., Ben Gurion Univ. of the Negev, Beer Sheva, Israel) *Biochim. Biophys. Acta* 465, 46–53 (1977). The osmotic fragility of human erythrocytes is well known to decrease as the temperature is elevated. The cellular site for the temperature effect was studied by assessing possible roles of hemoglobin and of membrane lipids and by taking advantage of the unique response of camel erythrocytes to temperature. It is concluded that the erythrocyte membrane is the site for the temperature effect on osmotic fragility. The human erythrocyte is likely to rupture in protein-lipid boundary regions in the membrane, from which cholesterol is apparently excluded.

LABELLING OF EGG PHOSPHATIDYLCHOLINE VESICLES AND MYELIN MEMBRANE WITH A PHOTOREACTIVE LIPOPHILIC REAGENT. K.M. Abu-Salah and J.B.C. Findlay (Dept. of Biochem., Univ. of Leeds, 9 Hyde Terrace, Leeds LS29LS, U.K.) *Biochem. J.* 161,

223–8 (1977). The preparation and isolation of [^{3}H]phenyl azide, a photosensitive non-polar probe, is reported. The reagent partitions into the lipid bilayer of egg phosphatidylcholine vesicles and bovine myelin membranes. On photoactivation to generate the nitrene grouping, as much as 90% of the covalently attached label is associated with the fatty acyl residues of the constituent phospholipid molecules. The remainder is found in the polar head groups. The cholesterol component of myelin membranes is also heavily labelled. These results suggest that such reagents may be used to probe the hydrophobic regions of natural membranes.

THE ACTION OF A CARBONSUBOXIDE DIMERIZED GRAMICIDIN A ON LIPID BILAYER MEMBRANES. E. Bamberg and K. Janko (Dept. of Biol., Univ. of Konstanz, D-7750 Konstanz, G.F.R.) *Biochim. Biophys. Acta* **465**, 486–99 (1977). Gramicidin A was dimerized with carbon-suboxide as bifunctional reagent. The effect of the resulting malonyl-bis-desformyl-gramicidin on lipid bilayer membranes was investigated and compared with the effect of the monomer gramicidin. It was found that the single channel conductance and the ion selectivity are very similar to the behaviour of the monomer molecule, whereas the channel forming kinetics and the life time of the single channel of the malonyl-bis-desformylgramicidin differ strongly from the behaviour of the monomer gramicidin. The electrical relaxations are very small and possibly associated with some structural changes of the membrane after a voltage jump. The single channel lifetime of the malonyl-bis-desformylgramicidin is measured in minutes, whereas for the same lipid system the single channel lifetime in the case of the monomer gramicidin is restricted to 1–2 s. It is concluded that the malonyl-bis-desformylgramicidin-molecule itself (as a single molecule) forms an ionic channel without further association.

THE EFFECT OF CHOLESTEROL INCORPORATION ON THE TEMPERATURE DEPENDENCE OF WATER PERMEATION THROUGH LIPOSOMAL MEMBRANES PREPARED FROM PHOSPHATIDYLCHOLINES. M.C. Blok, L.L.M. Van Deenen and J. De Gier (Lab. of Biochem., State Univ. of Utrecht, Transitorium 3, Padualaan 8, Utrecht, The Netherlands) *Biochim. Biophys. Acta.* **464**, 509–18 (1977). The permeation of water through liposomal membranes composed of phosphatidylcholine plus varying amounts of cholesterol was studied as a function of temperature. Increasing amounts of cholesterol caused a gradual disappearance of the abrupt change in the rate of water permeation near the gel to liquid-crystalline phase transition temperature of dipalmitoylphosphatidylcholine and dimyristoylphosphatidylcholine liposomes. At cholesterol concentrations above about 30 mol % there was no longer a discontinuity in the rate of water permeation. In systems containing unsaturated phosphatidylcholines cholesterol also enhanced the activation energy of the water permeation although to a lesser extent. The results indicate that the position of the *cis*-double bond in the fatty acid chain is very important in this respect.

THE PREFERENTIAL INTERACTION OF CHOLESTEROL WITH DIFFERENT CLASSES OF PHOSPHOLIPIDS. R.A. Demel, J.W.C.M. Jansen, P.W.M. Van Dijck and L.L.M. Van Deenen (Lab. of Biochem., State Univ. of Utrecht, Univ. Centre "De Uithof", Padualaan 8, Transitorium 3, Utrecht, The Netherlands) *Biochim. Biophys. Acta* **465**, 1–10 (1977). By differential scanning calorimetry a preferential affinity of cholesterol for sphingomyelin was established in mixtures of sphingomyelin and phosphatidylcholine where sphingomyelin was either the higher or the lower melting phospholipid. A preferential affinity of cholesterol for sphingomyelin was also found in mixtures of sphingomyelin and phosphatidylethanolamine where sphingomyelin was either the higher or the lower melting phospholipid. The sphingomyelin used was isolated from beef erythrocytes or synthetic palmitoyl sphingomyelin. In mixtures of phosphatidylserine with phosphatidylethanolamine, or phosphatidylserine with phosphatidylcholine, cholesterol showed the highest affinity for the lower melting phospholipid.

ANALOGS OF NATURAL LIPIDS. III. NONEQUIVALENCE OF METHYL GROUPS IN METHYLATED PHOSPHOLIPIDS. A.A. Gallo, A.J. Hancock and H.Z. Sable (Dept. of Biochem., Case Western Reserve Univ. School of Med., Cleveland, Ohio 44106) *J. Lipid Res.* **18**, 77–80 (1977). The dimethyl esters of a series of diastereoisomeric cyclopentanoid analogs of phosphatidic acid have been studied by proton NMR spectroscopy at 60, 100, and 300 MHz. The signals of the P–O–CH_3 protons near δ 3.80 show the expected doubling due to the ^{31}P-^{1}H coupling. In addition, the spectra of three of the isomers show additional multiplicity, the line separation (in Hz) being proportional to the frequency of the spectrometer. This multiplicity is due to the nonequivalence of the two methoxyl groups on phosphorus, predictable from their diastereotopic nature. The same explanation is proposed for similar observations on other compounds made by other authors The practical utility of symmetry considerations in lipid chemistry is discussed briefly.

PROTON MAGNETIC RESONANCE STUDY OF CHOLESTEROL TRANSFER BETWEEN EGG YOLK LECITHIN VESICLES. N. Haran and M. Shporer (The Weizmann Inst. of Sci., Rehovot, Israel) *Biochim. Biophys. Acta* **465**, 11–8 (1977). The intensity of the proton magnetic resonance signal of the $(CH_2)_n$ chain in phospholipids of sonicated lecithins is sensitive to the cholesterol content in the resulting vesicles. In the present study this signal has been used to monitor transfer of cholesterol between phospholipid vesicles. Vesicles prepared from pure egg yolk lecithin were mixed with vesicles that contained equimolar amounts of cholesterol and lecithin, and the time evolution of the $(CH_2)_n$ signal intensity was followed. The results show that a homogenous distribution of cholesterol among vesicles is reached after about 4 h at 37°C and 60 h at 4°C. In order to determine the mechanism of the cholesterol transfer process, experiments were performed over a 2.5-fold range of vesicles concentrations. The accuracy of the kinetic results was not sufficient however to decide on the order of the reaction with respect to vesicle concentration. Simultaneous observation of the choline proton resonance in the presence of Eu^{+3} and Pr^{+3} indicates that fusion between vesicles does not occur during cholesterol transfer.

ANALYSIS OF THE DEFECT STRUCTURE OF GEL-PHASE LIPID. A.G. Lee (Dept. of Physiol. and Biochem., Univ. of Southampton, Southampton SO9 3TU, United Kingdom) *Biochemistry* **16**, 835–41 (1977). The partitioning of the spin label 2,2,6,6-tetramethylpiperidinyl-1-oxy (Tempo) into phosphatidylcholine bilayers and the monomer-aggregate equilibrium for chlorophyll a incorporated into phosphatidylcholine bilayers have been interpreted in terms of the formation of defects in the gel-phase lipid, starting some 20°C below the temperature of the main gel to liquid crystalline phase transition. By contrast, defects seem to be largely absent from bilayers of dipalmitoylphosphatidylethanolamine in the gel phase. The defect structure accounts for the continuous nature of the phase transition for phosphatidylcholines, and also for the increase in width of the transition caused by the addition of alcohols.

A COMPARISON OF SIMPLIFIED METHODS FOR LIPOPROTEIN QUANTIFICATION USING THE ANALYTIC ULTRACENTRIFUGE AS A STANDARD. F.T. Lindgren, A. Silvers, R. Jutagir, L. Layshot, and D.D. Bradley (Donner Lab., Lawrence Berkeley Lab., Univ. of California, Berkeley, Calif.) *Lipids* **12**, 278–82 (1977). Two simplified methods for quantitative lipoprotein analysis have been calibrated and compared with each other using analytic ultracentrifugation as a standard reference procedure. The first method was the Friedewald procedure and the second was an automated agarose gel electrophoresis system. Both procedures offer comparable quantitative lipoprotein analysis with potential for large scale screening purposes at low cost ($4.00–$5.00 per analysis). There were advantages and limitations to both procedures. The Friedwald procedure can be used on frozen sera but requires 3 ml sera. In contrast, the electrophoresis system must be used with fresh serum but requires only 50 μl serum and the electrophoretic slides may be quantitatively analyzed several years retrospectively.

COMPOSITION OF NOVEL TRIESTERS FROM THE SKIN OF THE RHINO MUTANT MOUSE. M.K. Logani, D.B. Nhari, and R.E. Davies (Skin and Cancer Hosp. of Philadelphia, Temple Univ. Health Sci. Ctr., Philadelphia, Penn. 19140) *Lipids* **12**, 283–7 (1977). Composition of two novel triesters, derived from the skin of the rhino mutant mouse, is described. Chemical and spectroscopic analysis of the products of pancreatic hydrolysis of the triesters showed that these are comprised predominantly of isomer I (92.7 mole %). The syntheses of two reference compounds, 1-0-hexadecanoyl-2-[(14-hexadecanoyloxy)0-tetradecanoyl] 1,2-hexadecanediol (Ia) and 2-0-hexadecanoyl-1-[(14-hexadecanoyloxy)0-tetradecanoyl]-1,2-hexadecanediol (IIa), corresponding in their structures to isomers I and II of the triester, wax have also been described.

SURFACE PHASE SEPARATION AND COLLAPSE OF THE STEARATE ANION-ALKALINE EARTH CATION COMPLEX. G.S. Patil and D.G. Cornwell (Dept. of Physiol. Chem., Ohio State Univ., Columbus, Ohio 43210) *J. Lipid Res.* **18**, 1–5 (1977). The surface properties of fatty acid and fatty acid-alcohol mixtures were examined at 22–24°C. At pH 12, sodium stearate forms a rigid

surface film that generates an equilibrium spreading pressure of 16.5 dynes/cm. At pH 12, stearate-alkaline earth cation films collapse at the air-water interface and do not generate significant equilibrium spreading pressures. The rate of film collapse depends on the counterion decreasing in the sequence $Ba^{2+} > Sr^{2+} > Ca^{2+}$. Stearate-stearyl alcohol mixtures form solid (condensed) films that are relatively stable and behave initially as homogeneous surfaces in their selectivities for counterions. Stearate-oleyl alcohol mixtures form fluid (expanded) films that are unstable. Lateral phase separations occur rapidly in fluid films and the stearate-alkaline earth cation phase collapses. The rate of film collapse in the fluid mixtures also depends on the counterion decreasing in the sequence $Ba^{2+} > Ca^{2+}$. These surface properties suggest how a lipid anion may function as an ionophore in the translocation of alkaline earth cations.

CLUSTERING OF NITROXIDE SPIN LABELS IN LIPID BILAYER MEMBRANES. P. Rey and H.M. McConnell (Dept. of Chem., Stanford Univ., Stanford, Calif. 94305) *J. Am. Chem. Soc.* **99**, 1637–42 (1977). Three new nitroxide (spin-label) biradicals have been prepared; N,N′-dipalmitoyl-N,N′-bis(1-oxly-2,2,6,6-tetramethylpiperindin-4-yl)-1,10-diaminodecane; N,N′-dimethyl-N,N′-dihexadecyl-N,N′-bis(1-oxly-2,2,6,6-tetramethylpiperidin-4-yl)-1,10-diammoniumdecane diiodide; N,N′-dipalmitoyl-N,N′-bis [N-(1-oxyl-2,2,6,6-tetramethylpiperidin-4-yl)acetamide-2-yl]-1,10-diammoniumdecane. It is shown that these biradicals bind strongly to phosphatidylcholine-cholesterol bilayer membranes and exhibit a degree of clustering in the plane of the membrane that depends on the particular biradical, the concentration of the biradical in the plane of the membrane, the lipid composition of the membrane, and the temperature. The results illustrate one approach to controlling the lateral distribution and motion of membrane components that is relevant to current studies of membrane immunochemistry.

TRIACONTANOL: A NEW NATURALLY OCCURRING PLANT GROWTH REGULATOR. S.K. Ries, Violet Wert, C.C. Sweeley and R.A. Leavitt (Dept. of Hort., Michigan St. Univ., East Lansing, Mich. 48824) *Science* **195**, 1339–41 (1977). Alfalfa meal and chloroform extracts of the meal have increased the growth and yield of several plant species. A crystalline substance isolated from the active fraction of alfalfa meal increased the dry weight and water uptake of rice seedlings when sprayed on the foliage or applied in nutrient culture. The substance was identified as triacontanol by mass spectrometry. Sprays containing this compound also increased the growth of corn, and barley grown in soil. Authentic triacontanol produced a similar response over a wide range of concentrations on rice grown in nutrient cultures and tomatoes grown in soil.

CALCIUM ION-FLUX ACROSS PHOSPHATIDYLCHOLINE MEMBRANES MEDIATED BY IONOPHORE A23187. J. Wulf and W.G. Pohl (Univ. Konstanz, Fachbereich Biol., D-7750 Konstanz, G.F.R.) *Biochim. Biophys. Acta* **465**, 471–85 (1977). The antibotic A23187 carries Ca^{2+} across Müller-Rudin membranes made from 1,2-dierucoyl-*sn*-glycero-3-phosphocholine and *n*-decane. The conductance of the membranes is not increased by the Ca^{2+}-transport. The flux depends linearly on Ca^{2+} concentration and ionophore concentration (above pH 6). It increases with increasing pH, approximately by a factor of 4–5 between pH 6 and pH 8. Maximal Ca^{2+}-fluxes of about 10^{-10} mol · cm^{-2} · s^{-1} were found. A counter transport of H^+ could not be detected. The complex formation between A23187 and Ca^{2+} in egg phosphatidylcholine vesicles was studied spectroscopically. The results are consistent with the formation of a 2:1 complex. Optical absorption measurements on single phosphatidylcholine membranes were used to calculate the concentration of membrane-bound ionophore A23187.

HYDROCARBON CHAIN TRANS-GAUCHE ISOMERIZATION IN PHOSPHOLIPID BILAYER GEL ASSEMBLIES. N. Yellin and I.W. Levin (Lab. of Chem. Phys., Nat'l. Inst. of Arthritis, Metab. and Digestive Diseases, Nat'l. Insts. of Health, Bethesda, Md. 20014) *Biochemistry* **16**, 642–7 (1977). The vibrational Raman spectra of dimyristoyl (DMPC)-, dipalmitoyl (DPPC)-, and distearoylphosphatidylcholine (DSPC)-water bilayer systems were used to probe lipid hydrocarbon chain trans-gauche isomerization dynamics below the gel-liquid crystalline phase transition temperature. In addition to the 1090-1085 cm^{-1} vibrational transitions, which appear with increasing temperatures and are characteristic of gauche conformers within the acyl chains, a new feature arises in all three bilayer systems at ~1122 cm^{-1}. This carbon-carbon stretching mode is associated with the formation of a gauche bond rotation of the terminal methyl group oriented toward the center of the bilayer. Estimates of the enthalpy differences (ΔH) between hydrocarbon chains in an all-trans conformation and chain configurations containing gauche forms may be made from peak height intensities of vibrational features associated with the appropriate rotational isomers. For the DMPC-H_2O, DPPC-H_2O, and DSPC-H_2O assemblies, the Raman data yield enthalpy differences of 2.9 ± 0.6, 3.4 ± 0.5, and 9.9 ± 1.2 kcal/mol, respectively. These values are interpreted to reflect approximately two gauche bonds per lipid molecule for the DMPC-H_2O and the DPPC-H_2O gels and six gauche bonds per molecule for the DSPC-H_2O gels.

INTERRELATIONSHIP OF DIETARY VITAMIN D_3 WITH ZINC AND IRON IN YOUNG TURKEYS. A. Aksoy and T.W. Sullivan (Dept. of Poultry and Wildlife Sci., Univ. of Neb., Lincoln, Neb. 68583) *Poult. Sci.* **56**, 491–8 (1977). Three experiments were conducted with large white turkeys to four weeks of age. The interrelationship of zinc and vitamin D_3 was studied in two experiments. Iron and vitamin D_3 were involved in one experiment. One group of ten male and one group of ten female poults were randomly assigned to each treatment within an experiment. All experiments involved a factorial arrangement of two dietary variables, D_3 and either zinc or iron. For example, all possible combinations of four zinc levels; 31, 46, 76, and 106 p.p.m. and three vitamin D_3 levels; 600, 1200, and 3600 I.C. units/kg. were fed in the second zinc experiment. Significant ($P < 0.05$) weight gain differences occurred among both zinc and D_3 levels in this experiment. Four week body weights were 356, 436, 459 and 444 g., respectively, for Zn levels; and 409, 410, and 452 g., respectively, for D_3 levels. Significant ($P < 0.05$) interactions occurred between dietary zinc and D_3 levels, and between iron and D_3 levels relative to body weight gain. Vitamin D_3 tended to increase tibia zinc levels in poults receiving higher zinc levels; the opposite occurred with lower levels of the element. Zinc level in feces was decreased with vitamin D_3. Hemoglobin level was not influenced by D_3 but was increased by iron.

ETHER-LINKED GLYCEROLIPIDS IN HUMAN BRAIN TUMORS. D.H. Albert and C.E. Anderson (Dept. of Biochem. and Nutr., School of Med., Univ. of North Carolina, Chapel Hill, N.C. 27514) *Lipids* **12**, 188–92 (1977). In this investigation, the lipid composition of a number of human brain tumors was determined and compared to that of normal adult brain. Glioblastomas (11 samples), astrocytomas (4 samples), an acoustic neurinoma, an oligodendroglioma, and a meningioma were analyzed. All of the tumors had substantial levels (0.8–3.4% of total phospholipids) of choline plasmalogen which was present in only trace amounts in normal brain. With the exceptions of the acoustic neurinoma and the meningioma, the concentration of alkylacylglycerophosphorylcholine was also higher in the tumors than in normal brain. Neutral lipids of brain tumors also contained high concentrations of both alkyl (1.6–4.8% of total neutral glycerolipids) and alkenyl diacyl glycerol (3.8–10.1%). The results from this investigation indicate that increases in ether-linked glycerolipids may be characteristic of human brain tumors.

RATE CONSTANTS FOR THE UPTAKE OF CHOLESTEROL FROM VARIOUS INTESTINAL AND SERUM LIPOPROTEIN FRACTIONS BY THE LIVER OF THE RAT IN VIVO. J.M. Andersen, F.O. Nervi and J.M. Dietschy (Dept. of Med., the Univ. of Texas Health Sci. Center at Dallas, Dallas, Texas 75235 U.S.A.) *Biochim. Biophys. Acta* **486**, 298–307 (1977). The increase in the mass of cholesterol esters in the liver was used to estimate hepatic net uptake rates of cholesterol from various serum and intestinal lipoprotein fractions. Initial uptake rates equalled essentially zero for high density serum lipoproteins and for large chylomicrons while administration of both low density serum lipoproteins and smaller chylomicrons produced a significant increase in hepatic cholesterol ester levels. The rate of uptake of both serum lipoprotein fractions did not change over a 5 h interval after injection: in contrast, the rates of uptake of the intestinal fractions increased 10–25-fold during this interval. These observations are consistent with the view that the liver is capable of taking up cholesterol from chylomicron remnants and, at significantly lower rates, low density serum lipoproteins.

A FACILE HYDROLYSIS-SOLVOLYSIS PROCEDURE FOR CONJUGATED BILE ACID SULFATES. G.P. van Berge-Henegouwen, R.N. Allan, A.F. Hofmann and P.Y.S. Yu (Gastroenterol. Unit., Mayo Clinic and Mayo Found., Rochester, Minn. 55901) *J. Lipid Res.* **18**, 118–22 (1977). Methods for hydrolyzing and solvolyzing conjugated bile acid sulfates were compared on

reference mixtures of conjugated and unconjugated bile acid sulfates using gas-liquid chromatography to assess recovery, and thin-layer chromatography and zonal scanning to define the products occurring after hydrolysis. Conventional methods in which solvolysis preceded vigorous alkaline saponification gave incomplete recoveries. However, essentially complete recovery of primary and secondary bile acid sulfates was obtained with a mild alkaline saponification procedure followed by acidification and extraction into ether, in which complete solvolysis was shown to occur within 12 hours. Based on these findings, we developed and validated a simple hydrolysis-solvolysis procedure; the method features mild alkaline hydrolysis, acidification to pH 1, and extraction with ether followed by a 1-hour incubation.

CARNITINE UPTAKE INTO HUMAN HEART CELLS IN CULTURE. T. Bohmer, K. Eiklid and J. Jonsen (Dept. of Med. B and Inst. of Surgical Res., Rikshospitalet, Univ. Hosp., Oslo, Norway) *Biochim. Biophys. Acta* **465**, 627–33 (1977). The uptake of radiolabeled carnitine and butyrobetaine has been studied in human heart cells (CCL 27). The uptake of carnitine is 3–10-fold higher in heart cells than in fibroblasts (pmol · μg DNA^{-1}). The uptake of carnitine increases with temperature coefficient K_T of 1.6 in the interval 10–20°C and with a negligible uptake at 4 and 10°C. The uptake of carnitine follows Michaelis-Menten kinetics with a K_M of 4.8 ± 2.2 μM and V = 8.7 ± 3.2 pmol · μg DNA^{-1} · h^{-1}. Carnitine uptake is suppressed 90% by NaF (24 mM). Butyrobetaine is taken up into heart cells to the same extent as carnitine with a K_M of 5.7–17.3 μM and V = 8.7–9.3 pmol · μg DNA^{-1} · h^{-1}. Butyrobetaine inhibits competitively the uptake of carnitine and carnitine inhibits the uptake of butyrobetaine to the same extent. No conversion of radiolabeled butyrobetaine to carnitine, or carnitine to methyl choline was observed intra- or extracellulary during incubation. These data are compatible with a selective transport mechanism for carnitine which is also responsible for the uptake of butyrobetaine.

RELATIVE ACTIVITIES OF SOME METABOLITES AND ANALOGS OF CHOLECALCIFEROL IN STIMULATION OF TIBIA ASH WEIGHT IN CHICKS OTHERWISE DEPRIVED OF VITAMIN D. A. Boris, J.F. Hurley and T. Trmal (Dept. of Cell Biol., Roche Res. Ctr., Nutley, New Jersey 07110) *J. Nutr.* **107**, 194–8 (1977). Nine metabolites and analogs of cholecalciferol (CC) were tested for ability to increase tibia ash weight in chicks otherwise deprived of vitamin D. All of the compounds promoted bone mineralization in a linear log dose-response relationship. The maximal response obtained for any compound was an approximate doubling in bone ash weight compared to vehicle-treated controls. Relative potencies, based upon the calculated ash weight doubling dose, were as follows: 1α,25-$(OH)_2$-CC = 1α-OH-CC > CC > 25-OH-CC > 24R,25-(OH_2)-CC = 1α,24R,25-$(OH)_3$-CC > 5,6-trans-25-OH-CC > 1α,24S,25-$(OH)_3$-CC > 5,6-trans-CC > 24S,25-$(OH)_2$-CC.

THE ZUCKER-FATTY RAT: A REVIEW. G.A. Bray (Dept. of Med., UCLA School of Med., Harbor General Hosp. Campus, Torrance, Calif. 90509) *Fed. Proc.* **36**, 148–53 (1977). The Zucker (fatty) rat is one of a group of animals that inherit obesity as an autosomal Mendelian recessive trait. These rats are obese, hyperphagic, and hyperinsulinemic, but blood glucose remains at normal levels. Although these rats eat more than normal rats, their response to the addition of adulterants to the food or after exposure to the cold is more like that of normal rats than rats with hypothalamic obesity. The hypertriglyceridemia which characterizes these animals is due to the increased hepatic production of very low density lipoproteins. Adipocytes are increased in size and in number with the subcutaneous fat depot showing the largest increase in the number of fat cells. Lipogenesis from glucose is brisk in the young animals but declines with age. Enzymatic patterns of glycolysis and gluconeogenesis appear to reflect the altered internal milieu rather than specific defects. Endocrine changes in the fatty rat include hyperinsulinemia, reduced levels of glucagon, hypothyroidism, and impaired reproductive function. A model is presented in which the features of the genetically obese (Zucker) fatty rat are compared with those of animals with hypothalamic obesity.

GROWTH AND ACYLTRANSFERASE ACTIVITY OF RABBIT MAMMARY GLAND DURING PREGNANCY AND LACTATION. M. Caffrey and J.E. Kinsella (College of Agr. and Life Sci., Stocking Hall, Cornell Univ., Ithaca, N.Y. 14853) *J. Lipid Res.* **18**, 44–52 (1977). A bimodal change in yield and microsomal protein content of rabbit mammary gland was observed with the progress of pregnancy and lactation. The initial stimulus took place on day 22 of pregnancy and the second during early lactation. Palmitoyl-CoA:monopalmitoyl-*sn*-glycerol 3-phosphate palmitoyltransferase activity was monitored concurrently. This enzyme in rabbit mammary microsomes is composed of two isoenzymic species that differ with respect to the physical nature of the substrates with which each interacts. The activities of the two isoenzymes were recorded at progressive stages of pregnancy, lactation, and involution to determine if a regulatory role could be assigned to either or both species. Although the patterns were indefinite, total transacylase activity did increase over this period, i.e., the specific activity of LPAT-α was 12 and 24 nmoles/mg protein per min in pregnancy and lactation, respectively, while that of LPAT-β rose from zero to 90 nmoles/mg protein per min over the same period. The time of harvesting in relation to the interval between nursing periods is discussed in the light of these results.

EFFECTS OF PROLONGED FENFLURAMINE ADMINISTRATION IN OBESE AND NONOBESE MICE. R.H. Carr, M. Ipaktchi and S.W. Thenen (Dept. of Nutr., Harvard School of Public Health, Boston, Mass. 02115) *Proc. Soc. Exp. Biol. Med.* **154**, 116–20 (1977). Oral administration of fenfluramine for 8 weeks reduced food intake and body weight in ob/ob, GTG, and nonobese mice. The hyperglycemia and hyperinsulinemia present in the ob/ob mice were significantly reduced by this drug. Plasma glucose, but not insulin, was significantly reduced in GTG mice; neither was significantly reduced in nonobese mice. Fenfluramine reduced plasma triglycerides, but had no effect on liver glycogen concentration and did not reduce the percentage of body fat.

CELL SURFACE CHANGES IN DIABETIC RATS. STUDIES OF LECTIN BINDING TO LIVER CELL PLASMA MEMBRANES. V. Chandramouli, S. Williams, J.S. Marshall and J.R. Carter, Jr. (Dept. of Med., Case Western Reserve Univ. School of Med., Cleveland, Ohio) *Biochim. Biophys. Acta* **465**, 19–33 (1977). We have previously reported changes in the chemical composition of cell surface membranes in diabetic rats. To examine the possible implications of these changes for cell surface structures, we have measured the binding of labeled lectins and desialylated glycoproteins to plasma membranes prepared from the livers of streptozotocin-diabetic and control rats. Lectins were chosen which have affinities for different carbohydrate moieties. The binding of ricin and concanavalin A to liver cell membranes from the diabetic rats was significantly reduced, but no change in the binding of wheat germ agglutinin was noted. Binding of desialylated thyroxine-binding globulin, previously shown to be dependent on membrane sialic acid residues, was strikingly reduced in liver membranes from diabetic rats. These results strongly suggest that insulin deficiency leads to generalized changes in cell surface glycoproteins, at least in this animal model of diabetes.

CHOLIC ACID BIOSYNTHESIS: CONVERSION OF 5β-CHOLESTANE-3α,7α,12α,25-TETROL INTO 5β-CHOLESTANE-3α,7α,12α,24β,25-PENTOL BY HUMAN AND RAT LIVER MICROSOMES. F.W. Cheng, S. Shefer, B. Dayal, G.S. Tint, T. Setoguchi, G. Salen and E.H. Mosbach (College of Med. and Dentistry of New Jersey, New Jersey Med. School, Newark, N.J. 07103) *J. Lipid Res.* **18**, 6–13 (1977). This paper describes the conversion of 5β-cholestane-3α,7α,12α,25-tetrol into 5β-cholestane-3α,7α,12α,24β,25-pentol by liver microsomes. A sensitive radioactive assay for measuring the formation of 5β-cholestane-3α,7α,12α,24β,25-pentol was developed. Optimal assay conditions for human and rat microsomal systems were established. A higher 24β-hydroxylation activity was detected in rat than in human liver under the conditions employed. The hydroxylation of 5β-cholestane-3α,7α,12α,25-tetrol by the rat liver microsomal fraction fortified with NADPH was stimulated about two-fold by administration of phenobarbital. Phenobarbital treatment also stimulated hydroxylations at C-23, C-24α, and C-26. Carbon monoxide markedly inhibited all side-chain hydroxylations. In contrast, side-chain hydroxylase activities were not affected in animals deprived of food for 48 hr. These results are consistent with a previously postulated cholic acid biosynthetic pathway involving 5β-cholestane-3α,7α,12α,24β,25-pentol as a key intermediate in man and in the rat.

BIOSYNTHESIS OF FATTY ACIDS IN PERFUSED HEARTS OF RATS FED A FAT-SUPPLEMENTED OR A FAT-FREE DIET. J.G. Coniglio and J.M. O'Bryan (Dept. of Biochem., School of Med., Vanderbilt Univ., Nashville, Tenn. 37232) *Proc. Soc. Exp. Biol. Med.* **154**, 131–3 (1977). Incorporation of ^{14}C from [1-^{14}C]butyrate into lipids of the perfused rat heart was slightly greater than that from [1-^{14}C]acetate. Of the ^{14}C incorporated into total

lipids, a greater proportion was in phospholipids when [^{14}C] acetate was the substrate than when [^{14}C]butyrate was used as substrate. Perfusion with [^{14}C]-butyrate resulted in more ^{14}C incorporation into shorter chain fatty acids and less incorporation into palmitic and arachidonic acids than with [^{14}C] acetate. Feeding a fat-free diet did not affect the metabolism of acetate in the perfused heart. However, it did increase the relative incorporation of ^{14}C from butyrate into phospholipids as well as the total ^{14}C incorporation into total lipids of perfused hearts of rats compared to those fed a fat-supplemented diet.

GOLD THIOGLUCOSE OBESITY SYNDROME. A.F. Debons, I. Krimsky, M.L. Maayan, K. Fani and F.A. Jimeniz (Nuclear Med. and Anat. Pathol., Veterans Admin. Hosp., Brooklyn, N.Y. 11209) *Fed. Proc.* **36**, 143–7 (1977). Parenteral administration of gold thioglucose to mice produces an area of necrosis in the ventromedial portion of the hypothalamus. The lesion, like lesions produced by electrocautery of this area, causes hyperphagia and consequent obesity. The glucose moiety of gold thioglucose is essential for production of the lesion. Glucose analogues (2-deoxyglucose, sodium thioglucose and phlorizin) prevent the gold thioglucose-induced lesion, and by themselves produce a transient hyperphagia. Insulin deficiency prevents the lesion. Either adrenalectomy or hypophysectomy counteracts the effect of insulin deficiency. Electron microscopic studies, in which general necrosis is avoided by administration of aspirin before gold thioglucose or by administration of subnecrotic doses of gold thioglucose, reveal that gold thioglucose primarily affects neural elements contiguous with capillaries in the ventromedial hypothalamus. The experimental observations indicate the presence of special glucoreceptor cells in the ventromedial hypothalamus that are involved in the regulation of food intake.

PLASMALOGENASE ACTIVITIES IN NEUTONAL PERIKARYA, ASTROGLIA, AND OLIGODENDROGLIA ISOLATED FROM BOVINE BRAIN. R.V. Dorman, A.D. Toews and L.A. Horrocks (Dept. of Physiol. Chem., The Ohio State Univ., Columbus, Ohio 43210) *J. Lipid Res.* **18**, 115–7 (1977). Plasmalogenase (EC 3.3.2–, 1-alk-1′-enyl-2-acyl-*sn*-glycero-3-phosphorylethanolamine aldehydohydrolase) activities were assayed with the plasmalogens in dispersed myelin as the substrate. The activities were 6.7 μmoles/mg protein per hr in oligodendroglia from white matter, and 1.1 and 0.6 for astroglia and neuronal perikarya from gray matter. Myelin had no plasmalogenase activity. Thus, both the normal catabolism of myelin plasmalogens and the accelerated hydrolysis of plasmalogens in demyelination is probably carried out by oligodendroglial plasmalogenase.

EFFECT OF AGE ON HYPERPHAGIA IN THE GENETICALLY OBESE ZUCKER RAT. B.A. Dilettuso and P.J. Wangsness (Dept. of Dairy and Animal Sci., The Pennsylvania State Univ., Univ. Park, Penn. 16802) *Proc. Soc. Exp. Biol. Med.* **154**, 1–5 (1977). Two experiments were conducted to determine if and when hyperphagia occurs in the genetically obese Zucker rat. In Experiment 1, lean and obese rats were offered food *ad libitum* from 7 to 19 weeks of age. Obese rats consumed significantly more total food than lean rats throughout the experiment. However, when food intake (FI) was expressed per unit of body weight (FI/BW) to compensate for the large differences in body weight between lean and obese rats, hyperphagia in the obese rats was only evident from 7 to 10 weeks of age. Thereafter, FI/BW became similar and at 19 weeks of age, FI/BW was significantly lower for obese rats compared to leans. Experiment 2 studied younger rats (3 to 11 weeks of age) and produced similar results. However, the increased FI/BW in obese rats was more pronounced at the early age and was evident from 3 to 7 weeks of age. It is concluded that obese rats were hyperphagic compared to lean rats, but only at an early age.

EFFECTS OF DIETARY CARBOHYDRATE ON SODIUM CYCLAMATE TOXICITY IN RATS FED A PURIFIED, LOW-FIBER DIET. B.H. Ershoff (Inst. for Nutr. Studies, Culver City, Calif. 90230) *Proc. Soc. Exp. Biol. Med.* **154**, 65–8 (1977). Immature male rats were fed a purified, low-fiber diet containing 5% sodium cyclamate but varying as to the source of dietary carbohydrate. On diets containing sucrose or dextrose as the dietary carbohydrate, animals exhibited a highly significant retardation in weight gain, lack of grooming, varying degrees of alopecia, extensive diarrhea, and high mortality within an experimental period of 14 days. With the exception of growth retardation during the first week of feeding, these effects except for a slight diarrhea were completely counteracted when the diet contained cornstarch as the source of dietary carbohydrate, and animals appeared grossly normal in all respects and comparable to those fed a purified, low-fiber, cyclamate-free diet. Intermediate results were obtained in rats fed sodium cyclamate in conjunction with a diet containing dextrin as the source of dietary carbohydrate.

PLASMA CHOLESTEROL LEVELS IN SUCKLING AND WEANED KITTENS, PUPPIES, AND GUINEA PIGS. R.M.G. Hamilton and K.K. Carroll (Dept. of Biochem., Univ. of Western Ontario, London, Ontario, Canada, N6A 5C1) *Lipids* **12**, 145–8 (1977). Plasma cholesterol levels in kittens and puppies were low at birth, rose during the suckling period, and then decreased at about the time of weaning. The increase during the suckling period was much greater in puppies than in kittens. No significant differences in plasma cholesterol levels were observed in puppies fed two different types of diet, horse meat or dog chow, after weaning. Guinea pigs had lower plasma cholesterols than either kittens or puppies. The level was highest on the first day after birth, decreased during the next 3 wk, and then remained fairly constant after the animals were weaned.

FAT-CONTAINING UTERINE SMOOTH MUSCLE CELLS IN "TOXEMIA": POSSIBLE RELEVANCE TO ATHEROSCLEROSIS? M.D. Haust, J.L. Heras and P.G. Harding (Depts. of Pathol. and Paediatrics and Dept. of Obstetrics and Gynaecol., Univ. of Western Ontario, London, Ontario, Canada) *Science* **195**, 1353–4 (1977). Uterine smooth muscle cells in "toxemia of pregnancy" contain varying amounts of fat-a feature to date believed to characterize only the arterial smooth muscle cells in atherosclerotic lesions. Thus, the smooth muscle cells at these two sites do not differ essentially in their reactivity to certain forms of injury; hypoxia may represent an injurious factor common to both "toxemia" and atherosclerosis. These observations imply that the view that the arterial smooth muscle cells are biologically different than are those elsewhere may no longer be tenable.

DIFFERENTIAL BIOSYNTHESIS OF MOLECULAR SPECIES OF 1,2-DIACYL-SN-GLYCEROLS AND PHOSPHATIDYLCHOLINES IN COLD AND WARM ACCLIMATED GOLDFISH (CARASSIUS AURATUS L.). B.J. Holub, J. Piekarski and J.F. Leatherland (Dept. of Nutr., Univ. of Guelph, Guelph, Ontario, Canada) *Lipids* **12**, 316–8 (1977). The initial incorporation of glycerol-^{3}H into the molecular species of liver 1,2-diacyl-*sn*-glycerols and phosphatidylcholines was studied in vivo using goldfish acclimated to 10 C and 30 C. A 1.5- and 2.2-fold higher proportion of the total radioactivity in the diacylglycerols from cold acclimated fish was found to be associated with the trienoic and pentaenoic species, respectively, when compared to warm acclimated fish. In the phosphatidylcholines, 1.9- and 1.3-fold greater percentages of the newly-incorporated radioactivity were found in tetraenoic and pentaenoic molecules, respectively, from cold relative to warm acclimated fish which suggests a preferential synthesis of these molecules relative to other molecular species in response to a lowering of environmental temperature. The present results indicate, therefore, that environmental temperature influences the complement of molecular species of diacylglycerols and phosphatidylcholines which fish produce by way of de novo biosynthesis in vivo.

STIMULATORY ACTION OF CYCLOHEXIMIDE ON GLUCOSE METABOLISM IN THE RAT EPIDIDYMAL FAT PAD. J.A. Garcia-Sainz, E. Pina and Victoria Chagoya de Sanchez (Dept. de Biol. Exp., Inst. de Biol., Univ. Nacional Autonoma de Mexico, Mexico 20, D.F.) *J. Lipid Res.* **18**, 93–8 (1977). The action of cycloheximide on some parameters of glucose and lipid metabolism was studied in vitro in epididymal fat pads from fasted rats. Incubation of fat pads with cycloheximide (1 μg/ml) for 2 hours resulted in a two-fold increase in glucose uptake, glucose oxidation, incorporation of glucose into lipids, and reesterification of free fatty acid. The increase in glucose oxidation was evident in experiments in which [U^{14}C], [1-^{14}C], or [6-^{14}C]glucose was added to the media, but it was absent when the media were supplemented with pyruvate. Incorporation of glucose into glycogen and accumulation of lactate in the medium were not seriously modified by the presence of cycloheximide. The stimulatory effect of cycloheximide on incorporation of glucose into lipids was absent when insulin or cortisol was added to the medium. A cycloheximide-mediated increase in glucose uptake seems to be responsible for the subsequent changes in glucose metabolism, and would seem to be independent of an inhibition in protein synthesis; puromycin and actinomycin-D did not mimic the cycloheximide action on glucose incorporation into lipids.

ANALYSIS OF RAT SERUM APOLIPOPROTEINS BY ISOELECTRIC FOCUSING. I. STUDIES ON THE MIDDLE MOLECULAR WEIGHT SUBUNITS. L.I. Gidez, J.B. Swaney and S. Murnane (Depts. of Biochem. and Med., Albert Einstein Col. of Med., Yeshiva Univ., Bronx, N.Y. 10461) *J. Lipid Res.* **18**, 59–68 (1977). Analytical isoelectric focusing (IEF) has been applied to the study of the apolipoprotein components of rat serum high density and very low density lipoproteins. The apoliproteins were separated on 7.5% polyacrylamide gels containing 6.8% urea, with a pH gradient of 4–6. The middle molecular weight range apolipoproteins purified by electrophoresis on gels containing sodium dodecyl sulfate (SDS). The A-I protein focused as 4 to 5 bands from pH 5.46 to 5.82; the A-IV protein and the arginine-rich protein each focused as 4 to 6 bands from pH 5.31 to 5.46. The low molecular weight proteins focused from pH. 4.43 to 4.83 and are the subject of a separate communication. Comparisons of the IEF method with SDS gel electrophoresis, polyacrylamide gel electrophoresis in urea, and Sephadex chromatography are also reported. Additional studies were also carried out that tend to rule out carbamylation or incomplete unfolding of the proteins in the presence of urea as the causes of the observed heterogeneity.

DIURNAL CHANGES IN PLASMA AND LIVER LIPIDS AND LIPOPROTEIN LIPASE ACTIVITY IN HEART AND ADIPOSE TISSUE IN RATS FED A HIGH AND LOW FAT DIET. P. De Gasquet, S. Griglio, E. Pequignot-Planche and M.I. Malewiak (Unité de Recherches Diététiques, Inst. Nat'l. de la Santé et de la Recherche Med. Hopital Bichat, 170 Boulevard Ney, F-75018 Paris, France) *J. Nutr.* **107**, 199–212 (1977). In order to evaluate the respective roles of adipose and muscle lipoprotein lipase (LPL) in the clearing of alimentary lipemia and the role of the resulting nonesterified fatty acids (NEFA) in controlling hepatic ketogenesis and liver triglyceride content, a number of parameters related to lipid metabolism were studied over the 24 hour period (the dark period being from 1930 to 0730 hours), in rats ad libitum fed either a low-fat (LF) or a high-fat (HF) diet containing respectively 1.1% and 41.5% lard. The findings clearly indicate that in HF rats, muscle LPL controls the postprandial rise in plasma NEFA concentrations, which in turn appear to determine the extent of ketonemia and liver triglyceride changes. The possible control of these metabolic events by insulin is discussed.

MEASUREMENT OF THE ABSOLUTE RATES OF CHOLESTEROL BIOSYNTHESIS IN ISOLATED RAT LIVER CELLS. G.F. Gibbons and C.R. Pullinger (Med. Res. Council Lipid Metab. Unit., Hammersmith Hosp., London W12 0HS, U.K.) *Biochem. J.* **161**, 321–30 (1977). Triparanol [2-(4-chlorophenyl)-1-(4-diethylaminoethoxyphenyl)-1-*p*-tolylethanol] at a concentration of 2 μM has no effect on the overall conversion of [2-^{14}C]acetate into C_{27} sterols by isolated liver cells. In the presence of triparanol, however, the formation of radioactive cholesterol is inhibited by 85–90% and the balance of radioactivity appears in the C_{27} sterol desmosterol (cholesta-5,24-dien-3β-ol). The very small weights of desmosterol which accumulate under these conditions were, as a routine, quantitatively converted into the heptafluorobutyrate 3-enol ester of cholesta-4,24-dien-3-one. This derivative has a high electron-capturing capability, a property that enables extremely small quantities (< 0.25 pmol) of the material to be accurately measured by gas chromatography with electron-capture detection. Measurements of the mass and specific radioactivity of the newly biosynthesized desmosterol formed in the presence of triparanol provides an accurate assessment of the amount of cholesterol that would be synthesized by the liver cells in the absence of the drug.

SITE OF SYNTHESIS OF PHOSPHATIDIC ACID AND DIACYLGLYCEROL IN SPINACH CHLOROPLASTS. J. Joyard and R. Douce (Dept. de Recherche Fondamentale, Biol. Vegetale, Centre d'Etudes Nucleaires et Univ. Sci. et Med. de Grenoble 85X, F 38041, Grenoble-Cedex, France) *Biochim. Biophys. Acta* **486**, 273–85 (1977). The enzymatic synthesis of lysophosphatidic acid, phosphatidic acid, monoacylglycerol and diacylglycerol from *sn*-[^{14}C]glycerol 3-phosphate occurs in purified chloroplasts. The results indicate that: the chloroplast extract contains a soluble acylase (acyl-CoA: *sn*-glycerol 3-phosphate acyltransferase); the envelope fraction contains an acyl-CoA synthetase, a bound acylase (acyl-CoA: acyl-*sn* glycerol 3-phosphate acyltransferase) and a phosphatidic acid phosphatase; without chloroplast extract in the incubation medium, the envelope is unable to incorporate *sn*-glycerol 3-phosphate into phosphatidic acid and diacylglycerol; addition of chloroplast extract to the incubation medium induced a fast increase of the incorporation of *sn*-glycerol 3-phosphate into phosphatidic acid and diacylglycerol; thylakoids being unable to incorporate *sn*-glycerol 3-phosphate (in presence or absence of soluble chloroplast extract in the incubation medium) our results indicate that the envelope of spinach chloroplast is the site of phosphatidic acid and diacylglycerol synthesis; diacylglycerol actively synthesized by the envelope is also the substrate for the first galactosylation enzyme.

KINETIC STUDY OF LIPOXYGENASE-HYDROPEROXYLINOLEIC ACID INTERACTION. H. Aoshima, T. Kajiwara, A. Hatanaka, H. Nakatani and K. Hiromi (Dept. of Chem., Faculty of Liberal Arts, Yamaguchi Univ., Yamaguchi 753, Japan) *Biochim. Biophys. Acta* **486**, 121–6 (1977). Interaction of lipoxygenase with hydroperoxylinoleic acid, which is the product of this enzyme reaction and acts as an activator, was studied kinetically by the fluorescence stopped-flow method. The kinetic features are consistent with a two-step mechanism involving a fast bimolecular association process followed by a slow unimolecular process. The dissociation constant of the bimolecular process was 3 ($\pm$ 2) · 10^{-5}M, which was appreciably dependent on temperature and pH, in contrast to the rate constant of the latter process. The enthalpy and the entropy of activation for the unimolecular process were estimated to be 21 kcal/mol and 20 e.u., respectively. The pH dependence of the rate constant indicated that an ionizable group with pK of about 8.6 is involved in the interaction. Linoleic acid, the substrate of lipoxygenase, and oleic acid inhibited the interaction between the lipoxygenase and the hydroperoxylinoleic acid by reducing the rate. A series of saturated monohydric alcohols also reduced the rate of the interaction as the chain length of the alcohols increases, though methanol and ethanol increased the rate of the interaction.

DIETARY-INDUCED OVEREATING IN EXPERIMENTAL ANIMALS. R.B. Kanarek and E. Hirsch (Dept. of Nutr., Harvard School of Public Health, Boston, Mass. 02115) *Fed. Proc.* **36**, 154–8 (1977). Modification in dietary conditions can result in small, but consistent, increases in caloric intake which over time accumulate to substantial increases in body weight. Allowing rats access to either high fat diets or a variety of highly palatable foods can lead to obesity. Recent experiments also have shown that providing access to sweet carbohydrate solutions in addition to a complete diet can cause weight gains in normal neurologically intact adult rats. Although animals increase consumption of carbohydrate on this dietary regime, they continue to maintain minimum requirements for other dietary nutrients. It appears that increasing the palatability of the diet can lead to overeating, but cannot induce the rat to select a diet that is deficient in protein, fat, vitamins, or minerals. Further exploration of the conditions associated with dietary-induced overeating in animals may provide insights into conditions related to obesity in man.

OUTSIDE-INSIDE DISTRIBUTION AND TRANSLOCATION OF LYSOPHOSPHATIDYLCHOLINE IN PHOSPHATIDYLCHOLINE VESICLES AS DETERMINED BY ^{13}C-NMR USING (N-$^{13}CH_3$)-ENRICHED LIPIDS. B. de Kruyff, A.M.H.P. Van Den Besselaar and L.L.M. Van Deenen (Inst. of Mol. Biol., Univ. of Utrecht, Univ. Ctr. "De Uithof", Transitorium 3, Paduallan 8, Utrecht, The Netherlands) Biochim. Biophys. Acta **465**, 443–53 (1977). The outside-inside distribution of palmitoyl lysophosphatidylcholine and dioleoyl phosphatidylcholine in mixed sonicated vesicles is measured with (N-$^{13}CH_3$)-labelled lipids using ^{13}C NMR and Dy^{3+} as an impermeable shift reagent. Palmitoyl lysophosphatidylcholine is preferentially localised in the outside layer of the vesicle membrane. Incorporation of cholesterol in the vesicle diminished the extent of lysophosphatidylcholine asymmetry. Palmitoyl lysophosphatidylcholine added to dioleoyl phosphatidylcholine vesicles is incorporated in the outer monolayer of the vesicle. Even after 40 h less than 2% of the lysophosphatidylcholine could be detected in the inner monolayer. Since in the cosonicated vesicles 17% of the lysophosphatidylcholine is present in the inner monolayer it can be concluded that the transmembrane movement of lysophosphatidylcholine across the lipid bilayer of these vesicles is an extremely slow process.

INTESTINAL REGULATION OF HEPATIC CHOLESTEROL SYNTHESIS: AN HYPOTHESIS. C.L. Krumdieck and K.-J. Ho. (Dept. of Biochem. and Pathol., Univ. of Alabama in Birmingham, School of Med., Birmingham, Ala. 35294) *Am. J. Clin. Nutr.* **30**, 255–61 (1977). The serum cholesterol level in man is predominantly dependent on the rate of endogenous synthesis. The liver is the main site of cholesterol synthesis and this activity is in turn intimately related to the ingestion of food. When no food is present in the intestine, as during periods

of total parenteral nutrition, both normal individuals and patients with familiar hypercholesterolemia show a pronounced lowering of their serum cholesterol. The establishment of a portacaval shunt makes this a permanent change. This and other evidence lead us to postulate the existence of an intestinal factor capable of stimulating hepatic cholesterogenesis. The function of this factor would be to increase the synthesis of cholesterol whenever there is a need for increased amounts of intraluminal bile acids.

MITOCHONDRIAL BIOGENESIS IN CULTURED MAMMALIAN CELLS. III. SYNTHESIS OF MITOCHONDRIAL PHOSPHOLIPIDS BY SUBCELLULAR FRACTIONS ISOLATED FROM NORMAL AND CHLORAMPHENICOL-TREATED BHK-21 CELLS. J.H. Lipton and W.C. McMurray (Dept. of Biochem., Univ. of Western Ontario, London, Ontario, N6A 5C1, Canada) *Biochim. Biophys. Acta* **486**, 228–42 (1977). The capacity of subcellular fractions isolated from chloramphenicol-treated BHK-21 cells to synthesize various mitochondrial phospholipids in vitro was examined. Both mitochondria and microsomes showed the capacity to acylate *sn*-glycerol 3-phosphate and dihydroxyacetone phosphate to lysophosphatidic acid and acyldihydroxyacetone phosphate and subsequently to phosphatidic acid. Both processes are inhibited in mitochondria from chloramphenicol-treated cells. The synthesis of CDPdiacylglycerol in mitochondria or microsomes, and the synthesis of phophatidylinositol and of phosphatidylcholine in microsomes were stimulated in treated cells. A slight stimulation was also observed in the synthesis of phosphatidylglycerol and diphosphatidylglycerol when the labelled precursor was *sn*-glycerol 3-phosphate in treated cells, although the process was inhibited with labelled glycerol as the precursor. Conversion of phosphatidylglycerol phosphate to phosphatidylglycerol by mitochondria was rate limiting unless the post-microsomal supernatant fraction was added. These results are discussed in regard to the observed inhibition of phospholipid synthesis in BHK-21 cells in culture by chloramphenicol.

SPECIFIC REQUIREMENT OF LYSOPHOSPHATIDYLCHOLINE FOR PALMITOYL-CoA SYNTHETASE OF CHICKEN INTESTINE. B.R. Lokesh, A.M. Rao and S.K. Murthy (Dept. of Biochem., Indian Inst. of Sci., Bangalore-560 012 India) *Biochim Biophys. Acta* **486**, 341–50 (1977). The presence of palmitoyl-CoA synthetase (EC 6.2.1.3) in the brush border-free particulate fraction of chicken intestinal mucosa is demonstrated. The enzyme was dependent on the simultaneous presence of lysophosphatidylcholine and Triton X-100 as well as ATP, CoA and Mg^{2+} for maximal activity. Lysophosphatidylcholine could not be replaced by other lipids. Enzyme preparations solubilized by Triton X-100 or lysophosphatidylcholine were still dependent on the presence of detergents for maximal activity.

A COMPARATIVE IN VIVO STUDY OF INTESTINAL ABSORPTION OF BILIARY PHOSPHATIDYLCHOLINES AND MICELLAR PHOSPHATIDYLCHOLINES IN THE RAT. G. Nalbone, D. Lairon, H. Lafont, J. Amic, N. Domingo and J.C. Hauton (I.N.S.E.R.M., Groupe de Res. sur le Transport des Lipides, 46, Bd de la Gaye, 13009-Marseille, France) *Lipids* 12, 149–52 (1977). An *in vivo* study was performed using rats with the purpose of comparing the absorption of native biliary and purified phosphatidylcholines. The latter were purified from bile and solubilized in the form of mixed micelles of bile salts-phosphatidylcholines-cholesterol. The animals all bore bile duct diversions, and were divided into two groups: one had a normal pancreatic secretion while in the other group the pancreatic duct was ligated. Animals with normal pancreatic secretion showed comparable rates of absorption of micellar and biliary phosphatidylcholines. In the absence of normal pancreatic secretion, the rate of absorption of biliary phosphatidylcholines was unchanged, whereas that of micellar phosphatidylcholines markedly decreased. The results are consistent with the concept that some biliary phosphatidylcholines are absorbed independently of pancreatic secretion in an unhydrolyzed form.

INDUCTION OF HMG CoA REDUCTASE BY THE ADMINISTRATION OF 20,25-DIAZACHOLESTEROL. R. Langdon, S. El-Masry and R.E. Counsell (Lab. of Med. Chem., College of Pharmacy, Univ. of Mich., Ann Arbor, Mich. 48109) *J. Lipid Res.* **18**, 24–31 (1977). This paper describes the direct examination of HMG CoA reductase activity in rats treated with 20,25-diazacholesterol. Conversion of acetyl CoA and HMG CoA to mevalonate increased to over 200% of control values in the microsomes and in the 12,000 *g* supernatant of liver homogenates after 5 days of treatment. The time course of induction coincided with the development of hypocholesterolemia. Animal weights, liver weights, and microsomal protein content did not vary significantly between animal groups. Incubations to which the compound was introduced in vitro in concentrations as great as 0.5 mM produced no significant difference from control incubations. Similar treatment of the animals with 7-ketocholesterol, a cholesterol derivative reported to repress HMG CoA reductase activity in tissue cultures, produced no appreciable difference in reductase activity or serum steroid levels in vivo.

EFFECT OF DIET COMPOSITION ON METABOLIC ADAPTATIONS TO HYPOCALORIC NUTRITION: COMPARISON OF HIGH CARBOHYDRATE AND HIGH ISOCALORIC DIETS. S.B. Lewis, J.D. Wallin, J.P. Kane and J.E. Gerich (Clin. Invest. Ctr., Naval Regional Med. Ctr., Oakland, Calif. 94627) *Am. J. Clin. Nutr.* **30**, 160–70 (1977). The metabolic consequences of two hypocaloric diets were assessed in 10 obese men. The study, performed on a metabolic ward, compared the response of these men to two cholesterol-free liquid formula diets of differing composition (10 kcal/kg per day, 70% carbohydrate, 20% protein, 10% fat versus 70% fat, 20% protein, 10% carbohydrate) but identical in calories. These were administered for 14 days in a random order and each diet was preceeded by a 7-day control weight maintenance diet (30 kcal/kg per day, 40% carbohydrate, 20% protein, 40% fat). The low calorie diets were well tolerated by the men and effected similar losses of nonaqueous body weight. Fasting glucose and insulin decreased significantly in these men after they ingested either weight loss diet for 14 days, but the change in each parameter was greater for high fat as compared to high carbohydrate (15% versus 7% and 67% versus 35%, respectively, $P < 0.01$). In contrast, fasting glucagon concentration decreased in these subjects to a greater extent in response to the high carbohydrate diet (35% versus 16%, $P < 0.01$).

TURBIDIMETRIC ULTRACENTRIFUGATION. APPLICATION TO THE STUDY OF HUMAN SERUM VERY LOW DENSITY LIPOPROTEIN DISTRIBUTIONS. S.K. Ma, V.N. Schumaker and C.M. Knobler (Dept. of Chem. and Mol. Biol. Inst., Univ. of California, Los Angeles, Calif. 90024) *J. Biol. Chem.* **252**, 1728–31 (1977). In this communication it is shown that the sedimentation coefficient distribution may be accurately measured for very large particles using turbidimetric techniques and the ultraviolet-scanning analytical ultracentrifuge. A principal advantage is that turbidity is a function of the product of concentration and molecular weight; thus, large particles may be observed even when present in very small amounts. We propose to call this method of analysis "turbidimetric ultracentrifugation." We have used turbidimetric ultracentrifugation to determine the sedimentation coefficient distribution for a sample of human serum very low density lipoproteins. This distribution is compared to that found with conventional schlieren techniques with good agreement.

CHROMATIN PHOSPHOLIPIDS IN NORMAL AND CHRONIC LYMPHOCYTIC LEUKEMIA LYMPHOCYTES. F.A. Manzoli, N.M. Maraldi, L. Cocco, S. Capitani, and A. Facchini (Inst. of Histology and Gen. Embryology, Univ. of Chieti and Bologna, Italy) *Cancer Res.* **37**, 843–9 (1977). Certain phospholipids are associated with the nonhistone chromosomal proteins extracted from normal B- and chronic lymphocytic leukemia lymphocytes. The ratio of phospholipids to nonhistone chromosomal proteins was constant with the different methods used for isolating nuclei and extracting the chromatin, although the various methods allowed a different recovery of total lipids from chromatin. Three phospholipids were extractable from the nonhistone protein fraction, but their respective ratios varied in chronic lymphocytic leukemia compared to normal B- lymphocytes. The most significant variation concerns the reduction of sphingomyelin content in leukemic lymphocytes, since this phospholipid in vitro affects both DNA stability and transcription.

CHARACTERIZATION OF DOG SMALL INTESTINAL FUCOLIPIDS WITH HUMAN BLOOD GROUP A ACTIVITY. DIFFERENCES IN DOG AND HUMAN A-ACTIVE FUCOLIPIDS. J.M. McKibbin, E.L. Smith, Jan-E. Mansson, and Yu-Teh Li (from the Dept. of Biochem., Univ. of Ala. in Birmingham, Birmingham, Ala. 35294) *Biochemistry* **16**, 1223–8 (1977). Glycolipids containing fucose (fucolipids) which carried human blood group A activity were isolated from a number of dog small intestines and analyzed. On the basis of sugar analysis, methylation, periodate oxidation, enzyme degradation, mass spectrometry, and immunologic studies, a structure is proposed for these substances. The ceramides of the dog fucolipids contained only hydroxylated fatty acids with 85% saturated and 15% monoenoic acids

ranging from 16 to 25 carbon atoms. Sphingosine and phytosphingosine comprised 48% each of the long chain bases. An A-active fraction isolated from human small intestine was shown to have two components, one of which was immunologically distinct and the other identical with the dog intestinal fucolipids. The human fraction differed from the dog fucolipids in migration on thin-layer chromatography and contained two types of amino sugar substitution. It is proposed that the human fraction was composed of two fucolipids with incomplete structures.

CHOLESTEROL β-D-GLUCOSIDE-6′-*O*-PALMITATE, A METABOLITE OF PYTHIUM SYLVATICUM. T.C. McMorris and R.H. White (Dept. of Chem., Univ. of Cal., San Diego, La Jolla, Calif. 92093 U.S.A.) *Biochim. Biophys. Acta* **486**, 308–12 (1977). Cholesterol β-D-glucoside-6′-*O*-palmitate has been identified as a polar metabolite in the mycelium of mated cultures of *Pythium sylvaticum* grown in the presence of cholesterol. The structure was confirmed by synthesis of the metabolite. Similar steroid β-D-glucoside-6′-*O*-palmitates were obtained from β-sitosterol and campesterol when these sterols were added to cultures of *P. sylvaticum*. Corresponding esters of myristic and stearic acids were also detected.

SOLUBLE RAT ADIPOCYTE PHOSPHATIDATE PHOSPHATASE ACTIVITY: CHARACTERIZATION AND EFFECTS OF FASTING AND VARIOUS LIPIDS. F. Moller, P. Green and E.J. Harkness (Dept. of Biochem., Queen's Univ., Kingston, Ontario, Canada) *Biochim. Biophys. Acta* **486**, 359–68 (1977). Phosphatidate phosphatase (phosphatidate phosphohydrolase, EC 3.1.3.4) was present at very high specific activity in the soluble fraction of isolated rat adipocytes. Using phosphatidate in aqueous dispersion 90% of its hydrolysis depended on the presence of Mg^{2+}. Mg^{2+} appeared to almost saturate the enzyme at 20–40mM with no indication of an optimum. The substrate concentration was optimum at 1.2mM and the pH at 6.8. Initial rates were linear for only 4–5 min at optimum conditions. Increasing inhibition occurred at high phosphatidate concentrations. At optimum conditions acid or alkaline phosphatase activity was not measurable. The Mg^{2+}-dependent activity was enhanced by 3-*sn*-phosphatidylcholine and inhibited by albumin, 3-*sn*-phosphatidylethanolamine, 3-*sn*-phosphatidylinositol, diacylglycerol, oleoyl-CoA, and oleate. Oleoyl-CoA was the most potent "effector". Fasting for 24, 48 and 72h decreased the activity both relative to the protein and to DNA. The activity thus decreased to about one-third of that of the fed rat during 72h of fasting. The effects of Mg^{2+}, various lipids, and fasting may indicate that some form of control of glyceride synthesis can be exerted through the soluble phosphatidate phosphatase.

LIPID METABOLISM IN PLASMA, LIVER, AND ADIPOSE TISSUE OF RATS WITH EXPERIMENTAL CHRONIC NEPHROTIC SYNDROME. R.J. Morin, W.D. Davidson, S.J. Rorke and L.S.S. Guo (Depts. of Pathol. and Med., Harbor Gen. Hosp./UCLA School of Med., Torrance, Calif. 90509) *Lipids* **12**, 208–14 (1977). Plasma, liver, and adipose tissue lipid composition and synthesis from [1-^{14}C]acetate were studied three months following induction of nephrotic syndrome in rats by injection of antiglomerular basement membrane protein. Plasma triglyceride concentrations and specific radioactivities were elevated, and the triglycerides contained increased proportions of oleic acid. Plasma cholesterol and phospholipid concentrations were also increased, but free fatty acid levels were not. Liver triglyceride concentrations were decreased and incorporation of [1-^{14}C]acetate into liver triglycerides was also depressed below that of normal controls. Nephrotic rat liver triglycerides contained a higher proportion of oleic acid and lower arachidonic acid than did controls. Incorporation of [1-^{14}C]acetate into adipose tissue lipids of the nephrotic rats was increased, and the proportion of palmitic acid was decreased. In the chronic nephrotic rat, the major source of the increased plasma triglycerides may be fatty acids mobilized from adipose tissue stores.

EFFECTS OF ANTI-MICROTUBULAR AGENTS AND CYCLOHEXIMIDE ON THE METABOLISM OF CHYLOMICRON CHOLESTERYL ESTERS BY HEPATOCYTE SUSPENSIONS. A. Nilsson (Dept. of Physiol. Chem., Univ. of Lund, Lund, Sweden) *Biochem. J.* **162**, 367–77 (1977). Post-heparin plasma that promoted rapid hydrolysis of about 90% of the triacylglycerol markedly stimulated the uptake or binding of chylomicron cholesteryl ester by suspended hepatocytes. The net hydrolysis of chyle cholesteryl ester after the uptake by the cells was, however, slower than *in vivo*. The cholesteryl ester uptake in the presence of post-heparin plasma was larger if the cells had been preincubated for 2 hr. It was inhibited by the presence of colchicine, vinblastine or cycloheximide during the preincubation, and by mild trypsin treatment of the preincubated cells. The results suggested that the anti-microtubular agents, but not cycloheximide, also inhibited the hydrolysis of chyle cholesteryl ester after uptake or binding to the cells. The uptake of isolated chylomicron remnant particles was more efficient than that of native chyle lipoproteins. It was, however, still stimulated by heparin alone and by post-heparin plasma. The heparin-stimulated uptake was markedly decreased if cycloheximide was present during the preincubation period.

FATTY ACID SYNTHESIS IN ESCHERICHIA COLI IS INDIRECTLY INHIBITED BY PHENETHYL ALCOHOL. W.D. Nunn (Dept. of Mol. Biol. and Biochem., Univ. of California, Irvine, Calif. 92717) *Biochemistry* **16**, 1077–81 (1977). Experiments were performed to determine how phenethyl alcohol inhibits phospholipid synthesis in *E. coli*. At a nonbacteriostatic concentration, the drug reduces the rate of de novo fatty acid and phospholipid synthesis by 60 to 70%. The inhibition of fatty acid synthesis was found to be a secondary consequence of the inhibition of phospholipid synthesis. Phenethyl alcohol reduces the rate of incorporation of exogenous fatty acids into the phospholipids of a fatty acid auxotroph by 60%. These results indicate that this drug controls phospholipid synthesis beyond the level of fatty acid synthesis. Phenethyl alcohol inhibits the synthesis of phospholipids containing saturated fatty acids to a greater extent than it does the synthesis of phospholipids containing unsaturated fatty acids. It controls the synthesis of phospholipids containing saturated fatty acids at both the level of fatty acid synthesis and the level of incorporation of the saturated fatty acids into phospholipids. The synthesis of phospholipids containing unsaturated fatty acids is inhibited at the level of incorporation of the fatty acids into phospholipids.

PROSTAGLANDIN RECEPTOR-ADENYLATE CYCLASE SYSTEM IN PLASMA MEMBRANES OF RAT LIVER AND ASCITES HEPATOMAS, AND THE EFFECT OF GTP UPON IT. N. Okamura and H. Terayama (Zoo. Inst., Faculty of Sci., Univ. of Tokyo, Tokyo, Japan) *Biochim. Biophys. Acta* **465**, 54–67 (1977). Adenylate cyclase in plasma membranes from rat liver was stimulated by prostaglandin E_1, and to a lesser extent by prostaglandin E_2. Prostaglandin $F_{1\alpha}$, $F_{2\alpha}$ and A_1 did not stimulate the cyclase. The prostaglandin E_1-mediated activation was found to require GTP when the substrate ATP concentration was reduced from 3 mM to 0.3 mM in the reaction mixture. Adenylate cyclase of the plasma membranes from rat ascites hepatomas AH-130 and AH-7974 was not stimulated by prostaglandin E_1 in the presence or the absence of GTP, although the basal activity of adenylate cyclase as well as its stimulation by GTP alone were similar to normal liver plasma membranes. GTP alone was found to increase V of adenylate cyclase of liver plasma membranes, while GTP plus prostaglandin E_1 was found to decrease K_m of adenylate cyclase in addition to the increase of V to a further extent.

INVOLVEMENT OF CYTOCHROME b_5 IN THE OXIDATIVE DESATURATION OF LINOLEIC ACID TO γ-LINOLENIC ACID IN RAT LIVER MICROSOMES. T. Okayasu, T. Ono, K. Shinojima and Y. Imai (The Second Dept. of Biochem., School of Med., Hokkaido Univ., Sapporo 060, Japan) *Lipids* **12**, 267–71 (1977). The effects of antibodies against microsomal electron-transport components on the in vitro activity of Δ^6-desaturation of linoleic acid to γ-linolenic acid have been studied in intact microsomal membranes of rat liver. Reduced nicotinamide adenine dinucleotide (NADH) or reduced nicotinamide adenine dinucleotide phosphate (NADPH) (0.87mM) served as electron donors, and effectively prompted the Δ^6-desaturase activities with yields of about 1.1 to 1.3 nmol per mg of protein in 10 min. Of the two antibodies studied under the same in vitro conditions, i.e., rabbit antisera preparations against rat liver microsomal hydrophilic parts of cytochrome b_5 and NADPH-cytochrome c reductase, only the antibody against cytochrome b_5 demonstrated a marked ability to inhibit the Δ^6-desaturase activity. This evidence supports a participation of cytochrome b_5 in the Δ^6-desaturation of linoleic acid and suggests a pathway analogous to the Δ^9-desaturation of stearyl-CoA.

ISOLATION, CHEMICAL CHARACTERIZATION, AND BIOPHYSICAL PROPERTIES OF THREE DIFFERENT ABNORMAL LIPOPROTEINS: LP-X_1, LP-X_2, AND LP-X_3. J.R. Patsch, K.C. Aune, A.M. Gotto and J.D. Morrisett (Depts. of Med. and Biochem., Baylor College of Med., The Methodist Hosp., Houston, Tex. 77030) *J. Biol. Chem.* **252**, 2113–20 (1977). Three different but related abnormal lipoprotein species, LP-X_1, LP-X_2, and LP-X_3, have been isolated from cholestatic plasma by ethanol precipitation

and zonal ultracentrifugation. All three populations are rich in phospholipids (64.9 to 67.5%) and cholesterol (23.0 to 26.8%) but poor in cholesteryl esters (0.4 to 1.9%), triglycerides (1.8 to 3.2%), and protein (3.2 to 6.7%) with differences in chemical composition which result in buoyant densities (1.038, 1.049, and 1.058, respectively) to allow their separation. LP-X_1, LP-X_2, and LP-X_3 exhibited apparent flotation rates of 17.3, 9.7, and 3.2 Svedbergs and Stokes radii of 339, 343, and 294 Å, respectively. As determined from circular dichroic measurements, the protein constituents of all three particles possessed a high degree of α helical structure (41 to 65%). Each LP-X particle exhibited abnormally low fluidity as evaluated by electron paramagnetic resonance. All of the particles contained human serum albumin and the C-proteins as major protein constituents, but only LP-X_2 and LP-X_3 contained apolipoprotein A-I and apolipoprotine E.

LIPID METABOLISM IN EARLY DEVELOPMENT USING LABELED PRECURSORS INCORPORATED DURING OOGENESIS AND IN CELL-FREE EMBRYO HOMOGENATES. A.M. Pechen and N.G. Bazan (Inst. de Investigaciones Bioquimicas, Consejo Nacional de Investigaciones Cientificas y Tecnicas, Univ. Nacional del Sur, Bahia Blanca, Argentina) *Lipids* 12, 131–4 (1977). Embryos of the toad, *Bufo arenarum, Hensel*, taken during early stages of development used to survey the [U-^{14}C]glycerol and ^{32}P lipid labeling. When precursors were supplied at the time of oogenesis, large differences in specific activities of phospholipid were observed. Using ^{32}P, a steep rise as a function of development was evidenced. Triglycerides contained much higher proportions of [U-^{14}C]glycerol than phospholipids when administered to the female toad along with a pituitary homogenate. However, lack of [U-^{14}C]glycerol uptake into lipids was observed when cell-free homogenates of eggs at different stages of development ranging from unfertilized oocyte to midgastrula were incubated in unsupplemented amphibian Ringer. At the later stage, significant de novo biosynthesis of lipids from glycerol began to be measurable, whereas during cell cleavage intracellular redistribution of preformed phosphoglycerides was used for membrane assembly.

LIPOPROTEIN LIPASE ACTIVITY AT ONSET OF DEVELOPMENT OF WHITE ADIPOSE TISSUE IN NEWBORN RATS. E. Pequignot-Planche, P. De Gasquet, A. Boulange and N.T. Tonnu (Unite de Recherches Dietetiques, Inst. Nat'l. de la Sante et de la Recherche Med., Hopital Bichat, 170 Boulevard Ney, 75018 Paris, France) *Biochem. J.* 162, 461–3 (1977). The low triacylglycerol concentration in inguinal tissue of newborn rats did not change during the first 6h after birth, despite the relatively high lipoprotein lipase activity in the tissue. Subsequently triacylglycerol concentration and enzyme activity rose in parallel. The results show that lipoprotein lipase activity was present in the tissue before fat accumulation.

THE REACTIVITIES OF TYROSINE AND TRYPTOPHAN RESIDUES IN LIPID-BOUND CYTOCHROME b_5. J. Poensgen and V. Ullrich (Dept. of Physiol. Chem., Univ. of the Saarland, Homburg/Saar, G.F.R.) *Biochim. Biophys. Acta* **465**, 34–45 (1977). Purified cytochrome b_5 from rabbit liver microsomes was bound to liposomes prepared from microsomal lipids. Tyrosyl and tryptophyl side chains of the protein were modified by water-soluble reagents and the reactivities of these amino acid residues in the liposome-bound cytochrome b_5 were compared to those of the free protein. At pH 13, 80% of the tyrosines in lipid-free cytochrome b_5 ionized immediately, whereas in the lipid-bound protein only 65% ionized within the first minute. In contrast, acetylation with acetylimidazole resulted in the conversion of all 5 tyrosine groups of lipid-free as well as lipid-bound cytochrome b_5 into 0-acetylated derivatives, which upon treatment with hydroxylamine were completely deacetylated. It was concluded that the two tyrosines in the region linking the protein to the membrane are not shielded by the lipid bilayer, but that of the three tryptophans in the same region one is completely buried in the membrane, whereas the remaining two tryptophans may be both partly exposed to the solvent or alternatively, one may be partially and the other completely exposed.

A PHOSPHOLIPID DERIVATIVE OF CYTOSINE ARABINOSIDE AND ITS CONVERSION TO PHOSPHATIDYLINOSITOL BY ANIMAL TISSUE. C.R.H. Raetz, M.Y. Chu, S.P. Srivastava and J.G. Turcotte (Dept. of Biochem., College of Agr. and Life Sci., Univ. of Wisconsin, Madison, Wis. 53706) *Science* **196**, 303–5 (1977). We have synthesized an analcg (ara-CDP-DL-dipalmitin) of cytidine diphosphate diglyceride (CDP-diglyceride) in which the antitumor drug, cytosine arabinoside, is substituted for the cytidine moiety. Enzymes in rat and human liver convert this analog to phosphatidylinositol, thereby releasing cytosine arabinoside-5'-monophosphate, an obligatory intermediate in the activation of cytosine arabinoside. Unlike cytidine diphosphate diglyceride, however, ara-CDP-DL-dipalmitin is not an efficient substrate for phosphatidylglycerophosphate synthesis in liver or phosphatidylserine in *Escherichia coli*. The antitumor activity of ara-CDP-DL-dipalmitin in mice bearing L5178Y leukemia is described.

ACTION OF THREE BILE ACIDS ON HEPATIC AND INTESTINAL CHOLESTEROGENESIS IN THE RAT. M.O. Reynier, C.H. Marteau, J.L. Vigne, A. Mule, C. Crotte and A. Gerolami (Unite de Res. de Pathologie Digestive, U 31 INSERM 46, chemin de la Gaye, 13009 Marseille, France) *Lipids* 12, 254–7 (1977). Incorporation of [1-^{14}C]acetate into cholesterol by subcellular particles from the liver and the small intestine of rats with a biliary diversion and a duodenal perfusion of sodium taurocholate, taurochenodeoxycholate or taurodehydrocholate, was studied in vitro. In the liver, taurochenodeoxycholate prevented the increase of cholesterol synthesis induced by biliary drainage. Taurocholate had no action on cholesterol synthesis at any time, day or night. Intestinal synthesis of cholesterol was reduced by taurocholate and taurochenodeoxycholate but was not modified by taurodehydrocholate infusion.

ORIGIN OF FATTY ACIDS OF CHOLESTERYL ESTER ACCUMULATED BY Fu5AH CELLS IN CULTURE. J.M. Rosen and G.H. Rothblat (The Wistar Inst., 36th and Spruce Sts., Philadelphia, Penn. 19104) *Lipids* 12, 222–7 (1977). The Fu5AH hepatoma cell line accumulates cholesteryl ester (CE) upon incubation in medium supplemented with hyperlipemic serum or hyperlipemic serum lipoproteins. This cell line was used to investigate the origin of the fatty acids esterified to cholesterol in intracellular accumulations of CE. The intracellular CE-fatty acid distribution was found to be markedly different from that of the lipoprotein which stimulated the accumulation. Free fatty acids added to the culture medium were found esterified to cholesterol in the cells, demonstrating that cellular esterification contributes to the accumulation of CE. Using a subline of Fu5AH cells containing radioactively labeled intracellular fatty acids, it was found that about one-third of the fatty acid moiety of CE accumulated by the cells during a 24 hr incubation with hyperlipemic serum was derived from endogenous fatty acids. The drug chloroquine was found to inhibit cellular cholesterol esterification, so that only 4% of CE-fatty acids were derived from endogenous fatty acids. Evidence is presented suggesting a major role for cellular esterification in CE accumulation by Fu5AH cells.

GLYCEROKINASE IN HUMAN ADIPOSE TISSUE. R.L. Ryall and R.B. Goldrick (Dept. of Clinical Sci., The John Curtin School of Med. Res., The Australian National Univ., Canberra, Australia 2600) *Lipids* 12, 272–7 (1977). The presence of glycerokinase has been demonstrated in human omental and subcutaneous adipose tissue. The enzyme reaction showed a linear time course for 5 min at 30 C and pH optima at pH 7.6 and 9.0. Saturation of the enzyme was observed at 1.8 mM adenosine triphosphate (ATP) and the double reciprocal plot of activity vs. ATP concentration was nonlinear giving two apparent Km values of 0.094 and 0.518 mM. The apparent Km values for glycerol, 0.112 mM, was obtained from a linear double reciprocal plot, and the enzyme was saturated at about 0.4 mM glycerol. The activity of glycerokinase in human adipose tissue excised under general anaesthesia was low and was unrelated to adipose cell size or the degree of obesity of the subject from whom the fat was obtained.

SIMPLE ASSAY FOR GLYCEROPHOSPHOLIPID HYDROLASE ACTIVITY OF POSTHEPARIN PLASMA. W.A. Shaw and W.R. Harlan (Avanti Biochem., Inc., Birmingham, Ala. 35226) *J. Lipid Res.* 18, 123–7 (1977). An assay using radioactive substrates is described that permits rapid determinations of glycerophospholipid-hydrolyzing activity in postheparin plasma or its fractions. Optimal conditions are described for hydrolysis of phosphatidylcholine and phosphatidylethanolamine. A minimum of only 2 μl of normal postheparin plasma is used and no extraction of the reaction products is required before their separation by thin-layer chromatography. We found that after an optimal heparin dose of 60 units/kg body weight the rate of hydrolysis for diacyl glycerophosphocholine and diacyl glycerophosphoethanolamine is 1.16 μmoles/ml per hr and 22.4 μmoles/ml per hr, respectively.

ENERGY UTILIZATION OF A LOW CARBOHYDRATE DIET FED GENETICALLY OBESE RATS AND MICE. S.W. Thenen and J. Mayer

(Dept. of Nutr., Harvard School of Public Health, Boston, Mass. 02115) *J. Nutr.* **107,** 320–9 (1977). Genetically obese Zucker rats, ob/ob mice and non-obese littermates were fed low carbohydrate (2%, 48%, and 50% of energy as carbohydrate, protein, and fat, respectively) and control (60%, 19%, and 21%, as carbohydrate, protein, and fat) diets. The oxidation of the energy components of these diets was measured by adding D-[U-^{14}C]glucose, L-[U-^{14}C]glutamic acid, and glyceryl tri-[1-^{14}C]oleate to test meals given intragastrically and collecting respiratory CO_2 for 4 hours. Obese Zucker rats oxidized less fat than non-obese rats when fed both diets, while obese mice oxidized fat to the same extent as non-obese mice. Feeding the low carbohydrate diet significantly increased body weight in the obese mice, but not in obese rats and non-obese mice and rats. The effects of obesity and the low carbohydrate diet on food intake, serum glucose and lipid values and CO_2 production are also reported.

13-CIS-RETINOIC ACID: INHIBITION OF BLADDER CARCINOGENESIS IN THE RAT. M.B. Sporn, R.A. Squire, C.C. Brown, J.M. Smith, M.L. Wenk and S. Springer (Nat'l. Cancer Inst., Bethesda, Md. 20014) *Science* **195,** 487–9 (1977). Transitional cell and squamous cell cancer of the bladder was induced in Wistar/Lewis female rats by direct instillation of *N*-methyl-*N*-nitrosourea into the bladder. Feeding of the synthetic retinoid, 13-*cis*-retinoic acid, inhibited the incidence and extent of bladder cancer in these rats, even when 13-*cis*-retinoic acid administration was begun after completion of the carcinogen treatment.

SERUM LIPOPROTEIN RESPONSES TO EXOGENOUS CHOLESTEROL IN SPIDER MONKEYS: EFFECT OF LEVELS OF DIETARY PROTEIN. S.R. Srinivasan, B. Radhakrishnamurthy, E.R. Dalferes, L.S. Webber and G.S. Berenson (Dept. of Med., Louisiana State Univ. Med. Ctr., New Orleans, La. 70112) *Proc. Soc. Exp. Biol. Med.* **154,** 102–6 (1977). The interacting effects of high (25%), low (8%), and very low (4%) dietary protein on the serum lipoprotein concentrations have been studied in spider monkeys (*Ateles* Sp.). Although the level of dietary protein per se had no appreciable main effect on the basal levels of serum total cholesterol, very low protein diet produced significant elevation in α-lipoprotein cholesterol. The serum lipoprotein responses to exogenous cholesterol varied at different levels of protein intake, the degree of response being highest at 4% protein level and lowest at 8% protein level. The changes were observed mainly in β- and α-lipoproteins.

STEROL COMPOSITION AND PHYTOSTEROL UTILIZATION AND METABOLISM IN THE MILKWEED BUG. J.A. Svoboda, S.R. Dutky, W.E. Robbins, and J.N. Kaplanis (Insect Physiol. Lab., Plant Protection Inst., ARS, USDA, Beltsville, Md. 20705) *Lipids* 12, 318–21 (1977). Analysis of the sterols of the milkweed bug, *Oncopeltus fasciatus* (Dallas) and dietary sunflowerseeds revealed that there is little, if any, conversion of dietary C_{28} or C_{29} phytosterols to cholesterol in this phytophagous insect. The dietary sterols are apparently utilized with little alteration both during development to the adult stage and egg production, and cholesterol comprises $< 1\%$ of the sterols in either adult males and females or in the eggs. The significance of these findings are discussed in light of the recent discovery that the C_{28}-ecdysone, makisterone A, is the predominant molting hormone in the enbryonated egg of the milkweed bug.

ANALYSIS OF RAT SERUM APOLIPOPROTEINS BY ISOELECTRIC FOCUSING. II. STUDIES ON THE LOW MOLECULAR WEIGHT SUBUNITS. J.B. Swaney and L.I. Gidez (Depts. of Biochem. and Med., Albert Einstein Col. of Med., Yeshiva Univ., Bronx, N.Y. 10461) *J. Lipid Res.* **18,** 69–76 (1977). The low molecular weight proteins of rat apo HDL and apo VLDL have been isolated and analyzed by the technique of isoelectric focusing. Sephadex fractions from apo HDL (HS-3) and apo VLDL (VS-3) that contain these proteins reveal three major bands with apparent isoelectric points of pH 4.50, 4.67, and 4.74, as well as three minor bands at pH 4.43, 4.57, and 4.61. In addition, apo HDL has a major band at pI of 4.83. DEAE-Cellulose chromatography was used to prepare purified fractions of these components that were characterized by *N*-terminal analyses and molecular weight determinations by SDS gel electrophoresis. The major low molecular weight components of apo HDL were focused on a slab gel and the bands were identified as A-II (pI 4.93), C-II (pI 4.74), C-III-0 (pI 4.67), and C-III-3 (pI 4.50). Neuraminidase treatment of apo HDL, followed by isoelectric focusing, suggested that the other bands, which have not previously been reported, may be additional forms of the C-III protein, differing only in their content of sialic acid.

ABSENCE OF THE BIOCHEMICAL SYMPTOMS OF ESSENTIAL FATTY ACID DEFICIENCY IN SURGICAL PATIENTS UNDERGOING PROTEIN SPARING THERAPY. L.D. Stegink, J.B. Freeman, J. Wispe and W.E. Connor (Depts. of Biochem., Pediatrics, Surgery and Internal Med., Univ. of Iowa College of Med., Iowa City, Iowa 52242) *Am. J. Clin. Nutr.* **30,** 388–93 (1977). The biochemical symptoms of essential fatty acid deficiency appear within 7 to 10 days of fat-free total parenteral nutrition using glucose-amino acid mixtures. The linoleic acid (C18:2W6) content of all plasma lipid fractions decreases greatly and plasma eicosatrienoic acid (C20:3W9) increases. We have measured the fatty acid composition of the plasma lipid fractions in six surgical patients receiving parenteral nutritional solutions containing only amino acids, and completely free of glucose, before and after 10 to 14 days of such therapy. Biochemical symptoms of essential fatty acid deficiency did not occur. The fatty acid composition of all plasma lipid fractions remained unchanged after this time period. Mobilization of linoleic acid from the adipose tissue occurred with this treatment in contrast to the inhibition of lipolysis of adipose tissue triglyceride produced by continuous infusions of hypertonic glucose-amino acid mixtures.

EFFECT OF CHOLESTEROL FEEDING ON TISSUE LIPID PEROXIDATION, GLUTATHIONE PEROXIDASE ACTIVITY AND LIVER MICROSOMAL FUNCTIONS IN RATS AND GUINEA PIGS. A.C. Tsai, G.M. Thie and C.R.-S. Lin (Human Nutr. Program, School of Public Health, Univ. of Mich., Ann Arbor, Mich. 48109) *J. Nutr.* **107,** 310–9 (1977). The effect of cholesterol feeding on liver and aortic nonenzymatic lipid peroxidation and glutathione peroxidase activities, and on liver microsomal NADPH-dependent lipid peroxidation, codeine hydroxylation and cytochrome P-450 levels was examined in rats and guinea pigs. One percent cholesterol was added to a casein-sucrose-soybean oil basal diet for rats or a stock diet with 2% soybean oil for guinea pigs. The effect of vitamin E and cholestyramine was also examined in some experiments. Cholesterol feeding increased the rate of lipid peroxidation in liver and aortic homogenate both in rats and guinea pigs when fed nonvitamin E supplemented basal diets. Vitamin E supplementation prevented the increase in the aorta, but not as completely in the liver in rats, while the reverse was true in guinea pigs. Vitamin E supplementation increased liver and serum cholesterol levels in guinea pigs, but had no such effect in rats. Results of this study indicate that cholesterol feeding can result in various metabolic alterations in rats and guinea pigs. The implication of these alterations in atherogenesis requires further investigations.

EVIDENCE FOR THE PREFERENTIAL INTERACTION OF GLYCOPHORIN WITH NEGATIVELY CHARGED PHOSPHOLIPIDS. E.J.J. van Zoelen, R.F.A. Zwaal, F.A.M. Reuvers, R.A. Demel and L.L.M. Van Deenen (Lab. of Biochem., State Univ. of Utrecht, Transitorium III, Padualaan 8, De Uithof, Utrecht, The Netherlands) *Biochim. Biophys. Acta* **464,** 482–92 (1977). Glycophorin, extracted from the erythrocyte membrane after treatment with lithium-diiodo-salicylate, still contains a significant amount of phospholipid, consisting predominantly of phosphatidylserine. Methods are described which lead to a full delipidation of the protein. After treatment with neuraminidase, delipidated glycophorin shows a preferential interaction with monolayers of negatively-charged phospholipids. This lipid-protein interaction is decreased by the presence of cholesterol *in* the lipid film.

CHARACTERIZATION OF ANTIBODY TO HUMAN PHOSPHATIDYLCHOLINE:CHOLESTEROL ACYLTRANSFERASE. K.G. Varma, A.H. Nowotny and L.A. Soloff (Lipid Research Lab., Dept. of Med. and Dept. of Microbiol. and Immunology, Temple Univ. Health Sci. Center, Philadelphia, Pa. 19140 U.S.A.) *Biochim. Biophys. Acta* **486,** 378–84 (1977). Purified preparations of phosphatidylcholine(lecithin):cholesterol acyltransferase (EC 2.3.1.43) were injected into goats to produce antisera reacting with this enzyme. The antisera and the γ-globulin derived therefrom were examined by the technics of immunodiffusion, immunoelectrophoresis and immunoinhibition of the enzyme. The antisera gave no precipitation lines with human high density lipoproteins (HDL) and human low density lipoproteins (LDL). A weak antibody titer towards human serum albumin was noted only after prolonged immunization. The enzymatically active band isolated from acrylamide gels gave a single arc in immunodiffusion and immunoelectrophoresis. The γ-globulin derived from the antisera inhibited human phosphatidylcholine: cholesterol acyltransferase activity.

LIPOSOME-CELL INTERACTION: TRANSFER AND INTRACELLULAR

RELEASE OF A TRAPPED FLUORESCENT MARKER. J.N. Weinstein, S. Yoshikami, P. Henkart, R. Blumenthal and W.A. Hagins (Lab. of Theroretical Biol., Nat'l. Cancer Inst., Nat'l. Insts. of Health, Bethesda, Md. 20014) *Science* **195**, 489–92 (1977). When small, unilamellar lipid vesicles containing a high concentration of the fluorescent dye 6-carboxyfluorescein are incubated with either frog retinas or human lymphocytes, fluorescence distributes widely throughout each cell. Since "self-quenching" largely prevents the dye from fluorescing as long as it remains sequestered in vesicles, it is clear that a considerable amount of dye is released from the vesicles and diluted into the much larger volume of the cell.

DIMETHYLHYDRAZINE-INDUCED COLON TUMORS IN RATS FED DIETS CONTAINING BEEF FAT OR CORN OIL WITH AND WITHOUT WHEAT BRAN. R.B. Wilson, D.P. Hutcheson and L. Wideman (Dept. of Vet. Microbiol. and Pathol., Washington State Univ., Pullman, Wash. 99163) *Am. J. Clin. Nutr.* **30**, 176–81 (1977). Male rats of the Sprague-Dawley strain were fed semisynthetic diets containing either 20% beef fat or corn oil with and without 20% wheat bran. Half of the animals received four weekly doses and the other half received eight weekly doses of dimethylhydrazine, 30 mg/kg, by intragastric intubation. The percentage of rats with tumors of the colon of all types was significantly higher in animals fed no bran than in those fed bran. Likewise, the percentage of rats with polypoid neoplasms of the colon was higher in rats fed no bran, but there was no significant difference in the percentage of rats with malignant tumors of the colon with respect to the feeding of bran. No significant differences were found between rats fed corn oil and those fed beef fat with respect to either the incidence or the kinds of colon tumors. Malignant tumors of the colon, causing death, occurred earlier in rats fed corn oil as compared to those fed beef fat. The percentage of rats with tumors of the colon and the numbers of tumors per tumor-bearing rat were significantly increased in rats given eight doses of dimethylhydrazine or compared to those given four.

INCORPORATION OF DIETATY CIS AND TRANS ISOMERS OF OCTADECENOATE IN LIPID CLASSES OF LIVER AND HEPATOMA. R. Wood, F. Chumbler and R. Wiegand (Div. of Gastroenterol., Depts. of Med. and Biochem., Univ. of Missouri School of Med., Columbia, Mo 65201) *J. Biol. Chem.* **252**, 1965–70 (1977). Groups of rats bearing Morris minimal deviation hepatoma 7288CTC were fed a fat-free diet supplemented with either 0.5% safflower oil (diet A), 15% safflower oil or free fatty acids (diets B and C), or 15% safflower oil or free safflower fatty acids (diet D) for 4 weeks. A group of normal rats was also fed diet D. The positional isomers of the *trans*-octadecenoate fractions from liver and hepatoma triglycerides and cholesteryl esters exhibited the same approximate distribution as the *trans* fatty acids of diet D. In contrast, the 10-*trans*-octadecenoate, like 10-*cis*-octadecenoate, was almost excluded from the phospholipids of liver and plasma. Unlike liver, the hepatoma phospholipids contained 10-*trans*-octadecenoate at approximately half the percentage of neutral lipids.

ELECTRON IMPACT INDUCED FRAGMENTATION OF CHOLESTEROL AND RELATED C-5 UNSATURATED STEROIDS. S.G. Wyllie and B.A. Anos and L. Tokes (Chem. Dept., Hawkesbury Agr. College, Richmond, New South Wales 2753) *J. Org. Chem.* **42**, 725–32 (1977). Comparison of the mass spectra of cholest-5-ene and various C-5 unsaturated 3β-hydroxy sterols indicates that the fragmentations leading to the characteristic $(M-85)^+$ and $(M-111)^+$ ions in these sterols are triggered solely by the double bond. With the aid of 11 deuterium labeled analogues all diagnostic cleavages of this biologically important class of compounds have now been identified. Both $(M-85)^+$ and $(M-111)^+$ ions of cholesterol are due to very complex fragmentations involving the loss of ring A and part of ring B by cleavages of the 1–10, 5–10 and 5–6 or 7–8 bonds, respectively, with a hydrogen transfer mainly from C-6 in the $(M-85)^+$ ion. Fragmentations of such complexity can be revealed only with the aid of isotope labeling. The mechanisms of these fragmentations and the syntheses of the deuterium labeled compounds are discussed.

SERUM AND LIVER LIPID RESPONSES TO 3-HYDROXY-3-METHYLGLUTARIC ACID IN RATS ON DIFFERENT CARBOHYDRATE DIETS. S.Y.K. Yousufzai and M. Siddiqi (Biochem. Division, Dept. of Chem., Aligarh Muslim Univ., Aligarh 202001, India) *Lipids* **12**, 267–71 (1977). Groups of male adult albino rats were administered 3-hydroxy-3-methylglutaric acid (HMG) intraperitoneally along with six diets differing only in the type of carbohydrate used. Groups not treated with HMG served as controls. HMG showed a significant cholesterol- and triglyceride-lowering effect in the whole serum, serum β-lipoproteins, and liver of animals on all types of dietary carbohydrates. The effect was more marked in glucose, fructose, sucrose, and lactose. The phospholipid levels in whole serum, serum β-lipoproteins, as well as in liver were also significantly lowered on all types of carbohydrates except dextrin and starch fed animals, where it had no effect on liver levels. The lipid-lowering effect of HMG seems to be independent of the type of carbohydrate in diet.

TISSUE LIPID RESPONSES TO 3-HYDROXY-3-METHYLGLUTARIC ACID WITH DIFFERENT DIETARY FATS. S.Y.K. Yousufzai and M. Sissiqi (Biochem. Division, Dept. of Chem., Aligarh Muslim Univ., Aligarh 202001, India) *Lipids* **12**, 258–61 (1977). Simultaneous administration of 3-hydroxy-3-methylglutaric acid (HMG) for 4 weeks to rats fed 20% saturated fats prevented rise of serum cholesterol, triglycerides, and phospholipids. Except phospholipids, other liver lipids were significantly decreased. HMG administration for 4 weeks along with atherogenic diet significantly decreased cholesterol and phospholipids of serum, liver, aorta, and heart. The phospholipids of epididymal fat and brain were also significantly lowered. The triglyceride levels in serum, liver, and epididymal fat were significantly decreased. The maximal hypolipidemic effect of HMG was observed in serum.

ACTIVITY OF PHOSPHOLIPID-SYNTHESIZING ENZYMES IN RAT LIVER PLASMA MEMBRANES AND THE SOURCE OF BILIARY LECITHIN. I.M. Yousef, M.M. Fisher, J. Piekarski and B.J. Holub (Depts. of Pathol. and Med., Univ. of Toronto, Toronto, Ontario, Canada) *Lipids* **12**, 140–4 (1977). The potential for the synthesis of phosphatidylcholine by the bile canalicular membrane of the liver cell was assessed by measuring the activity of a number of phospholipid synthesizing enzymes in isolated bile canalicular membrane fractions from rat liver. The activity of these various enzymes was compared to that present in noncanalicular liver cell plasma membranes and in microsomes. The CDP-choline:1,2-diacyl-*sn*-glycerol-cholinephosphotransferase was virtually absent from the bile canalicular membranes but the specific activities of S-adenosyl-L-methionine:phosphatidylethanolamine N-methyltransferase and acyl-CoA:1-acyl-*sn*-glycero-3-phosphorylcholine acyltransferase were 11–15% of those found in the microsomes. The bile canalicular membranes also contained detectable acyl-CoA:*sn*-glycero-3-phosphate acyltransferase activity and the ability to potentiate the Ca^{++}-stimulated exchange of bases between different phospholipids. These findings indicate that the bile canalicular membranes have a very limited capacity for the formation of phosphatidylcholine under the assay conditions employed.

ALTERATIONS OF PROSTAGLANDIN E_2-9-KETOREDUCTASE ACTIVITY IN PROLIFERATING SKIN. V.A. Ziboh, J.T. Lord and N.S. Penneys (Depts. of Dermatol. and Biochem., Univ. of Miami School of Med., Miami, Fla. 33136) *J. Lipid Res.* **18**, 37–43 (1977). The activity of an NADPH-dependent PGE_2-9-ketoreductase has been demonstrated in rat and human skin. This activity is localized in the high speed supernatant fraction, indicating the presence of an active PGE_2-9-ketoreductase associated with the cytoplasmic fraction of the skin. Transformation of PGE_2 into $PGF_{2\alpha}$ is enhanced by skin specimens from psoriatic plaques and EFA-deficient rats, both characterized by excessive cellular proliferation and increased NADPH production. Incubations of the 105,000 *g* supernatant fractions from normal and EFA-deficient rats demonstrated that the activity of the PGE_2-9-ketoreductase was elevated in high speed preparations from EFA-deficient rats. Results from these studies suggest that the increased activity of PGE_2-9-ketoreductase observed in skin from human psoriatic plaques and EFA-deficient rats may be due in part to the increased generation of NADPH by these tissues and in part to alteration of the PGE_2-9-ketoreductase by the excessive proliferation of the tissues.

A STUDY ON THE PROTEOLYSIS OF FAT GLOBULE MEMBRANE PROTEINS ISOLATED FROM BUFFALO MILK. A.K. Bandyopadhyay and N.C. Ganguli (National Dairy Research Institute, Karnal) *J. Food Sci Technol.* **13**, 205 (1976). Fat globule membrane proteins (FGMP) of buffalo milk exhibited time and concentration dependent hydrolysis by trypsin. Except protein components V and VI all other components were almost completely cleaved at higher concentration of trypsin. Glycoproteins were relatively more resistant to proteolysis than other protein components. Proteins of ultracentrifugally classified membrane pellets exhibited similar behaviour towards proteol-

ysis indicating apparent similarities between these fractions. The rate of proteolysis was higher in FGMP from cow milk than from buffalo milk. However, buffalo milk FGMP and its fractions showed similarities in the rate of proteolysis. Sterilization of milk retarded proteolysis of FGMP, whereas chilling accelerated such enzymatic cleavage.

COLORIMETRIC METHOD FOR DETERMINING TOTAL LIPIDS IN HUMAN FLUIDS. C.S. Frings, T.W. Fendley and R.T. Dunn (Damon Corp.). *U.S. 4,012,196.* A process for determining total lipid concentration in an animal fluid containing at least 1 mg lipid per ml comprises reacting a sample of the animal fluid with sulfuric acid in a container and adding directly to the container a phospho-vanillin reagent to form a chromogen. The light absorbence of the chromogen is then measured and the concentration of total lipids determined. The mole ratios of vanillin to phosphoric acid and phosphoric acid to sulfuric acid are such as to allow direct addition of the phospho-vanillin reagent, thus eliminating the necessity of transfering a specific volume of the sulfuric acid-animal fluid mixture to a second container prior to addition of the chromogen.

EGG SUBSTITUTES: USE AND PREFERENCE—WITH AND WITHOUT NUTRITIONAL INFORMATION. J. Ostrander, C. Martinsen, J. McCullough, and M. Childs (School of Home Economics, Univ. of Washington, Seattle). *J. Am. Diet. Assoc.* 70, 267–9 (1977). Three egg substitutes, plus fresh and dried whole eggs, were evaluated by a consumer panel before and after receiving information on cost and caloric and cholesterol contents of the products, and before and after sensory evaluations. Initially, most of the thirty panelists believed that the egg substitutes would cost the same or less than fresh eggs. After receiving pertinent information, the panelists perceived no significant differences in the nutritional value of the products. After tasting, the consumers judged the nutritional value of all products higher, but their preferences were lower for all except fresh whole eggs. Presentation of information on the caloric and cholesterol contents of the egg substitutes without explanation of possible benefits in current health programs appeared to be insufficient to change consumers' perceptions of the fat-modified products.

TRANSBILAYER DISTRIBUTION AND MOVEMENT OF LYSOPHOSPHATIDYLCHOLINE IN LIPOSOMAL MEMBRANES. A.M.H.P. Van Den Besselaar, H. Van Den Bosch and L.L.M. Van Deenen (Biochem. Lab., Padualaan 8, De Uithof, Utrecht, The Netherlands) *Biochim. Biophys. Acta* 465, 454–65 (1977). Single bilayer vesicles were prepared by sonication of 5 mol% 1-palmitoyl lysophosphatidylcholine and 95 mol% egg phosphatidylcholine. Incubation with lysophospholipase results in a fast hydrolysis of 80–90% of lysophosphatidylcholine. The remaining lysophosphatidylcholine is only very slowly hydrolysed. These results are interpreted as lysophosphatidylcholine being asymmetrically distributed over the two halves of the bilayer. The slow phase of lysophosphatidylcholine hydrolysis sets an upper limit to the rate of transbilayer movement of lysophosphatidylcholine. The half time of this process at 37°C is estimated to be about 100 h. In handshaken liposomes consisting of 5 mol% 1-palmitoyl lysophosphatidylcholine and 95 mol% egg phosphatidylcholine 15–20% of lysophosphatidylcholine is readily available for exogenous lysophospholipase.

SPIN-LABEL STUDIES ON THE AQUEOUS REGIONS OF PHOSPHOLIPID MULTILAYERS. A.D. Deith, W. Snipes, and D. Chapman (Biochem. and Biophys. Dept., Pennsylvania State Univ., University Park, Penn.) *Biochemistry* 16, 634–41 (1977). Water-soluble spin labels were used to study dimyristoyllecithin (DML) phospholipid multilayers. Previous studies report that there is a "bound" water region associated with dimyristoyllecithin containing about 10 molecules of water per phospholipid, a "trapped" water region located between the lamellae containing approximately 11 molecules per phospholipid, and a "free" water region external to the lamellae. The results of this investigation show that certain water-soluble spin-label molecules have their motional properties differentially modified by these three water environments. Furthermore, the labels also reveal the onset of lipid-phase transitions even though they have high water solubility. A phosphate-containing spin label demonstrated strong anistropic motion in the lipid-water system above the phase transition but not below. The resulting interaction was different at the two pH values. These water-soluble spin labels mimic ionic or nonionic solutes. Upon freezing, the spin labels are shown to be expelled from the ice regions into the remaining aqueous regions. The usefulness of this approach in studying solute behavior when freezing occurs and potential studies involving aqueous regions of cytoplasm are considered.

SYNTHESIS AND FUNCTION OF 9,12,15-OCTADECATRIEN-6-YNOIC ACID IN THE MOSS CERATODON PURPUREUS. J.L. Gellerman, W.H. Anderson and H. Schlenk. (The Hormel Inst., Univ. of Minnesota, Austin, Minn.) *Biochemistry* 16, 1258–62 (1977). Biosynthesis of *all-cis*-9,12,15-octadecatrien-6-ynoic acid in the moss, *Ceratodon purpureus,* was studied using protonemata cultures with labeled 9,12,15-octadecatrienoic (linolenic) and 6,9,12,15-octadecatetraenoic acids as substrates. Both acids were efficiently converted into the acetylenic and into 5,8,11, 14,17-eicosapentaenoic acids. Accordingly, the introduction of a triple bond in position 6 of linolenic acid involves formation of a double bond as a discrete step. Acetylenic acid triglycerides are reserve lipids in the moss. Under suitable growth conditions the acetylenic acids are catabolized and partly reused via acetate for de novo synthesis of fatty acids. They are not used for more direct syntheses of the common polyunsaturated fatty acids.

QUALITY EVALUATION OF FROZEN STORED CHANNEL CATFISH GROWN BY TANK CULTURE: EFFECTS OF DIETARY FAT, FREEZING METHOD AND STORAGE TEMPERATURE. T.A. Gibson, R.E. Worthington, E.K. Heaton and L.R. Beuchat (Dept. of Food Sci., Univ. of Georgia College of Agr. Experiment Stations, Experiment, GA) *J. Food Sci.* 42, 352–4 (1977). Channel catfish *(Ictalurus punctatus)* were grown in tank culture on diets containing either tallow or menhaden oil as 10% of diet. Processed fish were frozen in air at −18°C or in nitrogen spray at −77°C, sealed in polyethylene and stored at either −18 or −35°C. Sensory evaluations of fresh fish revealed no differences in flavor, texture, or aroma. Fish reared on tallow were rated slightly superior for flavor after 94 days storage and for aroma and flavor after 312 days. Fish frozen in air were rated slightly superior for aroma after 94 days and for texture and flavor after 312 days. Fish stored at −18°C were rated slightly superior in texture after 312 days. Initial microbial counts were approx 900 per 10 cm^2 of flesh surface; these values decreased by 65% after storage for 102 days and by 95% or more after storage for 365 days.

LIPID CHANGES IN FROZEN STORED CHANNEL CATFISH GROWN BY TANK CULTURE: EFFECTS OF DIETARY FAT, FREEZING METHOD, AND STORAGE TEMPERATURE. T.A. Gibson and R.E. Worthington (Dept. of Food Sci., Univ. of Georgia College of Agr. Experiment Stations, Experiment, GA) *J. Food Sci.* 42, 355–8 (1977). Channel catfish *(Ictalurus punctatus)* were grown in tank culture on diets containing either tallow or menhaden oil at 10% of diet. Processed fish were frozen in air at −18°C or in nitrogen spray at −77°C, sealed in polyethylene, and stored at either −18 or −35°C. Levels and fatty acid profiles were determined for total lipid, triglyceride, phospholipid, and free fatty acids in fresh fish that had been held on ice for 2 days and in frozen fish at 0, 94, 220, and 312 days. Dietary lipids strongly affected fatty acid profiles of all lipid fractions. Free fatty acid production occurred only in fish held on ice and in fish stored at −18°C. Changes in free fatty acid profiles and decreases in phospholipid levels indicated phospholipase activity. After 312 days storage at −18°C free fatty acid levels reached approx 0.93%. Measurable oxidative deterioration did not occur under either condition of frozen storage.

NATURE OF INTERACTION OF DEXTRAN SULFATE WITH LECITHIN DISPERSIONS AND LYSOLECITHIN MICELLES. Y.C. Kim and T. Nishida (Burnsides Res. Lab., Dept. of Food Sci., Univ. of Illinois, Urbana, Ill. 61801) *J. Biol. Chem.* 252, 1243–9 (1977). Lecithin and lecithin/cholesterol dispersions as well as lysolecithin micelles were used to provide basic information on the mechanism of the interaction of zwitterionic phospholipids with dextran sulfate. The addition of dextran sulfate to lecithin dispersions or lysolecithin micelles in the presence of Ca^{2+} produced insoluble complexes. The conversion of lecithin dispersions into insoluble complexes was very effective even at low Ca^{2+} concentrations. Approximately 70% of the lecithin was converted to the insoluble complex at $CaCl_2$ concentrations as low as 0.5 mM and the complete conversion was observed at $CaCl_2$ concentrations above 2.5 mM. It appears that the formation of the insoluble complex of lecithin or lysolecithin with dextran sulfate represents the mutually enhancing interactions involving both positive and negative charges of the zwitterionic phospholipids. These are the direct electrostatic interaction between the phospholipid choline nitrogen and the sulfate groups of dextran sulfate and the calcium cross-linking of the phosphate groups to the sulfate groups or to the phosphate groups of neighboring phospholipids.

ARRANGEMENT OF PHOSPHATIDYLSERINE AND PHOSPHATIDYLETHANOLAMINE IN THE ERYTHROCYTE MEMBRANE. G.V. Marinetti (Dept. of Biochem., Univ. of Rochester School of Med. and Dentistry, Rochester, N.Y. 14642) *Biochim. Biophys. Acta* **465**, 198–209 (1977). Cross-linking of phosphatidylethanolamine and phosphatidylserine in the erythrocyte membrane with the reagent difluorodinitrobenzene was studied as a function of temperature, time and concentration of difluorodinitrobenzene. The optimal extent of cross-linking of phosphatidylethanolamine to phosphatidylethanolamine, phosphatidylethanolamine to phosphatidylserine and phosphatidylserine to phosphatidylserine was expressed as molar ratios of these three different cross-linked species. The experimental results were compared to different models of a phospholipid monolayer containing phosphatidylethanolamine and phosphatidylserine in which phosphatidylserine was arranged primarily as singles (having 6 phosphatidylethanolamine neighbors) as clusters of dimers, trimer and tetramers or as large clusters. In the various model monolayers each lipid component has 6 neighbors. The models which are consistent with the experimental results are those in which phosphatidylserine and phosphatidylethanolamine occur as small clusters in a non-random array.

COOPERATIVITY OF THE PHASE TRANSITION IN SINGLE- AND MULTIBILAYER LIPID VESICLES. D. Marsh, A. Watts and P.F. Knowles (Max-Planck-Inst. fur biophysikalische Chemie, D-3400 Gottingen, G.F.R.) *Biochim. Biophys. Acta* **465**, 500–14 (1977). The effect of membrane morphology on the cooperativity of the ordered-fluid, lipid phase transition has been investigated by comparing the transition widths in extended, multibilayer dispersions of dimyristoyl phosphatidylcholine, and also of dipalmitoyl phosphatidylcholine, with those in the small, single-bilayer vesicles obtained by sonication. The electron spin resonance spectra of three different spin-labelled probes, 2,2,6,6-tetramethylpiperdine-*N*-oxyl, phosphatidylcholine and stearic acid, and also 90° light scattering and optical turbidity measurements were used as indicators of the phase transition. In all cases the transition was broader in the single-bilayer vesicles than in the multibilayer dispersions, corresponding to a decreased cooperativity on going to the small vesicles. The implications for the effect of membrane structure and morphology on the cooperativity of phase transitions in biological membranes, and for the possibility of achieving lateral communication in the plane of the membrane, are discussed.

FAILURE OF BRAN TO ALTER DIET-INDUCED HYPERLIPIDEMIA IN THE RAT. C. Arvanitakis, C.L. Stamnes, J. Folscroft and P. Beyer (Dept. of Medicine, Division of Gastroenterology and Dept. of Nutr. and Dietetics, Kansas Univ. Schl. of Medicine, Kansas City, Kan.) *Proc. Soc. Exp. Biol. Med.* **154**, 550–2 (1977). The effect of bran on diet-induced hyperlipidemia was examined in a long-term 16-week study in the rat. Animals were divided into four groups: group I (control diet with cornstarch); group II (cornstarch with 10% wheat bran); group III (atherogenic diet with sucrose); and group IV (atherogenic diet with sucrose and 10% wheat bran). The addition of bran did not affect serum and liver lipids (cholesterol and triglycerides) in the control or the experimental groups on atherogenic diet.

LIPOPROTEIN-MEDIATED REGULATION OF 3-HYDROXY-3-METHYLGLUTARYL COENZYME A REDUCTASE ACTIVITY AND CHOLESTERYL ESTER METABOLISM IN THE ADRENAL GLAND OF THE RAT. S. Balasubramaniam, J.L. Goldstein, J.R. Faust, G.Y. Brunschede and M.S. Brown (Div. of Med. Genetics, Dept. of Internal Med., Univ. of Texas Health Sci. Ctr., Dallas, Tex. 75235) *J. Biol. Chem.* **252**, 1771–9 (1977). In the adrenal gland of the rat, the activity of 3-hydroxy-3-methylglutaryl coenzyme A reductase, the rate-controlling enzyme of cholesterol synthesis, is shown to be regulated by cholesterol carried in plasma lipoproteins. When plasma cholesterol levels were lowered 90% by administration of the drug 4-aminopyrazolopyrimidine, the cholesteryl ester content of the adrenal gland declined by more than 90%, and this was associated with a 150- to 200-fold increase in the activity of adrenal 3-hydroxy-3-methylglutaryl coenzyme A reductase and a 30-fold increase in cholesterol synthesis from [^{14}C]acetate. The current data indicate that the low rate of cholesterol synthesis normally observed in the rat adrenal gland is due to a suppression of the activity of 3-hydroxy-3-methylglutaryl coenzyme A reductase that is mediated by plasma lipoproteins.

ON CELL MEMBRANE LIPID FLUIDITY AND PLANT LECTIN AGGLUTINABILITY. A SPIN LABEL STUDY OF MOUSE ASCITES TUMOR CELLS. B.L. Bales, E.S. Lesin and S.B. Oppenheimer (Centro de Fisica, I.V.I.C., Apartado 1827, Caracas, Venezuela) *Biochim. Biophys. Acta* **465**, 400–7 (1977). The fluidity of the plasma membrane of Sarcoma 180 mouse ascites tumor cells has been studied in viable cells using fatty acid spin labels. The order parameter was found to vary from 0.61, approximately four carbon bond lengths removed from the membrane surface, to 0.47 approximately eleven bond lengths removed at 22°C and from 0.55 to 0.33 at 37°C. Thus these cells show similar membrane fluidity to that found in other mammalian cells with the exception of human erythrocytes which are less fluid. The concanavalin A mediated agglutinability of Sarcoma 180 cells was altered by the addition of cytochalasin B and the fluidity was found to be the same as in unaltered cells.

ISOLATION AND INCORPORATION INTO LIPID VESICLES OF A CONCANAVALIN A RECEPTOR FROM HUMAN ERYTHROCYTES. D.G. Barratt, F.J. Sharom, A.E. Thede and C.W.M. Grant (Dept. of Biochem., Univ. of Western Ontario, London, Ontario, N6A 5C1, Canada) *Biochim. Biophys. Acta* **465**, 191–7 (1977). Affinity chromatography has been used to isolate a concanavalin A receptor portion of Band 3 from human erythrocytes in the presence of the readily-dialysable detergent, dodecyltrimethylammonium bromide. Addition of phospholipids to the isolated fraction and removal of detergent by dialysis leads to formation of vesicles containing the receptor. Intramembranous particles similar in size and shape to those seen in intact erythrocytes are a characteristic of the reconstituted preparations. Vesicles containing receptor bind concanavalin A with high affinity.

FLUIDITY OF THE LIPIDS NEXT TO THE ACETYLCHOLINE RECEPTOR PROTEIN OF TORPEDO MEMBRANE FRAGMENTS. USE OF AMPHIPHILIC REVERSIBLE SPIN-LABELS. A. Bienvenue, A. Rousselet, G. Kato, and P.F. Devaux (Biophys. Moleculaire, GPS-ENS Tour 23, Univ. Paris VII, 75221 Paris Cedex 05, France) *Biochemistry* **16**, 841–8 (1977). Choline esters of spin-labeled fatty acids (long-chain acylcholines) were used to probe the hydrophobic environment of the acetylcholine receptor protein in membrane fragments from *Torpedo marmorata*. These spin-labels competitively inhibit the binding of [^{3}H]acetylcholine to the receptor site. Their inhibition constants (K_I) were close to 200 nM. At the high membrane concentration required for electron spin resonance (ESR) experiments, the apparent inhibition constants (K_I^{app}) differed from K_I determined by using dilute membrane concentration. This is due to the amphiphilic character of long-chain acylcholine. For most spin-labels used, only difference ESR spectroscopy provided reliable spectra corresponding to receptor-bound spin-labeled acylcholines. Short-range spin-spin interactions were created between spin-labels bound to the receptor site and spin-labels in a fluid phase. This indicates that lipids next to the receptor protein are not completely immobilized in spite of the semicrystalline organization of the proteins in the postsynaptic region.

RAT LIVER PROTEINS CAPABLE OF TRANSFERRING PHOSPHATIDYLETHANOLAMINE. PURIFICATION AND TRANSFER ACTIVITY FOR OTHER PHOSPHOLIPIDS AND CHOLESTEROL. B. Bloj and D.B. Zilversmit (Div. of Nutr. Sci. and Sec. of Biochem., Mole. and Cell Biol., Div. of Biol. Sci., Cornell Univ., Ithaca, N.Y. 14853) *J. Biol. Chem.* **252**, 1613–9 (1977). Two proteins, one in a highly purified form, have been isolated from the soluble fraction of rat liver homogenate. These proteins accelerate the transfer of labeled phosphatidylethanolamine, phosphatidylcholine, phosphatidylinositol, sphingomyelin, and cholesterol from liposomes to mitochondria or erythrocyte ghosts. The relative transfer activities toward the different phospholipids remain constant throughout the last three steps of the purification procedure in spite of the extensive change in the electrophoretic profile of the protein mixture. The cholesterol transfer activity remains unchanged after the final heat treatment as well. This indicates that all of the transfer activities are present in a single protein.

UTILIZATION OF LONG-CHAIN FREE FATTY ACIDS AND GLUCOSE BY HUMAN LEUKEMIC BLAST CELLS. C.P. Burns, I.R. Welshman and A.A. Spector (Depts. of Medicine and Biochem., Univ. of Iowa College of Medicine, Iowa City, 10) *Cancer Res.* **37**, 1323–7 (1977). We have studied the utilization of free fatty acid and glucose by human leukemic blast cells. Palmitate was both incorporated into complex cellular lipids, primarily phospholipids and triglycerides, and oxidized to CO_2. The predominant phospholipid synthesized was phosphatidylcholine. Only a small proportion of the incoming fatty acid was modified structurally before incorportation into lipid esters. After incubation with (1-^{14}C) palmitate, 91% of the radioactivity recovered in cell lipids remained in fatty acids con-

taining 16 carbon atoms. Studies with labeled glucose revealed little *de novo* synthesis of fatty acid, and the majority of the radioactivity from glucose was located in the water-soluble fraction after saponification of the esters. We conclude that the free fatty acids contained in the extracellular fluid provide much of the fatty acid for required cellular lipid synthesis in human leukemic blast cells. Since there is little elongation of incoming palmitate before incorporation into cellular lipids, it may be possible to alter the fatty acid composition of membrane phospholipids by changing the proportion of the various free fatty acids available to the leukemic cells.

INFLUENCE OF (−)-HYDROXYCITRATE ON LIPOGENESIS IN CHICKENS AND RATS. H. Chee, D.R. Romsos and G.A. Leveille (Food Sci. and Human Nutr. Dept., Michigan State Univ., East Lansing, Mich. 48824) *J. Nutr.* **107**, 112–9 (1977). The influence of (−)-hydroxycitrate on in vitro rates of fatty acid synthesis in chicken and rat liver and in rat adipose tissue was investigated. Following acute and chronic administration of (−)-hydroxycitrate to chickens and rats, changes in rates of fatty acid synthesis in vivo and in lipogenic enzyme activities were also determined. In vivo rates of fatty acid synthesis in rat adipose tissue were not influenced by consumption of a diet containing 52.6 mmoles (−)-hydroxycitrate/kg diet. Plasma triglyceride levels were increased two-fold in chickens, but unchanged in rats, fed (−)-hydroxycitrate for 2 to 3 weeks. There are species-specific as well as organ-specific responses to (−)-hydroxycitrate.

CHANGES IN RAT HEART PHOSPHOLIPID COMPOSITION AFTER RAPESEED OIL FEEDING. P. Dewailly, G. Sezille, A. Nouvelot, J.C. Fruchart and J. Jaillard (Lab. U.E.R. de Physiopathol. des Lipides, Inst. Pasteur, 59012 Lille Cedex, France) *Lipids* **12**, 301–6 (1977). The influence of long duration rapeseed oil feeding with high or low levels of erucic acid has been investigated on rat heart phospholipids. The rats treated for 20 wk with rapeseed oil containing 46.2% erucic acid showed a twofold increase in the sphingomyelin content of the heart. Treatment with primor rapeseed oil (3.7% erucic acid) for 20 wk did not modify phospholipid composition of rat heart. The fatty acid patterns of phosphatidylethanolamine and phosphatidylcholine were slightly influenced by the high erucic rapeseed oil; eicosenoic acid was incorporated preferentially into position one, but erucic acid showed a random distribution in both. As cardiolipin is localized in the inner membrane of mitochondria and sphingomyelin in plasma and microsomal membranes, the acyl-moiety alterations of both phospholipids might be correlated to the pathological lesions of rat heart after a long duration of rapeseed oil feeding.

EFFECTS OF BILE ACIDS, LECITHIN, AND MONOOLEIN ON AMINO ACID ABSORPTION FROM THE HUMAN DUODENUM. E.P. Dimagno, J.-R. Malagelada and V.L.W. Go (Gastroenterol. Unit, Mayo Clinic and Mayo Found., Rochester, Minn. 55901) *Proc. Soc. Exp. Biol. Med.* **154**, 325–30 (1977). The effects of bile acid, lecithin, and monoolein (MO) on essential amino acid (EAA) absorption from the human duodenum were determined. It was found that: methionine was absorbed at the most rapid rate while isoleucine, leucine, and valine were absorbed slightly less rapidly and lysine and threonine at the slowest rates; duodenal EAA absorption rates are very similar to previously reported rates for jejunal absorption of EAA; taurocholate (TC) and taurodeoxycholate significantly depressed EAA absorption; and although emulsified MO had no significant effect on EAA absorption, the addition of MO or lecithin to the EAA + 10 m*M* TC perfusion mixture restored EAA absorption to normal. How bile acids inhibit EAA absorption and how lecithin and MO abolish this inhibition are not known and have not been elucidated by our studies. However, it is postulated that when either lecithin or MO is present, less bile salt is available to act at the surface membranes of intestinal cells, disruption of cell membranes is prevented, and EAA absorption proceeds normally.

UPTAKE AND DEGRADATION OF CHOLESTEROL ESTER-LABELLED RAT PLASMA LIPOPROTEINS IN PURIFIED RAT HEPATOCYTES AND NONPARENCHYMAL LIVER CELLS. C.A. Drevon, R. Berg, and K.R. Norum (Inst. for Nutr. Res., Schl. of Medicine, Univ. of Oslo, Blindern, Oslo, Norway) *Biochim. Biophys. Acta* **487**, 122–36 (1977). 1. A new method for isolation and purification of rat liver hepatocytes and nonparenchymal cells by differential centrifugation is described. 2. Cholesterol ester-labelled lipoproteins (prepared by the action of lecithin: cholesterol acyltransferase) intravenously injected were taken up by hepatocytes and nonparenchymal cells. 3. Hepatocytes and nonparenchymal cells in suspension were able to take up and hydrolyse the cholesterol ester portion of lipoproteins. 4. Uptake of cholesterol ester labelled whole rat plasma and high density lipoproteins (HDL) increased with increasing concentrations until a distinct saturation level was reached in hepatocytes. In nonparenchymal cells there was no saturation of lipoprotein uptake. 5. Concanavalin A inhibited cholesterol ester-labelled lipoprotein uptake in hepatocytes, indicating that the uptake at least partially depends on carbohydrate sites on the cell surface. The uptake in nonparenchymal cells was unaffected of concanavalin A. 6. The specific activity of the acid cholesterol ester hydrolase was the same in homogenates from hepatocytes and nonparenchymal cells while acyl-CoA: cholesterol acyltransferase was found almost exclusively in hepatocytes.

THE INFLUENCE OF CRUDE COTTONSEED OIL IN THE FEED ON THE BLOOD AND EGG YOLK LIPOPROTEINS OF LAYING HENS. R.J. Evans, C.J. Flegal, C.A. Foerder, D.H. Bauer and M. La Vigne (Depts. of Biochem. and Poult. Sci., Mich. State Univ., East Lansing, Mich. 48824) *Poult. Sci.* **56**, 468–79 (1977). Lipovitellin, very low density lipoproteins (VLDL), low density lipoproteins (LDL), high density lipoproteins (HDL), and proteins of $d > 1.20$ were isolated from blood plasma and egg yolks obtained from hens fed a normal diet or one containing 2.5% of crude cottonseed oil. The amounts and compositions of each fraction were determined. Hen blood plasma and egg yolk VLDL and LDL obtained from hens fed a normal diet contained similar levels of lipid, and the fatty acid compositions of those lipids were, for the most part, similar. The percentages of VLDL and LDL in total lipoproteins were similar for plasma and egg yolk obtained from hens fed the normal diet. The increased content of stearic acid increased the density of the lipoproteins so that a larger proportion of the lipoproteins were in the LDL and a smaller proportion were in the VLDL than in lipoproteins from normal plasma and eggs.

LIPOPROTEIN LIPASE. ISOLATION AND CHARACTERIZATION OF A SECOND ENZYME SPECIES FROM POSTHEPARIN PLASMA. P.E. Feilding, V.G. Shore and C.J. Fielding (Cardiovascular Res. Inst. and Dept. of Physiology, Univ. of Calif., San Francisco, Calif.) *Biochemistry* **16**, 1896–900 (1977). A lipoprotein lipase species (mol wt 69, 250) has been isolated from rat postheparin plasma, which differs from the low-molecular-weight species previously characterized in its amino acid composition and hexosamine content, and in its lower affinity for triglyceride-rich lipoprotein substrates. However, both enzymes are activated by the same coprotein (C-terminal glutamic acid, apo-C-2) from human very low density lipoprotein and have a similar specificity for lipid esters. Neither purified enzyme is activated by heparin. Both are inhibited by molar sodium chloride. Both enzyme species can be recovered from the same plasma samples. The possible relationship of these proteins to the different functional lipoprotein lipase activities of muscle and adipose tissues is discussed.

REGULATION OF LIPOLYSIS AND CYCLIC AMP SYNTHESIS THROUGH ENERGY SUPPLY IN ISOLATED HUMAN FAT CELLS. Y. Giudicelli, R. Pecquery, D. Provin, B. Agli and R. Nordmann (Ser. de Biochim. de la Faculte de Med. de Paris-Quest, Lab. de Biochim. du Ctr. Hosp., 78303 Poissy, France) *Biochim. Biophys. Acta* **486**, 385–98 (1977). The effects of glucose and of various inhibitors of glycolysis or of oxidative phosphorylation on stimulated lipolysis and on intracellular cyclic AMP and ATP levels were investigated in isolated human fat cells. The glycolysis inhibitors, NaF and monoiodoacetate, inhibited epinephrine- or theophylline-stimulated lipolysis and parallely reduced the intracellular cyclic AMP and ATP levels; however, neither NaF nor monoiodoacetate significantly affected dibutyryl cyclic AMP-induced lipolysis. Removal of glucose from the medium also reduced the rate of epinephrine-stimulated lipolysis and the intracellular cyclic AMP and ATP levels but failed to modify the lipolytic activity of dibutyryl cyclic AMP. When glycolysis was almost fully inhibited, human fat cells were insensitive to epinephrine but remained fully responsive to dibutyryl cyclic AMP. These results, showing a relationship between ATP availability, cyclic AMP synthesis and lipolysis, suggest a different ATP requirement for cyclic AMP synthesis and triacylglycerol lipase activation, a difference which could explain why ATP issued from glucose breakdown appears to be a determinant factor for cyclic AMP synthesis, but not for triacylglycerol lipase activation in human fat cells.

STIMULATION OF HUMAN PLATELET GUANYLATE CYCLASE BY FATTY ACIDS. D.B. Glass, W. Frey, II, D.W. Carr and N.D.

Goldberg Dept. of Pharmacol., Univ. of Minnesota, Minneapolis, Minn. 55455) *J. Biol. Chem.* 252, 1279–85 (1977). Guanylate cyclase from human platelets was over 90% soluble, even when assayed in the presence of Triton X-100. A time-dependent increase in activity occurred when the enzyme was incubated at 37° and this spontaneous activation was prevented by dithiothreitol. Arachidonic acid stimulated the soluble enzyme activity approximately 2- to 3-fold. These data indicate that the structural determinant required for stimulation by fatty acids of soluble platelet guanylate cyclase is a 1,4,7-octatriene group with its first double bond in the ω6 position. This structural group is similar to the substrate specificity determinants of fatty acid cyclooxygenase, the first enzyme of prostaglandin synthetase complex. Kinetic studies showed that the stimulation of guanylate cyclase by arachidonic acid is primarily an effect on maximal velocity. Arachidonic acid did not alter the concentration of free Mn^{2+} required for optimal activity. It is concluded that the activity of the soluble form of guanylate cyclase in cell-free preparations of human platelets can be increased by a lipid-protein interaction involving specific polyunsaturated fatty acids.

FEEDING POLYUNSATURATED VEGETABLE OILS TO LACTATING COWS. H.K. Goering, T.R. Wrenn, L.F. Edmondson, J.R. Weyant, D.L. Wood, and J. Bitman (U.S. Dept. of Agr., ARS, Beltsville, MD and Philadelphia, PA) *J. Dairy Sci.* 60, 739–47 (1977). Holstein cows fed concentrate: hay diets also were fed for 14 days supplements of soybean oil protected from ruminal hydrogenation by encapsulation in a casein-formaldehyde matrix, cottonseed oil plus casein, or cottonseed oil protected with casein-formaldehyde. The supplements were fed at rates to give a linoleic acid (18:2) intake of 225 g/day. Yields of milk and milk protein were not affected by treatment. Milk 18:2 was not increased by the unprotected soybean oil or cottonseed oil but was increased by protected soybean and cottonseed oil from a control of 2.3 to 5.7% of total milk fat. Milk 18:0 and 18:1 also increased. Compensatory declines were observed in milk 16:0 and 14:0 acids. In fecal fatty acids during the treatment periods, percentage of 18:2 of the total fat decreased and 18:0 markedly increased. These results indicate hydrogenation of the dietary oils in the alimentary tract or a differential absorption. Fecal 16:0 and 14:0 decreased.

MODIFICATION OF FATTY ACID COMPOSITION OF RAT HEART LIPIDS BY FEEDING COD LIVER OIL. S. Gudbjarnason and G. Oskarsdottir (Dept. of Chem., Sci. Inst., Univ. of Iceland, Dinhaga 3, Reykjavik) *Biochim. Biophys. Acta* 487, 10–5 (1977). Modification of the fatty acid composition of cardiac phospholipids and neutral lipids was studied in rats fed a diet containing 10% cod liver oil. The results reflect the dynamic state of esterified fatty acids in neutral lipids and phospholipids of heart muscle. In cardiac neutral lipids there was a moderate but significant increase in exogenous fatty acids, 20:1(*n*-9), 22:1(*n*-11), 20:5 (*n*-3) and 22:6(*n*-3), in animals fed cod liver oil, and a relative decrease in endogenous fatty acids, 16:0, 18:2(*n*-6) and 20:4(*n*-6). Increased dietary availability of 22:6(*n*-3) resulted in a major increase in the content of this fatty acid in phospholipids and replacement of 18:2(*n*-6) and 20:4(*n*-6). The 22:6(*n*-3) was able to replace one third of 18:2(*n*-6): further increase in 22:6(*n*-3) was accompanied by a decrease in 18:0. An inverse relationship between (*n*-6) and (*n*-3) polyene fatty acids in cardiac phospholipids suggests a replacement of (*n*-6) fatty acids by (*n*-3) fatty acids.

STUDIES ON INACTIVATION OF PYRUVATE DEHYDROGENASE BY PALMITOYLCARNITINE OXIDATION IN ISOLATED RAT HEART MITOCHONDRIA. R.G. Hansford (Lab. of Mole. Aging, Gerontol. Res. Ctr., Nat'l. Inst. on Aging, Nat'l. Inst. of Health, Baltimore City Hosp., Baltimore, Md. 21224) *J. Biol. Chem.* 252, 1552–60 (1977). The oxidation of an optimal concentration of palmitoylcarnitine, buffered with bovine serum albumin, by isolated rat heart mitochondria was found to give rise to an inactivation of pyruvate dehydrogenase, provided that the concentration of pyruvate present in the mitochondrial incubation was less than 250 μM. The greatest degree of inactivation was found at the lowest pyruvate concentration used, 50 μM, and this concentration was adopted for further studies in which the rate of mitochondrial respiration was varied.

EXPERIMENTALLY INDUCED "FATTY LIVER SYNDROME" CONDITION IN LAYING HENS. R.H. Harms, D.A. Roland, Sr., and C.F. Simpson (Dept. of Poultry Sci., Florida Agr. Exp. Station, Gainesville, Fla. 32611) *Poult. Sci.* 56, 517–20 (1977). Two experiments were conducted with ages laying hens to determine the influence of feeding 5,000 p.p.m. of iodine as potassium iodine (KI) and/or injecting 12 mg. of estradiol upon fat accumulation in the liver and serum cholesterol levels. The KI was fed for 8 days before making liver and blood determinations, and the estradiol was injected 3 days prior to making the determinations. The feeding of KI or injection of estradiol resulted in significantly increased liver weight. When the two treatments were combined a further significant increase in liver weights was obtained. The percent of fat in the liver was significantly increased by the injection of estradiol. However the feeding of KI in the presence or absence of estradiol did not affect the percentage of fat in the liver. Neither of the treatments significantly affected the fatty acid composition of the liver fat. Feeding of KI or injection of estradiol significantly increased total serum cholesterol levels. When the two treatments were combined a further increase in serum cholesterol level was observed. Histological changes of the livers of hens treated with KI and estradiol were similar to those previously described for the "fatty liver syndrome."

METABOLISM OF GAMMA-LINOLENIC ACID IN ESSENTIAL FATTY ACID-DEFICIENT RATS. A.G. Hassam, J.P. Rivers and M.A. Crawford (Dept. of Biochem., Nuffield Inst. of Comparative Medicine, Zoological Society of London, Regent's Park, London, England) *J. Nutr.* 107, 519–24 (1977). Female rats were weaned and fed a semi-purified diet lacking in essential fatty acids. After 160 days, the deficient diet was supplemented with varying amounts of gamma-linolenic acid. Changes in body weight and feed efficiency were measured. Total liver phospholipid fatty acids were also analyzed. Supplementation with gamma-linolenic acid to the deficient diet for 7 days led to improvements in body weight and feed efficiency of the deficient rats. The liver phospholipid fatty acid composition returned to a normal pattern. There was a reduction of 5,8,11-eicosatrienoic acid and an increase in the arachidonic acid. Thus, there was a fall in the triene:tetraene ratio with increasing dietary supplementation of gamma-linolenic acid. The essential fatty acid potency, the minimum dietary requirement for this fatty acid, and the widely accepted levels of the minimum requirements of dietary essential fatty acids are discussed.

MANNOSYLTRANSFERASE ACTIVITY IN CALF PANCREAS MICROSOMES. FORMATION OF ^{14}C-LABELED LIPID-LINKED OLIGOSACCHARIDES FROM GDP-D-[^{14}C]MANNOSE AND PANCREATIC DOLICHYL β-D-[^{14}C]MANNOPYRANOSYL PHOSPHATE. A. Herscovics, A.M. Golovtchenko, C.D. Warren, B. Bugge and R.W. Jeanloz (Lab. for Carbohydrate Res., Depts. of Biol. Chem. and Med., Harvard Med. School at the Massachusetts Gen. Hosp., Boston, Mass. 02114) *J. Biol. Chem.* 252, 224–34 (1977). Calf pancreas microsomes incorporated radioactive D-mannose from GDP-D-[^{14}C]mannose into lipid-bound oligosaccharides extracted with chloroform/methanol/water (10/10/2.5, v/v). Several products, which probably differed in the size of the oligosaccharide moiety, were labeled. These could be partially resolved by thin layer chromatography and DEAE-cellulose chromatography. The labeled lipid-bound oligosaccharides were retained on DEAE-cellulose more strongly than synthetic dolichyl α-D-[^{14}C]mannopyranosyl phosphate. The carbohydrate composition of the lipid-bound oligosaccharides of microsomal membranes was investigated by gas-liquid chromatography and by reduction with sodium borotritide. A heterogeneous mixture of oligosaccharides containing *N*-acetyl-D-glucosamine, D-mannose, and D-glucose varying in proportions from approximately 1/2.5/0.5 to 1/5/1.5 was obtained with glucosamine at the reducing end. Acid treatment of the lipid-bound oligosaccharide fraction yielded dolichyl pyrophosphate, suggesting that at least some of the oligosaccharides were linked to dolichol throught a pyrophosphate group.

ADEQUATE RESPONSE OF PLASMA 1,25-DIHYDROXYVITAMIN D TO PARTURITION IN PARETIC (MILK FEVER) DAIRY COWS. R.L. Horst, J.A. Eisman, N.A. Jorgensen and H.F. DeLuca (Depts. of Biochem. and Dairy Sci., College of Agr. and Life Sci., Univ. of Wisconsin-Madison, Madison 53706) *Science* 196, 662–3 (1977). The concentration of 1,25-dihydroxyvitamin D was measured by means of a radioactive receptor assay in the plasma of cows during the period immediately prior to, during, and following parturition. Nonparetic cows showed initially a slight decrease in plasma 1,25-dihydroxyvitamin D which was followed by a significant increase during parturition and 2 days postpartum. The highest concentration achieved in the control or nonparetic cows was 100 picograms per milliliter. In the paretic animals the plasma 1,25-dihydroxyvitamin D concentration increased sharply during the day preceding calving and reached a maximum of 200 picograms per

milliliter at parturition. This level was maintained during the ensuing 2.5 days. These results demonstrate that parturient paresis cannot be the result of insufficient synthesis or secretion of 1,25-dihydroxyvitamin D.

THE EFFECTS OF ORIGINAL AND RANDOMIZED RAPESEED OILS CONTAINING HIGH OR VERY LOW LEVELS OF ERUCIC ACID ON CARDIAC LIPIDS AND MYOCARDIAL LESIONS IN RATS. S. Hung, T. Umemura, S. Yamashiro, S.J. Slinger and B.J. Holub (Dept. of Nutr., Univ. of Guelph, Guelph, Ontario, Canada) *Lipids* 12, 215–21 (1977). The nutritional status of the very low-erucate rapeseed oil, *Brassica napus* var. 'Tower,' was compared with that of the high-erucate oil, *Brassica napus* var. 'Target,' as well as with corn oil. The effect of randomization on the nutritional qualities of rapeseed oil was investigated as well. The feeding of diets containing the original and randomized 'Tower' oil or the original 'Target' oil, at the 20% level by weight, gave growth rates which were not significantly different from that for corn oil. However, the randomized 'Target' oil gave growth rates which were significantly less than all other groups. The growth results could not be explained simply on the basis of food consumption. A much higher incidence of focal myocardial necrosis was found in animals receiving high-erucate rapeseed oil relative to animals given the corn oil. The incidence in rats fed diets containing very low-erucate rapeseed oil was intermediate between these latter two extremes.

PROPERTIES OF RAT LIVER MICROSOMAL STEAROYL-COENZYME A DESATURASE. R. Jeffcoat, P.R. Brawn, R. Safford and A.T. James (Basic Studies, Biosci. Div., Unilever Res., Colworth House, Sharnbrook, Bedford MK441LQ, U.K.) *Biochem. J.* 161, 431–7 (1977). Rat liver microsomal stearoyl-CoA desaturase activity was shown to be stimulated by both bovine serum albumin and a basic cytoplasmic protein from rat liver. Partially purified desaturase is unaffected by either of these two proteins. Bovine serum albumin appears to exert its effect on the crude system by protecting the desaturase substrate, stearoyl-CoA, from the action of endogenous thiolesterases. From amino acid analyses, a comparison was made of the hydrophobicity of the membrane portion of cytochrome b_5 with the hydrophobicity reported for stearoyl-CoA desaturase.

HYPERTRIGLYCERIDEMIA IN EHRLICH ASCITES CARCINOMATOUS MICE: TUMOR AND MOUSE STRAIN DIFFERENCES. R. Kannan and N. Baker (Tumor-Lipid Lab., Res. Service, Veterans Admin. Wadsworth Hosp. Ctr., Los Angeles, Calif. 90073) *Lipids* 12, 153–8 (1977). Ehrlich ascites carcinoma growth in mice induces hypertriglyceridemia. The degree of hypertriglyceridemia found in one laboratory (Spector's) was much greater than we observed in our laboratory. Moreover, major differences were reported with respect to fasting (no effect on tumor extracellular fluid triglyceride levels in Spector's tumor-bearing mice; marked decrease in ours). We have obtained tumorous CBA mice from Spector's laboratory and have studied them simultaneously with our Swiss-Webster mice. Triglyceride levels of the above two groups and from two controlled cross-over groups, included to evaluate the influence of mouse and tumor strains on hypertriglyceridemia, were determined. A mouse strain difference was also evident from a significant decrease in wet weights of adipose tissues like epididymal fat, inguinal fat, and intermuscular fat with tumor growth in the CBA strain which was not observed in the Swiss-Webster strain at the corresponding stage of tumor growth. Study of these strain differences may lead to an understanding of factors that regulate hyperlipidemia.

COMPARATIVE EFFECTS OF PHYSICAL TRAINING AND DIET IN NORMALIZING SERUM LIPIDS IN MEN WITH TYPE IV HYPERLIPOPROTEINEMIA. R.M. Lampman, J.T. Santinga, M.F. Hodge, W.D. Block, J.D. Flora, Jr, and D.R. Basset. (Dept. of Internal Medicine, Dept. of Bio. Chem., and the Dept. of Biostatistics, Univ. of Michigan Med. Center, Ann Arbor, Mich.) *Circulation* 55, 652–9 (1977). The effect of mild physical training (PT) (group A), Type IV hyperlipoproteinemia (HLP) diet (group B), and PT plus Type IV HLP diet on serum lipids (group C) in 46 men with Type IV HLP was studied. Significant reduction in mean triglyceride (TG) levels from 163, 229, 196, to 136, 145, 116 mg/100 ml serum were found for groups A, B, and C, respectively. Following six weeks of intervention, cholesterol levels also dropped for all groups with the greatest reductions occurring in groups B and C. Minimal weight losses were found for all groups while groups A and C displayed significant reductions in body fatness, but both of these changes appeared independant of lipid reductions. It was concluded that either mild PT or HLP diet or both are effective means of lowering TG levels in Type IV HLP individuals. Furthermore, it appears that patients need to participate regularly in formal programs in order to maintain adherence to these interventions.

HDL CHOLESTEROL AND OTHER LIPIDS IN CORONARY HEART DISEASE. W.P. Castelli, J.T. Doyle, T. Gordon, C.G. Hames, M.C. Hjortland, S.B. Hulley, A. Kagan and W.J. Zukel (National Heart, Lung, and Blood Inst., Landow Bldg., Room C841, 7910 Woodmont Ave., Bethesda, MD) *Circulation* 55, 767–78 (1977). The relation between coronary heart disease (CHD) prevalence and fasting lipid levels was assessed by a case-control study in five populations with a total of 6859 men and women of black, Japanese and white ancestry drawn from subjects aged 40 years and older from populations in Albany, Framingham, Evans County, Honolulu and San Francisco. In each major study group mean levels of high density lipoprotein (HDL) cholesterol were lower in persons with CHD than in those without the disease. The average difference was small-typically 3–4 mg/dl—but statistically significant. It was found in most age-race-sex specific groups. The inverse HDL cholesterol-CHD association was not appreciably diminished when adjusted for levels of low density lipoprotein (LDL) cholesterol and triglyceride. LDL, total cholesterol and triglycerides were directly related to CHD prevalence; surprisingly, these findings were less uniformly present in the various study groups than the inverse HDL cholesterol-CHD association.

LONG-CHAIN DIGLYCEROL TETRAETHERS FROM THERMOPLASMA ACIDOPHILUM. T.A. Langworthy (Dept. of Microbiol., Schl. of Medicine, Univ. of South Dakota, Vermillion, SD) *Biochim. Biophys. Acta* 487, 37–50 (1977). The C_{40} isopranol-containing glycerol ether residues which characterize the complex lipids of the extreme thermoacidophile *Thermoplasma acidophilum* were isolated and purified from the glycolipid and phospholipid fractions. The glycerol ether, as well as the acetate and methoxy derivatives were characterized by thin-layer, gel-permeation and gas-liquid chromatography, infrared, nuclear magnetic resonance and mass spectrometry and by vapor phase osmometry. The glycerol ethers are proposed to be unique fully saturated diglycerol tetraethers, primarily $C_{86}H_{172}O_6$, M_r 1300, which contain two *sn*-2,3-glycerol residues bridged through ether linkages by two C_{40} isopranoid branched diols.

OSMOTIC AND PEROXIDATIVE FRAGILITIES OF ERYTHROCYTES FROM VITAMIN E-DEFICIENT LEAD-POISONED RATS. O.A. Levander, R.J. Ferretti and Virginia C. Morris (Nutr. Inst., Agr. Res. Service, U.S. Dept. of Agr., Agr. Res. Ctr., Beltsville, Md. 20705) *J. Nutr.* 107, 373–7 (1977). Weanling male rats were fed either a vitamin E-deficient Torula yeast diet fortified with selenium or the same diet supplemented with 100 ppm vitamin E. One group of rats fed each diet received plain distilled water, whereas another group received 250 ppm lead as lead acetate in the drinking water. After a 3 month feeding period, erythrocyte osmotic and peroxidative fragilities were determined in an osmotic test recorder. Dietary vitamin E had little or no effect on the osmotic fragility of red cells. Lead in the drinking water, however, decreased the osmotic fragility of red cells from deficient rats. Lead poisoning also markedly decreased the elevated peroxidative fragility characteristic of erythrocytes from vitamin E-deficient rats.

COMPARATIVE EFFECTS OF SELENIUM AND VITAMIN E IN LEAD-POISONED RATS. O.A. Levander, V.C. Morris and R.J. Ferretti (Nutr. Inst., Agr. Res. Service, U.S. Dept. of Agr., Agr. Res. Ctr., Beltsville, Md. 20705) *J. Nutr.* 107, 378–82 (1977). Weanling male rats were fed a Torula yeast diet supplemented with selenium, vitamin E, or both for 3 months. Of rats fed each diet, one group received 250 ppm lead in the drinking water and another group did not. In rats not poisoned with lead, neither vitamin E nor selenium deficiency affected spleen weight, hematocrit value, or erythrocyte mechanical fragility. Vitamin E deficiency increased the splenomegaly, anemia, and mechanical fragility of red cells of lead-poisoned rats, whereas selenium deficiency did not. These results show that vitamin E status of rats is more important than selenium status in determining response to toxic levels of lead. Excess dietary selenium did protect partially against lead poisoning in vitamin E-deficient rats, but the levels of selenium used were toxic in themselves.

FILTERABILITY OF ERYTHROCYTES FROM VITAMIN E-DEFICIENT LEAD-POISONED RATS. O.A. Levander, V.C. Morris, and R.J. Ferretti (Nutr. Inst., Agr. Res. Service, U.S. Dept. of Agr., Agr. Res. Ctr., Beltsville, Md. 20705) *J. Nutr.* 107, 363–72

(1977). The time required for red blood cells (RBC) from vitamin E-deficient lead-poisoned (−E + Pb) rats to pass through polycarbonate filters after incubation in vitro was much greater than that of RBC from vitamin E-supplemented non-poisoned rats. Vitamin E deficiency per se (i.e., in non-poisoned rats) often increased filtration times, but in all such experiments the RBC from −E + Pb groups had even longer filtration times. Administration of lead to rats supplemented with vitamin E had little effect on the filtration rate of RBC. Addition of lead in vitro increased filtration times of RBC from both vitamin E-deficient and supplemented non-poisoned rats, but filtration times tended to be longer in the deficient group.

A CONFIRMATION OF THE PHASE BEHAVIOR OF ESCHERICHIA COLI CYTOPLASMIC MEMBRANE LIPIDS BY X-RAY DIFFRACTION. C.D. Linden, J.K. Blasie and C.F. Fox (Mole. Biol. Inst. and Dept. of Bacteriol., Univ. of California, Los Angeles, Calif. 90024) *Biochemistry* **16**, 1621–5 (1977). The lipid fatty acid composition of the cytoplasmic membranes of *Escherichia coli* can be varied by growing an unsaturated fatty acid auxotroph in the presence of different fatty acid supplements. Electron spin resonance (ESR) studies of spin-label partitioning into the cytoplasmic membranes of different lipid fatty acid compositions as a function of temperature have been interpreted as indicating a broad order-to-disorder transition in the membrane lipids, the end points of the transition depending upon the fatty acid composition. We have utilized x-ray diffraction to confirm the ESR studies for three different fatty acid supplements (oleic, elaidic, and bromostearic). We found that the characteristic end-point temperatures detected by ESR were indeed the end-point temperatures of a broad order-to-disorder transition of the cytoplasmic membrane lipids. In addition, Patterson functions calculated from lamellar x-ray diffraction from partially oriented cytoplasmic membranes indicate a decrease in average membrane thickness upon fatty acid chain melting.

CYCLIC FLUCTUATIONS OF PLASMA CHOLESTEROL IN THE FEMALE MINIATURE SWINE AND ITS RELATIONSHIP TO PROGESTERONE SECRETION. S. Lussier-Cacan, E. Bolte, M. Bidallier, Y.S. Huang and J. Davignon (Dept. of Lipid Metab. and Athero. Res., Clinical Res. Inst. of Montreal, 110 Pine Ave. West, Montreal, Quebec H2W 1R7, Canada) *Proc. Soc. Exp. Biol. Med.* **154**, 471–4 (1977). Large fluctuations in plasma cholesterol concentration were noted during a study on the metabolic effects of portacaval anastomosis in the female miniature swine. The fluctuations were cyclic and related to the estrous cycle, as shown by measurements of total plasma cholesterol and progesterone on samples obtained almost daily from six animals over several estrous cycles. Hormone concentrations indicated a 21-day estrous cycle consisting of a 7-day follicular phase and a 14-day luteal period. Plasma cholesterol fluctuated in a cycle which was the inverse of that for progesterone: High cholesterol concentrations were observed for 3–6 days during the follicular phase with peak values as much as 80%, and on the average 50%, higher than the mean levels observed during the luteal phase. Failure to recognize these plasma cholesterol cyclic fluctuations can totally confuse the interpretation of studies on cholesterol metabolism in swine, which are increasingly popular as experimental models of atherosclerosis and lipid metabolism.

FEEDING VALUE OF PROTECTED ANIMAL TALLOW FOR HIGH YIELDING DAIRY COWS. G.K. Macleod, Y. Yu and L.R. Schaeffer (Univ. of Guelph, Guelph, Ontario, Canada) *J. Dairy Sci.* **60**, 726–38 (1977). Supplement containing animal tallow protected with formaldehyde-treated soybean meal was introduced into concentrates at 0, 21, and 32% and fed ad libitum to Holstein cows in a changeover design during early lactation. Primary objective was to determine whether feeding protected tallow in concentrates would increase voluntary energy intake, milk yield, and fat test without adversely affecting other milk components or flavor. Cholesterol, formaldehyde, and flavor of milk were unaffected by treatment whereas cholesterol and triglycerides in blood plasma rose dramatically during short-term feeding of protected tallow. Blood glucose remained unchanged.

A FACTOR IN YOGURT WHICH LOWERS CHOLESTEREMIA IN MAN. G.V. Mann (Vanderbilt Univ. School of Med., Dept. of Biochem., Nashville, Tenn. 37232) *Atherosclerosis* **26**, 335–40 (1977). Large dietary intakes of yogurt are found to lower cholesteremia in man. This effect is associated with a reduction of incorporation of radioacetate into serum cholesterol. The effect appears slowly and persists after intake of the yogurt stops suggesting that the mechanism involves the synthesis of a regulatory protein rather than an allosteric effect. The effective agent is postulated to be hydroxymethyl glutarate which inhibits the regulatory enzyme hydroxymethyl glutaryl CoA reductase (EC 1.1.1.3.4).

EFFECTS OF DIETARY FIBER AND SALT MIXTURES ON THE CHOLESTEROL METABOLISM OF RATS. D. Mathe, C. Lutton, J. Rautureau, T. Coste, E. Gouffier, J.C. Sulpice and F. Chevallier (Lab. de Physiol. de la Nutr., Univ. Paris Sud, Batiment 447, F 91405 Orsay Cedex, France) *J. Nutr.* **107**, 466–74 (1977). The isotopic dilution method, which permits the in vivo measurements of the rates of the processes involved in cholesterol turnover, has been applied to rats fed a commercial stock diet or a basal semipurified diet in which either the nature and proportions of the source of dietary fiber or the salt mixture were changed. The cholesterolemia was about 100 mg/100 g in rats fed agar-agar, cellulose, bran or the stock diet. Pectin addition (5%) lowered significantly the plasma concentration of cholesterol (70 mg/100 g). Cholesterol biosynthesis and fecal excretion were inversely correlated to the absorption coefficient of dietary cholesterol in rats fed all of the semipurified diets indicating, as previously shown, that the intestine was the major source of biosynthesized cholesterol diverted into the plasma.

THE CHICK'S REQUIREMENT FOR 25-HYDROXYCHOLECALCIFEROL AND CHOLECALCIFEROL. J.L. McNaughton, E.J. Day, and B.C. Dilworth (Poultry Sci. Dept., Mississippi Agr. and Forestry Experiment Station, Miss. State Univ., MS) *Poult. Sci.* **56**, 511–6 (1977). Three experiments were conducted to compare the effect of 25-hydroxycholecalciferol (25-OHD) and cholecalciferol (D_3) on three week weight gains, feed utilization and tibia ash of broiler cockerels. Tibia ash statistically equal to the maximum response was achieved with 132 I.C.U./kg of 25-OHD_1 was found to be 1.5 times greater than D_3 based on requirement levels of both vitamin D sources.

A FACTOR IN MILK WHICH INFLUENCES CHOLESTEREMIA IN RATS. C.R. Nair and G.V. Mann (Vanderbilt Univ. School of Med., Dept. of Biochem., Nashville, Tenn. 37232) *Atherosclerosis* **26**, 363–7 (1977). Rats fed cholesterol show an adaptive response over several weeks with lowering of the initial rise of cholesteremia to near normal levels. This reduction of cholesteremia is augmented by including milk powder in the diet. A similar effect is obtained by feeding 0.1% β-hydroxy-β-methylglutaric acid (HMG) in the diet. The evidence suggests that milk powder contains HMG.

CHANGE IN RATS' SERUM TRIGLYCERIDE CONCENTRATION WITH GRADED LEVELS OF THYROXINE AND EXERCISE. H.K. Naito and D.R. Griffith (Dept. of Athero. and Thrombosis Res., Div. of Res., and Dept. of Biochem., Div. of Lab. Med., The Cleveland Clinic Found., Cleveland, Ohio 44106) *Proc. Soc. Exp. Biol. Med.* **154**, 372–6 (1977). Male thyroidectomized rats with varying levels of daily L-thyroxine injection were subjected to different amounts of physical activity to determine the influence of both factors on serum TG concentration. The study indicated that exercise is effective in lowering serum TG and that this hypotriglyceridemic response is directly related to the increasing severity of exercise. The control animals with intact thyroids had higher TG levels as compared to the thyroidectomized rats with differing replacement levels of L-T_4 irrespective of the degree of physical activity. The differing feed intake in the various groups could not account for this hypotriglyceridemic effect. The lower body weight of the thyroidectomized animals as compared to the control rats with intact thyroid glands, may be related, in part, to the lower TG concentration. The study suggests that the thyroid gland, in some manner, plays an important role in the hypotriglyceridemic effect of exercise.

PREGASTRIC ESTERASE AND OTHER ORAL LIPASES—A REVIEW. J.H. Nelson, R.G. Jensen and R.E. Pitas (Dairyland Food Lab., Inc., Waukesha, Wis. 53186) *J. Dairy Sci.* **60**, 327–62 (1977). The secretion of pregastric esterase and other oral lipases has been detected in 13 species. Research on secretion by the human, calf, kid goat, lamb, and rat of pregastric esterase has been significant. Secretion by calves is little affected by age or diet but is greater when calves are nipple fed than when pail fed.

REPRODUCTION AND LIPID COMPOSITION OF RATS FED CYCLOPROPENE FATTY ACIDS. J.E. Nixon, T.A. Eisele, J.D. Hendricks and R.O. Sinnhuber (Dept. of Food Sci. and Tech., Oregon State Univ., Corvallis, Ore.) *J. Nutr.* **107**, 574–83 (1977). The

effect of cyclopropene fatty acids fed in saturated or unsaturated lipid diets on reproduction and lipid composition of progeny was determined in rats. *Sterculia foetida* oil (50% cyclopropene fatty acids) fed at 0.2% of the diet for three generations and 0.5% fed to first generation rats did not significantly affect breeding. Two percent *S. foetida* oil fed with 3% corn oil did not appreciably affect conception rate and litter size in the first litters, but reduced pup survival 36% in the first litters and 78% in the second litters. Unsaturated lipids in the diet enabled rats to cope with the effects of moderate levels of cyclopropene fatty acids, but the combination of cyclopropene in saturated lipid diets caused detrimental effects.

RECOVERY OF BRAIN FROM DEFICIENCY OF ESSENTIAL FATTY ACIDS IN RATS. A.A. Odutuga (Dept. of Biochem., Ahmadu Bello Univ., Zaria, Nigeria) *Biochim. Biophys. Acta* **487**, 1–9 (1977). Rats were made visibly deficient in essential fatty acids by feeding a deficient diet and were then fed a diet containing 5.0 per cent by weight corn oil as a source of essential fatty acids. After 11–13 weeks on deficient diet the weights of the brain were 25% less than those of rats on control diets. After 5 weeks on the control diet, deficient animals had regained normal brain weight and composition and had lost the deficiency symptoms. Lipids were extracted from the brains and analysed qualitatively and quantitatively, the overall effect of essential fatty acid deficiency being a reduction in the proportion of cerebrosides and sphingomyelin, the appearance of a high proportion of eicosatrienoic acid (20:3) and the reduction of arachidonic acid (20:4) and the other essential fatty acids. It is considered that essential fatty acid deficiency retards maturing of brains and the present data show that this effect is reversible.

EFFECTS OF PROTEIN AND FAT IN THE DIETS ON HATCHABILITY OF EGGS AND CHICK GROWTH. M.B. Patel and J. McGinnis (Dept. of Animal Sci., Washington State Univ., Pullman, Washington) *Poult. Sci.* **56**, 529–37 (1977). An experiment was conducted using Single Comb White Leghorn (S.C.W.L.) hens to study the effect of fat and vitamin B_{12} (50 μg./kg.) additions to a 32% protein diet; on production parameters, hatchability and chick growth. Levels of protein, fat and vitamin B_{12} had no significant effect on egg production, however, vitamin B_{12} supplementation of the 32% protein diet gave a considerable improvement in egg weight. Feed consumption was highest on the 32% protein diet. Adding 8.5% fat to the 16 and 32% protein diets significantly reduced feed intake. The 32% protein diet supported the poorest hatchability. Animal fat (8.5%) or vitamin B_{12} (50 μg./kg.) added to the 32% protein diet supported satisfactory hatchability. On the contrary, addition of 8.5% fat to the 16% protein diet significantly reduced hatchability of eggs. Vitamin B_{12} content of egg yolks was increased by adding fat to the 32% protein hen diet. Hatchability of eggs from these hens was higher than for eggs from hens fed a similar diet without added fat. The vitamin B_{12} content of livers of day-old chicks was correlated with increased growth of chicks fed vitamin B_{12} deficient diets.

EFFECT OF DIETARY EGG ON SERUM CHOLESTEROL AND TRIGLYCERIDE OF HUMAN MALES. M.W. Porter, W. Yamanaka, S.D. Carlson and M.A. Flynn (Dept. of Nutr. and Dietetics, Schl. of Medicine, Univ. of Missouri-Columbia, MO) *Am. J. Clin. Nutr.* **30**, 490–5 (1977). One hundred fourteen male volunteers (mean age 44.6 years) consumed one whole egg daily in their customary diets for 3 months. Their final serum cholesterol (SCHOL) and triglycerides (STG) levels were compared with their initial levels on customary free choice diets and also with their levels after a 3-month elimination of dietary whole eggs All subjects had previously confirmed normal serum lipid levels and no history of heart disease. Four-day food records were kept during both experimental dietary periods. A Latin square design allowed analysis for seasonal effects on lipid levels. No significant change in mean SCHOL on either diet was found; there was a seasonal effect on mean STG. Significant linear associations of fat intake and of energy intake were found. There was no significant association of dietary cholesterol intake with either SCHOL or STG.

THE EFFECT OF A SHORT TERM SATURATED FAT DIET ON THE APOPROTEIN COMPOSITION AND RADIOIODINATION PROPERTIES OF RAT VERY LOW DENSITY LIPOPROTEINS. P. Poulis and N.H. Fidge (Dept. of Clin. Sci., The John Curtin School of Med. Res., The Australian Nat'l. Univ., Canberra, A.C.T. Australia) *Lipids* **12**, 288–92 (1977). The effect of a saturated fat diet on the apoprotein composition and radioiodination properties of plasma very low density lipoprotein (VLDL) was studied in rats. After feeding the diet for 10 days, the proportion of ^{125}I attached to VLDL lipid decreased from 50% (control animals) to 8%, the remainder (92%) being bound to the apoprotein components. The decreased lipid labelling was associated with proportional changes in the fatty acid composition of serum and VLDL lipids, the most notable change being a reduction in linoleic acid (30–8%) content which occurred in all the major lipid classes of both serum and VLDL. It is concluded that feeding a saturated fat diet to rats for 10 days significantly improved ^{125}I labelling of the apoprotein moiety while apparently not inducing changes in apoprotein composition.

TISSUE STORAGE AND CONTROL OF CHOLESTEROL METABOLISM IN MAN ON HIGH CHOLESTEROL DIETS. E.C.R. Quintao, S. Brumer and K. Stechhahn (Dept. of Med., Hosp. das Clinicas, Univ. of Sao Paulo Med. School, Sao Paulo, Brazil) *Atherosclerosis* **26**, 297–310 (1977). The possibilty of accumulation of tissue cholesterol in human beings submitted to high cholesterol feeding was investigated in liver biopsies and through fecal sterol balance studies. Feeding to 10 individuals 3.1 to 3.4 g/day of cholesterol for 3 weeks raised the mean serum level from 293 to 349 mg/100 ml, namely 19%, whereas the liver cholesterol content was 417 mg/100 g of wet weight. In 10 control cases eating 0.1–0.4 g/day of cholesterol serum cholesterol remained stable throughout the experimental period and the liver cholesterol content was 256 mg/100 g. Difference of liver cholesterol level between the two groups was 62%. In 7 patients submitted to two periods of balance investigation on a cholesterol-free synthetic formula diet respectively prior to (PI) and after (PIII) eating the high cholesterol solid food from 4 to 15 weeks (PII), fecal steroid excretion in PIII exceeded PI in 3 patients. Such data are a direct evidence for the existence of an efficient system to release acutely stored cholesterol. In one patient bile acid excretion accounted for the difference between PIII and PI.

EFFECTS OF VITAMIN B_6 DEFICIENCY ON LIVER, KIDNEY, AND ADIPOSE TISSUE ENZYMES ASSOCIATED WITH CARBOHYDRATE AND LIPID METABOLISM, AND ON GLUCOSE UPTAKE BY RAT, EPIDIDYMAL ADIPOSE TISSUE. J.D. Ribaya and S.N. Gershoff (Dept. of Nutr., Harvard School of Public Health, 665 Huntington Ave., Boston, Mass. 02115) *J. Nutr.* **107**, 443–52 (1977). Adipose tissue and liver from vitamin B_6-deficient rats have an increased lipogenic capacity. Whether this phenomenon is accompanied by changes in the activities of certain enzymes involved in the metabolism of carbohydrate and lipid, or by altered transport of glucose into adipocytes, has been studied. Five glycolytic enzymes (hexokinase, phosphoglucose isomerase, phosphofructokinase, aldolase, and pyruvate kinase), two pentose phosphate pathway enzymes (glucose-6-phosphate dehydrogenase and 6-phosphogluconate dehydrogenase), malic enzyme, and ATP citrate lyase were measured in the epididymal adipose tissue, livers and kidneys of vitamin B_6-deficient and control rats. Vitamin B_6 deficiency did not significantly affect the glycolytic enzyme levels in the tissues studied, or the dehydrogenases measured in adipose tissue and kidneys.

ROLE OF APOLIPOPROTEIN A-I IN THE STRUCTURE OF HUMAN SERUM HIGH DENSITY LIPOPROTEINS. RECONSTITUTION STUDIES. M.C. Ritter and A.M. Scanu (Dept.s of Med. and Biochem., Univ. of Chicago Pritzker School of Med., Chicago, Ill. 60637) *J. Biol. Chem.* **252**, 1208–16 (1977). For a better definition of the role of human serum apolipoprotein A-I (apo A-I) in high density lipoprotein structure, a systematic investigation was carried out on factors influencing the *in vitro* association of this apoprotein with lipids obtained from the parent high density lipoprotein (HDL); these lipids include phospholipids, free cholesterol, cholesteryl esters, and triglycerides. Following equilibration, mixtures of apo A-I and lipids in varying stoichiometric amounts were fractionated by sequential flotation, CsCl density gradient ultracentrifugation, or gel-permeation chromatography, and the isolated complexes were characterized by physicochemical means. The results indicate that *in vitro* complexation of apo A-I with lipids is under kinetic control; apo A-I can generate a lipid · protein complex with properties similar to those of the parent lipoprotein; the process requires well defined experimental conditions and, most importantly, the presence in solution of monomers and dimers of apo A-I; the number of apo A-I molecules incorporated into R-HDL determines the size and structure of the reassembled particle. All of these observations strongly support the essential role of apo A-I in the structure of human HDL.

EFFECTS OF UDP-GLUCOSE ADDITION ON THE SYNTHESIS OF MANNOSYL LIPID-LINKED OLIGOSACCHARIDES BY CELL-FREE FIBROBLAST PREPARATIONS. P.W. Robbins, S.S. Krag and T. Liu (Dept. of Biol. and Ctr. for Cancer Res., Massachusetts Inst. of Technol., Cambridge, Mass. 02139) *J. Biol. Chem.* **252**, 1780–5 (1977). The pattern of mannosyl lipid-linked oligosaccharides synthesized by cell-free enzyme preparations from cultured fibroblasts is altered substantially when 0.2 μM UDP-glucose is added to the incubation medium. Inclusion of UDP-glucose results in the appearance of a new labeled oligosaccharide, which is 1 or 2 glycose units larger than the lipid-linked oligosaccharide synthesized in the presence of only GDP-mannose (2 μM) and UDP-*N*-acetylglucosamine (20 μM). Label from UDP-[^{3}H]glucose is incorporated into the same larger oligosaccharide size class. The results can be explained most easily by assuming that the new mannosyl lipid-linked oligosaccharide contains 1 or 2 glucose residues in addition to 5 to 6 mannose residues. In addition to being incorporated into lipid oligosaccharides, glucose residues are also incorporated into endogenous glycoproteins. Incorporation of glucose into glycoproteins that give rise to pronase glycopeptides of the typical asparagine-linked size classes is almost completely dependent on the presence of GDP-mannose.

EFFECT OF SERUM AND LIVER EXTRACTS FROM HYPERCHOLESTEROLEMIC RATS ON THE SYNTHESIS OF COLLAGEN BY ISOLATED AORTAS AND CULTURED AORTIC SMOOTH MUSCLE CELLS. T. Ronnemaa and N.S. Doherty (Dept. of Med. Chem., Univ. of Turku, Turku 20520 52, Finland) *Atherosclerosis* **26**, 261–72 (1977). Rats were made hypercholesterolemic by feeding them a high-cholesterol, olive oil diet for one week. The effect of sera and 35,000 $\times$ *g* supernatants of liver homogenates on collagen synthesis was studied in isolated aortas, cultured arterial smooth muscle cells and the same cells in suspension. Compared to the preparations from normal rats, the liver preparations from hyperlipidemic rats stimulated collagen synthesis in both isolated aortas and cultured smooth muscle cells by about 25%. In these test systems hyperlipidemic serum was without effect but when added to smooth muscle cells incubated in suspension produced a significant increase in the amount of collagen secreted. Hyperlipidemic serum caused an increase of about 50% in the incorporation of [^{3}H]-thymidine by cultured smooth muscle cells.

SERUM LIPIDS AND PROTEINS IN LACTOSE MALABOSORPTION[1-8]. T. Sahi, J. Jussila, I.M. Penttila, S. Sarna, and M. Isokoski (Dept. of Epidemiology, Harvard Schl. of Public Health, 677 Huntington Ave. Boston, Mass.) *Am. J. Clin. Nutr.* **30**, 476–81 (1977). It has been suggested that dietary lactose may reduce the intestinal absorption of fat and protein in individuals with lactase deficiency. On the other hand, it is known that a high carbohydrate diet increases serum lipids. The purpose of this study was to examine whether there are differences in the fasting serum lipid and protein concentrations between people with lactose malabsorption and people with normal lactose absorption. Therefore in the connection of a family study serum lipids and proteins were measured in 409 subjects belonging to 11 families. Of these 288 were relatives of the 11 index persons and 121 were spouses or relatives of the spouses. It was hypothesized that increased intestinal motility may disturb the absorption of fats and cause the observed difference at least in the Finnish population.

POSITIONAL ISOMERS OF UNSATURATED FATTY ACIDS IN RAT LIVER LIPIDS. B. Schmitz, U. Murawski, M. Pfluger and H. Egge (Inst. of Physiol. Chem., Univ. Bonn, Nussallee 11, 5300 Bonn, German Fed. Rep.) *Lipids* **12**, 307–13 (1977). The fatty acids of liver lipids from rats raised on a fat free diet from the 30th to the 90th day after birth were analyzed with special regard to the detection of positional isomers of mono-, di-, tri-, and tetraenoic fatty acids. The methyl esters obtained after transesterification of total lipids were separated by argentation chromatography into five fractions: I saturated, II monoenoic, III dienoic, IV dienoic nonmethylene interrupted, V tri- and tetraenoic fatty acid esters. After hydroxylation of the double bonds with osmium tetroxide, the analysis of the poly-O-trimethylsilyl derivatives by gas liquid chromatography on S.C.O.T. columns combined with mass spectrometry revealed the presence of 19 monoenoic, 15 dienoic, and 9 trienoic as well as 3 tetraenoic fatty acid isomers including the normally occurring representatives of the (n-3), (n-6), (n-7), and (n-9) fatty acid families. The great number of isomers found in the (n-7) family indicates that the members of this family are actively metabolized in partial essential fatty acid deficiency.

THE EFFECT OF LIPIDS ON TAUROCHOLATE ABSORPTION FROM INTESTINAL LOOPS IN THE RAT. D. Sklan and P. Budowski (Faculty of Agr., The Hebrew Univ. of Jerusalem, Rehovot, Israel) *Lipids* **12**, 193–7 (1977). The rates of uptake and serosal transfer of [^{14}C]-labelled taurocholate (7.77 mM in bicarbonate buffer, pH 6.5) were determined in situ in ligated segments of rat intestine in the presence of lipids. Oleic acid, monoolein, lecithin, and lysolecithin enhanced taurocholate uptake and transfer in the jejunum, each lipid exhibiting an optimal concentration at which the bile acid fluxes were maximal. The maximal rates of bile acid uptake observed with the various lipids were close to four times the uptake rates found with the lipid-free taurocholate medium, whereas serosal transfer rates under optimal conditions were enhanced about six-fold. The optimal concentrations differed widely among the various lipids, being inversely related to the lipids' polarity.

STEREOCONFIGURATION OF BISPHOSPHATIDIC AND SEMILYSOBISPHOSPHATIDIC ACIDS FROM CULTURED HAMSTER FIBROBLASTS (BHK CELLS). P. Somerharju, J. Brotherus, K. Kahma and O. Renkonen (Dept. of Biochem., Lab. of Lipid Res., Univ. of Helsinki, SF-00290 Helsinki 29, Finland) *Biochim. Biophys. Acta* **487**, 154–62 (1977). Monolayers of hamster fibroblasts (BHK cells) were incubated in Eagle's minimal essential medium under conditions where and increase in the levels of all cellular bisphosphatidic acids takes place. Bisphosphatidic acid and semilysobisphosphatidic were isolated from these cells and subjected to strong alkaline hydrolysis. Steriochemical analysis of the hydrolysis products revealed that the majority of the molecules of both lipids are derivatives of *sn*-1-glycerophospho-*sn*-1'-glycerol, the structure previously found to be the "backbone" of lysobisphosphatidic acid, (bis(monoacylglycerol)phosphate) from BHK cells and other sources. This finding suggests a close metabolic relationship between the three bisphosphatidic acid derivatives of BHK cells.

STIMULATION OF CHOLESTEROL ESTERIFICATION IN RHESUS MONKEY ARTERIAL SMOOTH MUSCLE CELLS. R.W. St. Clair, Beth P. Smith and L.L. Wood (Arteriosclerosis Res. Ctr., Dept. of Pathol., Bowman Gray School of Med., Winston-Salem, N.C.) *Cir. Res.* **40**, 166–73 (1977). The influence of homologous high density lipoprotein (HDL) and low density lipoprotein (LDL) and of whole hypercholesterolemic serum on the esterification of oleic acid and cholesterol was studied in rhesus monkey arterial smooth muscle cells. These studies provide further evidence that a major consequence of the interaction of plasma LDL with the cellular elements of the arterial wall is a stimulation of cholesterol esterification. These studies, coupled with the observation that cholesteryl esters, more than any other single component, increase in the atherosclerotic artery, suggest an important role of a stimulation in cholesterol esterification in the pathogenesis of atherosclerosis.

CHLOROQUINE-INDUCED INTERFERENCE WITH DEGRADATION OF SERUM LIPOPROTEINS IN RAT LIVER, STUDIED IN VIVO AND IN VITRO. Y. Stein, V. Ebin, H. Bar-On and O. Stein (Lipid Res. Lab., Dept. of Med., B, Hadassah Univ. Hosp., Jerusalem, Israel) *Biochim. Biophys. Acta* **486**, 286–97 (1977). The effect of chloroquine, an inhibitor of certain lysosomal enzymes including cathepsin B (EC 3.4.22.1), on the degradation of serum lipoproteins in rat liver was studied in vivo and in liver homogenates. Chloroquine had no effect on the clearance from the circulation of ^{125}I-labeled rat or human very low density lipoproteins or human low density lipoproteins. Pretreatment with chloroquine for 3 h, resulted in a 2–2.5 fold increase in ^{125}I-labeled very low density lipoprotein recovered in the liver 45 min after injection of the homologous and heterologous lipoproteins. This effect was evident on both the ^{125}I-labeled protein and ^{125}I-labeled lipid moiety. Degradation of very low density and low density lipoproteins was completely inhibited at 0.05 M chloroquine, while less pronounced inhibition was seen with high density lipoproteins, apolipoproteins and apolipoprotein AI. These results indicate that liver acid hydrolases in vivo participate in the degradation of serum lipoproteins. Cathepsin B is apparently responsible for the degradation of apolipoprotein B, while other cathepsins might also be active in the degradation of this and the other apolipoproteins.

EVIDENCE FOR THE PARTICIPATION OF SACCHARIDE-LIPIDS IN THE SYNTHESIS OF THE OLIGOSACCHARIDE CHAIN OF OVALBUMIN. D.K. Struck and W.J. Lennarz (Dept. of Physiol. Chem., The Johns Hopkins Univ. School of Med., Baltimore, Md. 21205) *J. Biol. Chem.* **252**, 1007–13 (1977). To obtain information on

the mechanism of glycosylation of ovalbumin, three types of experiments were performed with either hen oviduct membrane preparations or tissue slices and the antibiotic tunicamycin. Finally, it was found that tissue slices incubated in the presence of *tunicamycin* synthesized the polypeptide chain of ovalbumin at almost normal rates. However, the protein newly synthesized *in vivo* did not contain labeled *N*-acetylglucosamine or mannose and had the properties of unglycosylated ovalbumin. These results indicate that saccharide-lipids participate in the assembly of the core oligosaccharide of the secretory glycoprotein, ovalbumin.

SERUM INSULIN CONCENTRATION, INSULIN RELEASE AND DEGRADATION, GLUCOSE TOLERANCE AND IN VIVO INSULIN SENSITIVITY IN CHOLESTEROL-FED RATS. A.C. Tsai (Human Nutr. program, Schl. of Public Health, Univ. of Michigan, Ann Arbor, Mich.) *J. Nutr.* **107**, 546–51 (1977). A series of experiments was conducted to elucidate the mechanism by which cholesterol feeding decreases serum insulin levels in rats and to determine the effect of this reduced insulin level on glucose metabolism. Rats were fed a casein-sucrose-soybean oil basal diet or this basal diet supplemented with 1% cholesterol (dissolved in hot oil) for periods longer than 30 days. Cholesterol feeding resulted in a decrease in serum insulin concentrations, although the decrease was not always significant. Cholesterol feeding did not affect fasting blood glucose levels, glucose tolerance, glucose-induced insulin release, pancreatic insulin content, in vivo insulin sensitivity, or in vitro glucose utilization in diaphragm and adipose tissue, but it significantly elevated the activity of liver glutathione-insulin transhydrogenase. On a per liver basis, the activity of this enzyme was approximately doubled. Results of this study suggest that cholesterol feeding has no significant effect on glucose utilization, but it can lead to a decrease in serum insulin concentration, probably by increasing the rate of insulin degradation in the liver.

SUPPLEMENTATION WITH VITAMIN E IN HYPERLIPIDEMIC PATIENTS TREATED WITH DIET AND CLOFIBRATE. EFFECTS ON SERUM LIPOPROTEIN CONCENTRATIONS, PLASMA FATTY ACID COMPOSITION AND ADIPOSE TISSUE LIPOPROTEIN LIPASE ACTIVITY. B. Vessby, H. Lithell, and J. Boberg (Dept. of Geriatrics, Univ. of Uppsala, Uppsala, Sweden) *Am. J. Clin. Nutr.* **30**, 517–22 (1977). Twelve hyperlipidemic patients on long term treatment with a lipid lowering diet enriched in polyunsaturated fatty acids and with clofibrate were supplemented with vitamin E (400 mg/day). The effect on serum lipoprotein concentration, plasma lipid fatty acid composition, and adipose tissue lipoprotein lipase activity was studied. No additional lipid-lowering effect was registered during a treatment period of 4 months. A slight increase in total serum cholesterol concentration and in high density lipoprotein concentrations was probably attributable to seasonal variations in serum lipoprotein concentrations. No major changes of fatty acid composition in plasma cholesteryl esters or triglycerides were recorded. However, an increased relative amount of arachidonic acid and a reduced amount of palmitic acid in the plasma phospholipids after 2 months was possibly caused by the vitamin E therapy.

EFFECT OF A DIET RICH IN SUNFLOWER OIL ON ASPECTS OF LIPID METABOLISM IN THE GENETICALLY-OBESE RAT. K.W.J. Wahle and J.D. Radcliffe (Rowett Res. Inst., Bucksburn, Aberdeen, AB2 9SB, Great Britain) *Lipids* **12**, 135–9 (1977). Aspects of the lipid metabolism of male, obese and lean Zucker rats were compared using animals which had been fed ad libitum for 32 days on a diet (HS) which contained 200 g sunflowerseed oil/kg or one (LS) which contained 50 g/kg of the oil. When compared with the LS diet, the HS diet decreased the characteristic lipid accretion in the liver of obese rats from 126 mg (LS) to 81 mg (HS)/g wet weight; corresponding values for the lean rats were 39 mg and 56 mg/g wet weight of liver, respectively. HS compared with the LS diet resulted in increased proportions of 18:2ω6 in liver lipids and adipose tissue triacylglycerols of obese and lean rats. The HS diet also increased the proportions of 20:4ω6 in adipose triacylglycerols of obese and lean rats and in liver lipids of obese animals but not in their lean littermates.

LIPIDS OF CULTURED HEPATOMA CELLS: VIII. UTILIZATION OF D-[1-^{14}C] GLUCOSE FOR LIPID BIOSYNTHESIS. C.L. Welch and R. Wood (Div. of Gastroenterol., Depts. of Med. and Biochem., Univ. of Missouri School of Med., Columbia, Mo. 65201) *Lipids* **12**, 245–53 (1977). Minimal deviation hepatoma 7288C cells (HTC) were incubated in serum-supplemented and serum-free Swim's 77 medium in the presence of D-[1-^{14}C] glucose for 1, 2, 4, 8, 12 and 24 hr. Glucose oxidation to CO_2, incorporation into total cell mass, and incorporation into cell and medium lipids were determined. The percentage distribution of total cell lipid radioactivity in individual neutral and polar lipid classes was followed as a function of time. Degradation studies of individual lipid classes were performed to ascertain the percentage of radioactivity in acyl and glycerol moieties. Glycerol from glyceride classes contained a higher percentage of radioactivity than the acyl moieties, with this percentage significantly elevated in serum-free cultures. The data indicate that, although glucose is a substrate for HTC cell lipids, other precursors present in the culture system also contribute to the lipid constituency of this hepatoma cell line.

THE EFFECT OF PALMITOYL-COENZYME A ON RAT HEART AND LIVER MITOCHONDRIA. OXYGEN CONSUMPTION AND PALMITOYLCARNITINE FORMATION. J. McMillin Wood, E.T. Wallick, A. Schwartz and C.-H. Chang (Depts. of Cell Biophys. and Pediatrics, Baylor College of Med., Houston, Tex. 77030) *Biochim. Biophys. Acta* **486**, 331–40 (1977). Rat heart and liver mitochondria, respectively, oxidized palmitoyl-CoA and palmitoylcarnitine optimally at 20–30 and 10–20 nmol substrate/mg. The oxidation of palmitoyl-CoA was accompanied by a lag in State 3 respiration that was proportional to the palmitoyl-CoA concentration. The delay in State 3 rates was more prolonged in liver than in heart at comparable palmitoyl-CoA levels. A similar range of palmitoyl-CoA concentrations produced significant inhibition of respiration in mitochondria oxidizing glutamate-malate. The inhibition was not due to a detergent effect of palmitoyl-CoA since addition of carnitine restored State 3 rates. The prolongation in time to reach equilibrium may account for the relatively greater respiratory sensitivity of liver mitochondria to increasing levels of palmitoyl-CoA.

MILK AND TISSUE LIPID COMPOSITION AFTER FEEDING COWS PROTECTED POLYUNSATURATED FAT FOR TWO YEARS. T.R. Wrenn, J. Bitman, J.R. Weyant and D.L. Wood, K.D. Wiggers, L.F. Edmondson (Nutrient Utilization Lab., Animal Physiology and Genetics Inst., ARS, USDA, Beltsville Agr. Res. Center, Beltsville, MD) *J. Dairy Sci.* **60**, 521–32 (1977). The long-term effects of feeding Holstein cows plant lipids protected from microbial hydrogenation in the rumen were studied. Of particular interest were cow health and changes in fatty acid and cholesterol concentrations of milk and meat. Safflower oil-casein or safflower oil-casein treated with formaldehyde to impede microbial attack were fed to two groups of three cows as 10% of the concentrate ration for two lactations. Production of milk fat of cows fed the protected concentrate increased significantly. Linoleic acid of milk fat was twice normal, providing a polyunsaturated milk. Cholesterol of milk or meat did not increase even though cholesterol of blood plasma was higher in both groups fed safflower oil than in control cows. Cardiovascular systems showed no marked abnormalities and no differences that could be due to treatment. All cows maintained normal health and milk production throughout the experiment.

MOLECULAR DYNAMICS OF THE LOCAL ANESTHETIC TETRACAINE IN PHOSPHOLIPID VESICLES. P.L. Yeagle, W.C. Hutton and R.B. Martin (Chem. Dept., Univ. of Virginia, Charlottesville, Va.) *Biochim. Biophys. Acta* **465**, 173–8 (1977). Upon introduction into phosphatidylcholine vesicles, the ^{13}C magnetic resonance peaks of the aromatic resonances of tetracaine are broadened while the T_1 relaxation times show little change. Addition of tetracaine to vesicles containing 30% cholesterol produces a similar broadening in the ^{13}C NMR spectrum of tetracaine. Nuclear magnetic resonance parameters of phosphatidylcholine in vesicles which are unchanged by the addition of equimolar tetracaine include ^{13}C T_1 relaxation time and ^{31}P linewidth, T_1 relaxation time, and nuclear Overhauser effect enhancement. These results are interpreted as indicating a hydrophobic interaction between hydrocarbon portions of the anesthetic and phospholipid bilayer. Using shift reagents and ^{31}P NMR, tetracaine has been shown to displace cations from the bilayer surface, and does not undergo fast flip-flop across the vesicle bilayer.

TOPOGRAPHICAL DISSECTION OF SHEEP ERYTHROCYTE MEMBRANE PHOSPHOLIPIDS BY TAUROCHOLATE AND GLYCOCHOLATE. D. Billington, R. Coleman and Y.A. Lusak (Dept. of Biochem., Univ. of Birmingham, P.O. Box 363, Birmingham B 15 2 TT, U.K.) *Biochim. Biophys. Acta* **466**, 526–30 (1977). Glycocholate and taurocholate removed significant amounts of membrane phospholipid from intact sheep erythrocytes before lysis of the cells occurred. The pre-lytic extract was enriched in spingomyelin and correspondingly depleted in phophatidylserine,

phosphatidylinositol and phosphatidylethanolamine when compared to the original membrane. In contrast, the phospholipid profiles of glycocholate and taurocholate extracts of unsealed ghosts, made at the same bile salt concentrations, were similar to that of the whole membrane. These observations are related to the topography of the phospholipids in the membrane and to some aspects of bile formation.

LIPID PHASE SEPARATION INDUCED BY A HYDROPHOBIC PROTEIN IN PHOSPHATIDYLSERINE-PHOSPHATIDYLCHOLINE VESICLES. J.M. Boggs, D.D. Wood, M.A. Moscarello and D. Papahadjopoulos (Biochem. Dept., Hosp. for Sick Children, Toronto, Ontario). *Biochemistry* 16, 2325–9 (1977). Differential scanning calorimetry (DSC) was used to detect phase separation induced by a hydrophobic myelin protein, lipophilin, in a mixture of phosphatidylserine (PS) and dipalmitoylphosphatidylcholine (DPPC). Preferential binding of PS to the boundary layer of lipophilin causes a decrease in the PS content of the remaining lamellar phase with a resultant shift in the phase-transition temperature to a higher temperature.

NANOSECOND TIME-DEPENDENT FLUORESCENCE DEPOLARIZATION OF DIPHENYLHEXATRIENE IN DIMYRISTOYLLECITHIN VESICLES AND THE DETERMINATION OF "MICROVISCOSITY." L.A. Chen, R.E. Dale, S. Roth and L. Brand (Bio. Dept. and McCollom-Pratt Inst., Johns Hopkins Univ., Baltimore, MD) *J. Biol. Chem.* 252, 2163–9 (1977). The nanosecond time dependence of the fluorescence depolarization of 1,6-diphenyl-1,3,5-hexatriene in L-α-dimyristoyllecithin vesicles was determined at temperatures above and below the midpoint of the gel-liquid crystalline transition. In neither case could the decay of the total fluorescent emission or the decay of the emission anisotropy be described adequately in terms of single exponential decay laws. At the lower temperature, the emission anisotropy did not approach zero in the time window available for measurement, a finding which may indicate that the range over which rotation of the probe can freely occur is restricted. The results are discussed in relation to the concept of microviscosity of bilayer membranes.

PROTEIN-LIPID INTERACTIONS: RECOMBINANTS OF THE PROTEOLIPID APOPROTEIN OF MYELIN WITH DIMYRISTOYLLECITHIN. W. Curatolo, J.D. Sakura, D.M. Small and G.G. Shipley (Biophys. Div., Dept. of Med., Boston Univ. Schl. of Med., Boston, MA). *Biochemistry* 16, 2313–8 (1977). Recombinants of the proteolipid apoprotein (PLA) of bovine myelin with dimyristoyllecithin (DML) have been prepared under conditions which maximize the opportunity for hydrophobic interactions. X-ray diffraction studies show that the overall lamellar structure of the lecithin is preserved when PLA is incorporated into the DML liposomes. Both above and below the temperature of the DML order-disorder transition, maximally hydrated DML/PLA recombinants have a larger bilayer repeat distance than maximally hydrated DML, attributable to increased intercalation of water between the bilayers. In a DML/PLA recombinant containing 6.3 wt% protein at 37°C, a single phase is present in which the protein is homogeneously distributed throughout the bilayers. At 10°C, two phases are present: a DML/PLA complex phase, and a small amount of free DML.

PROTEIN-CATALYZED EXCHANGE OF PHOSPHATIDYLCHOLINE BETWEEN SONCIATED LIPOSOMES AND MULTILAMELLAR VESICLES. P.E. DiCorleto and D.B. Zilversmit (Div. of Nutr. Sci, Div. of Biol. Sci., Cornell Univ., Ithaca, NY). *Biochemistry* 16, 2145–50 (1977). Phospholipid exchange protein from beef heart or beef liver does not catalyze the transfer of phosphatidylcholine from multilamellar vesicles of phosphatidylcholine. Certain combinations of phospholipids, however, do yield multilamellar vesicles that will exchange phosphatidylcholine with liposomes in the presence of exchange protein. Multilamellar vesicles of phosphatidylcholine: phosphatidylethanolamine: cardiolipin (70:25:5, mol %) can be used in place of mitochondria or erythrocyte ghosts as an improved acceptor particle in the study of liposome structure with phospholipid exchange proteins. These multilamellar vesicles act as a well-defined reservoir of unlabeled phosphatidylcholine with 7% exchangeable phospholipid.

PROPERTIES OF MIXED VESICLES OF LECITHIN: CHOLESTEROL UP TO A 1:2 MOLAR RATIO. B. Jundberg (The Research Institute of the Åbo Akademi Foundation, Åbo Akademi SF Åbo, Finland). *Chem. Phys. Lipids* 18, 212–20 (1977). The cholesterol solubilizing capacity of lecithin vesicles was studied and some physicochemical properties of the resulting mixed vesicles were investigated. The maximum association of ultrasonicated cholesterol and lecithin was found to be a cholesterol/lecithin molar ratio of 2:1, with a limiting concentration of colloidal lipid of approximately 34 mg/ml. The 2:1 dispersions were found to be rather stable with no change in cholesterol/lecithin ratios for long periods. The mixed 2:1 cholesterol/lecithin vesicles were separated by Sepharose 4 B chromatography to obtain homogeneous preparations. The homogeneity was further tested by analytical ultracentrifugation and electron microscopy. Light-scattering measurements showed an increase in particle weight with increasing cholesterol proportion. ^{1}H- and ^{13}C-NMR studies demonstrated an additional broadening, especially of chain resonances, when going from a cholesterol/lecithin molar ratio of 1:1 to 2:1.

MECHANISM OF CHOLESTEROL SIDE-CHAIN CLEAVAGE. THE ENZYMIC HYDROPEROXIDE-GLYCOL REARRANGEMENT OF THE EPIMERIC 20-HYDROPEROXYCHOLESTEROLS IN ^{18}O-ENRICHED WATER. J.E. van Lier, J. Rousseau, R. Langlois and G.J. Fisher (Biochem. Labs., Dept. of Nuclear Med. and Radiobiology, Centre Hospitalier Universitaire, Sherbrooke, Quebec, Canada). *Biochim. Biophys. Acta.* 487, 395–9 (1977). Incubation of 20α-hydroperoxycholesterol (I) and its 20β-isomer, 20β-hydroperoxy-20-*iso*cholesterol (II) with adrenocortical mitochondrial preparations in the absence of molecular oxygen, in normal and ^{18}O-enriched water, gave 20α,22*R*-dihydroxycholesterol (III) from I and 20β,21-dihydroxy-20-*iso*cholesterol (IV) from II. Mass spectral analysis of the persilylated glycol products III and IV showed no uptake of ^{18}O, indicating that the oxygen atoms of the C20-, C22- and C21-hydroxyl groups originated from the 20-hydroperoxy groups of the substrates. This work supports the suggestion that a ferryl-atomic oxygen complex is the intermediate in the enzymic oxidative reactions of cholesterol side-chain cleavage.

STUDIES ON SPIN-LABELLED EGG LECITHIN DISPERSION. L.M. Gordon and R.D. Sauerheber (Dept. of Chem., San Diego State Univ., San Diego, Calif.) *Biochim. Biophys. Acta* 466, 34–43 (1977). ESR spectra of egg lecithin dispersions labelled with 5-nitroxide stearic acid are recorded with a 50 G field sweep, and also with a new technique which "expands" the spectrum by (1) recording pairs of adjoining peaks with a smaller field sweep and (2) superposing the common peaks. The expansion technique improves the precision of the order parameters determined from the hyperfine splitting measurements, and may prove useful in future spin label membrane studies. Approximate order parameters are derived to describe the fluidity of fatty acid spin-labelled membranes in those cases where either the inner or outer hyperfine extrema are not well defined. The ability of these expressions to measure the fluidity of labelled egg lecithin dispersions for the temperature range 14–42°C is examined.

LACK OF FATTY ACID SPECIFICITY IN THE LIPOLYSIS OF OLIGO- AND POLYUNSATURATED TRIACYLGLYCEROLS BY MILK LIPOPROTEIN LIPASE. N. Morley and A. Kuksis (Banting and Best Dept. of Med. Res., Univ. of Toronto, Toronto, Ontario, M5G 1L6, Canada). *Biochim. Biophys. Acta.* 487, 332–42 (1977). Native soybean and rapeseed oils and native and rearranged cod liver and peanut oils were subjected to partial hydrolysis with milk lipoprotein lipase and the fatty acid composition and molecular association in the substrates and lipolysis products were determined. In both native and rearranged oils the lack of significant differences in the fatty acid composition and molecular association between the residual and total triacylglycerols suggested that all triacylglycerols were attacked by the lipoprotein lipase at about the same rate. In general, the products of the rearranged oils closely resembled the original triacylglycerols in the fatty acid composition. It is concluded that lipoprotein lipase does not show any detectable specificity for the unsaturated and polyunsaturated fatty acids with double bonds located at carbons 3 to 19 from the carboxyl end of the fatty acid molecules. These findings are compatible with the possible binding of the substrate to lipoprotein lipase through atoms involved in the acyl ester groups of the triacylglycerol molecules.

A SENSITIVE RADIOENZYMATIC ASSAY FOR GLYCEROL AND ACYLGLYCEROLS. P.B. Schneider (Thorndike Mem. Lab. and Nuclear Med. Unit, Dept. of Med., Beth Israel Hosp. and Harvard Med. Schl., Boston, MA). *J. Lipid Res.* 18, 396–9 (1977). A sensitive radioenzymatic assay for glycerol and acylglycerols is described. The assay depends on the quantitative phosphorylation of glycerol to glycerophosphate by glycerol kinase using [γ-^{32}P]ATP as a substrate. The ^{32}P content of the formed glycerophosphate is determined and gives a measure

of the original glycerol content. Acylglycerols can be determined by prior hydrolysis to glycerol. The assay is sensitive to about 0.1 nmol of glycerol and can be extended to 100 nmol. The assay can be applied to the determination of acylglycerols separated by thin-layer chromatography in amounts as low as 0.5 nmol. The assay is particularly useful in the determination of the specific activity of ^{14}C- or ^{3}H-labeled glycerol moeities.

STRUCTURAL PHASE RELATIONSHIPS IN LECITHIN-CHOLATE-WATER SYSTEMS. W. Shankland (Faculte de Medecine, Universite de Paris 6, Pitie-Salpetriere, Paris Cedex, France) *Chem. Phys. Lipids* 19, 20–42 (1977). In the first part of this paper the anomalies and discontinuities observed in different physical properties of the lecithin-cholate isotropic phase (light scattering, sodium ion activity, viscosity, conductivity) as well as the results of an enzymatic study are explained in terms of partial aggregation which sets in when the intermicellar solution is "unsaturated" in cholate. It is proposed that this aggregation process in the isotropic phase is exactly the inverse process of the disintegration of the hexagonal phase when the latter is diluted. The mixed micelles had previously been shown to be in the form of a bimolecular disc; both the cylindrical elements of the hexagonal phase and the entities in the aggregation process are stocks of mixed micellar discs, the existence of both of these states being governed by the same delicate hydrophilic-hydrophobic balance. The variations of both the water layer between discs in the cylinder and of the intercylinder water volume were shown to be coherent with changes in lecithin concentration and the amount of water present, in agreement with the above mentioned balance.

THERMAL BEHAVIOR OF HUMAN PLASMA HIGH DENSITY LIPOPROTEIN. A.R. Tall, R.J. Deckelbaum, D.M. Small and G.G. Shipley (Dept. of Med., Boston Univ. Schl. of Med., Boston, MA). *Biochim. Biophys. Acta* 487, 145–53 (1977). Human plasma low density lipoprotein displays a reversible thermal transition between 20 and 40°C, due to a phase transition of its core cholesterol ester from a smectic to a more liquid-like state. To determine if the cholesterol ester of high density lipoprotein (HDL) displays similar thermal behavior, the human lipoprotein and its extracted lipid have been examined by differential scanning calorimetry, low angle X-ray scattering and polarizing microscopy. Neither HDL_2 **(*d*1.063–1.125 g/ml) nor HDL_3 (*d*1.125–1.21 g/ml) show thermal transitions between 0 and 60°C. By contrast cholesterol ester isolated from HDL and mixtures of cholesterol oleate and linoleate show reversible liquid crystalline transitions between 20 and 40°C. Following the thermal disruption of HDL, reversible liquid crystalline transitions of cholesterol ester can be seen by differential scanning calorimetry and polarizing microscopy, showing the presence of large domains of cholesterol ester. The high temperature behavior of HDL indicates that apoprotein A-1 is less important than apoprotein A-2 in maintaining the HDL apolar lipids in the form of a stable microemulsion.

MECHANISTIC INTERPRETATION OF THE INFLUENCE OF LIPID PHASE TRANSITIONS ON TRANSPORT FUNCTIONS. L. Thilo, H. Träuble and P. Overath (Max-Planck-Inst. fur Biologie, Tubingen, West Germany). *Biochem. J.* 16, 1283–90 (1977). In an attempt to understand the mechanism by which a structural change of membrane lipids affects transport functions, the temperature dependence of transport rate has been measured to below the low temperature end of the fluid ⟷ ordered phase transition of the membrane lipids. The unsaturated fatty acid requiring *Escherichia coli* strain T105 was supplemented with either *trans*-Δ^9-octadecenoate or *trans*-Δ^9-hexadecenoate or supplemented with and subsequently starved for *cis*-Δ^9-octadecenoate. These experiments are interpreted in terms of a partitioning of transport proteins between ordered and fluid domains which is described by a lateral distribution coefficient, k. This distribution coefficient varies with the membrane lipid composition as well as with the transport system.

THE TRANSLOCATION OF Ca^{2+} ACROSS PHOSPHOLIPID BILAYERS INDUCED BY A SYNTHETIC NEUTRAL Ca^{2+}-IONOPHORE. P. Vuilleumier, P. Gazzotti, E. Carafoli and W. Simon (Labs. of Organic Chem., and Biochem., Swiss Federal Inst. of Tech., Zurich, Switzerland). *Biochim. Biophys. Acta* 467, 12–8 (1977). The effect of a neutral synthetic Ca^{2+}-ligand, which induced selective Ca^{2+} transport in electrodialysis experiments in bulk membranes, on the Ca^{2+} permeability of phospholipid bilayers has been investigated. The ligand is able to promote the transport of Ca^{2+} across synthetic phopholipid bilayers and can therefore be classified as a Ca^{2+}-ionophore. Its activity is enhanced by the uncoupler carbonyl cyanide *p*-trifleuoromethoxyphenylhydrazone (FCCP). The efficiency of the neutral carrier-mediated Ca^{2+} transport is rather low as compared with that of the charged Ca^{2+}-ionophore X537A. The Ca^{2+} selectivity of the neutral ionophore is decreased by its incorporation in the low dielectric ambient of the phopholipid bilayer.

A LINEAR FUNCTION FOR THE MELTING BEHAVIOR OF LIPIDS. H.M. Zacharis (Southern Regional Research Center, New Orleans, La.) *Chem. Phys. Lipids* 18, 221–31 (1977). The melting behavior of a variety of saturated long chain compounds is shown to be related to hydrocarbon chain length by the equation $TN = C_o + T\infty N$ where T is the absolute melting temperature, and N is the number of long chain carbon atoms. The constants Co and T∞ are determined graphically or analytically from TN vs. N data. The linear relationship, derived from fundamental thermodynamic principles, is empirically satisfied. For each homologous series considered, coefficients of the equation provide a rational means for correlation and comparison with other polymorphs and indicate the relative importance of chain length, chain parity (even or odd), and headgroup polarity to melting behavior.

REGULATION OF STEROL SYNTHESIS IN 15 TISSUES OF RAT. ROLE OF RAT AND HUMAN HIGH AND LOW DENSITY PLASMA LIPOPROTEINS AND OF RAT CHYLOMICRON REMNANTS. J.M. Andersen and J.M. Dietschy (Dept. of Internal Med., Univ. of Texas Health Sci. Ctr., Dallas, Texas). *J. Biol. Chem.* 252, 3652–9 (1977). These studies were undertaken to identify which, if any, of the circulating lipoproteins regulates sterol synthesis in various extrahepatic organ systems. Following administration of 4-aminopyrazolo [3,4-*d*]pyrimidine to rats for 72 h, the plasma cholesterol level fell to < 10 mg · dl^{-1} and sterol synthesis in 9 of 15 extrahepatic tissues increased 1.8- to 34.9-fold. Infusion of a preparation of either human or rat "whole" plasma lipoproteins, i.e. a fraction containing all plasma lipoproteins with a density of < 1.230, over a 40-h period to similarly treated animals suppressed synthesis in nearly all of these tissues, indicating that one or more of the specific lipoproteins in this fraction were responsible for suppression of synthesis in at least these tissues. On the basis of these studies and other published reports, three functionally different types of lipoprotein feedback regulation of sterol synthesis are proposed: regulation of hepatic cholesterogenesis by chylomicron remnants; regulation of sterol synthesis in a number of nonendocrine, extrahepatic tissues by LDL; and regulation of sterol synthesis in ovary and adrenal gland principally by HDL.

REGULATION OF STEROL SYNTHESIS IN 16 TISSUES OF RAT. J.M. Andersen and J.M. Dietschy (Dept. of Internal Med., Univ. of Texas Health Sci. Ctr., Dallas, Texas). *J. Biol. Chem.* 252, 3646–51 (1977). The rate of cholesterol synthesis in the liver is affected by many physiological variables. This study was undertaken to measure systematically the effect of these same variables on sterol synthesis in 15 extrahepatic tissues of the rat. Diurnal light cycling caused a significant change in the rate of cholesterogenesis in the liver but had no effect on the rate of sterol synthesis in any of the extrahepatic tissues tested. Fasting markedly reduced the rate of hepatic cholesterol synthesis and resulted in lesser degrees of reduction in the rate of sterol synthesis in kidney, intestine, and several other tissues. Experimental manipulations designed to alter markedly the amount of bile acid circulating in the enterohepatic circulation (such as biliary diversion, biliary obstruction, and cholestyramine or bile acid feeding) had no effect on the rate of sterol synthesis in any tissue assayed except the liver, intestine, and adrenal gland.

STIMULATION OF HEPATIC LIPOGENESIS BY EICOSA-5,8,11,14-TETRAYNOIC ACID IN MICE FED A HIGH LINOLEATE DIET. S. Abraham, H. McGrath and G.A. Rao (Bruce Lyon Memorial Res. Lab., Children's Hos. Med. Center of Northern Calif., Oakland, CA) *Lipids* 12, 446–9 (1977). Liver slices, from mice fasted for one day and then refed for three days either a 15% corn oil diet or a 15% corn oil diet containing eicosa-5,8,11,14-tetraynoic acid (TYA), were incubated with (1-^{14}C) acetate or (^{3}H)H_2O to determine lipogenic capacity. Dietary TYA produced a twofold stimulation in fatty acid and cholesterol synthesis. TYA also caused an increase in the relative proportion of linoleate ($C_{18:2}$) and a decrease in that of arachidonate ($C_{20:4}$) in liver. Thus, (a) despite high levels of $C_{18:2}$, hepatic lipogenesis can be increased, and (b) even short term feeding of TYA can alter the hepatic fatty acid composition presumably by inhibition of arachidonate synthesis from linoleate.

RECOGNITION OF DIFFERENT POOLS OF PHOSPHATIGLYCEROL IN INTACT CELLS AND ISOLATED MEMBRANES OF ACHOLEPLASMA LAIDLAWII BY PHOSPHOLIPASE A_2. E.M. Bevers, S.A. Singal, J.A.F. Op den Kamp, and L.L.M. van Deenen (Lab. of Biochem., St. Univ. Utrecht, Utrecht, The Netherlands). *Biochem. J.* **16**, 1290–5 (1977). Phospholipase A_2 (EC 3.1.1.4) from pig pancreas hydrolyzes phosphatidylglycerol in intact cells and isolated membranes of *Acholeplasma laidlawii*. Complete degradation of phosphatidylglycerol in intact cells at 37°C does not result in lysis as shown by the retention of intracellular K^+ ions and the cytoplasmic glucose-6-phosphatase, as well as the inability to detect activity of membrane-bound intracellular NADH-oxidase. *A. laidlawii* was grown on linoleic acid. Phospholipase A_2 treatment of these cells at 5°C, at which temperature the lipids are still in the liquid-crystalline state, results in a rapid breakdown of 50% of the phosphatidylglycerol. The residual phosphatidylglycerol can be hydrolyzed only at elevated temperatures and at much smaller rates, depending strongly on the incubation temperature.

LEUCINE CATABOLISM AND CO_2 FIXATION INTO FATTY ACIDS BY TETRAHYMENA. M. Borowitz, G. Raugi, T. Liang and J.J. Blum (Dept. of Derma., Univ. Oregon, Portland, OR). *J. Biol. Chem.* **252**, 3402–7 (1977). The pathway by which aerobic CO_2 fixation into fatty acids takes place in *Tetrhymena pyriformis* is investigated. It is found that leucine but not other amino acids stimulates this CO_2 fixation 7-fold and that isovalerate, (a precursor of isovaleryl coenzyme CoA, an intermediate in leucine degradation) stimulates $H^{14}CO_3$ incorporation into lipid 15-fold. These results are consistent with a quantitatively important fixation of CO_2 via β-methylcrotonyl-CoA carboxylase. The substrate for this fixation can either be exogenous HCO_3, or CO_2 produced endogenously from the oxidation of leucine or pyruvate. Interpretation of these results in light of what is known about acetyl-CoA metabolism in *Tetrahymena* suggests that the former reaction occurs in the mitochondria while the latter takes place in the cytosol or peroxisomes.

LIPROTEIN REGULATION OF 3-HYDROXY-3-METHYLGLUTARYL COENZYME A REDUCTASE ACTIVITY IN RAT LIVER CELL CULTURES. J.L. Breslow, D.A. Lothrop, A.W. Clowes, and S.E. Lux (Dept. of Pediatrics and Pathology, Harvard Med. Schl., Boston, Mass.). *J. Biol. Chem.* **252**, 2726–33 (1977). A primary cell culture technique was used to study the effects of lipoproteins on rat hepatocyte 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase activity. In this system, lipoproteins prepared from normocholesterolemic rat and human plasma, including low density lipoproteins, did not inhibit hepatocyte HMG-CoA reductase activity whereas very low density lipoproteins and high density lipoproteins isolated from the same sources were stimulatory. Primary cell culture of rat hepatocytes appears to be a useful system in which to study cholesterol metabolism in the liver.

PHOSPHOLIPID SYNTHESIS IN ISOLATED FAT CELLS. R. Coleman and R.M. Bell (Depts. of Biochem. and Pediatrics, Duke Univ. Med. Center, Durham, North Carolina). *J. Biol. Chem.* **252**, 3050–6 (1977). Diacylglycerol cholinephosphotransferase (EC 2.7.8.2) and diacylglycerol ethanolaminephosphotransferase (EC 2.7.8.1) activities were investigated in microsomes from isolated rat fat cells. Assays based on the conversion of CDP-[^{14}C]choline or CDP-[^{14}]ethanolamine to phosphatidylcholine or phosphatidylethanolamine utilized ethanol-dispersed diacylglycerols and 1 to 5 μg of protein. Cholinephosphotransferase and ethanolaminephosphotransferase activities had similar dependences on $MgCl_2$ and pH, and were inhibited similarly by $CaCl_2$, organic solvents, Triton X-100, Tween 20, and dithiothreitol. Ethylene glycol bis(β-amino-ethyl ether)-*N,N,N',N'*-tetraacetic acid stimulated both activities similarly. These data provide the first characterization of the cholinephosphotransferase and ethanolaminephosphotransferase activities from adipose tissue or fat cells and the first systematic investigation of the diacylglycerol dependences of these activities from any tissue. Taken as a whole, the data strongly suggest that the cholinephosphotransferase and ethanolaminephosphotransferase are separate microsomal enzymes.

RAT INTESTINAL GLYCOLIPIDS III. FATTY ACIDS AND LONG CHAIN BASES OF GLYCOLIPIDS FROM VILLUS AND CRYPT CELLS. J.F. Bouhours and R.M. Glickman (Thorndike Lab., Dept. of Med., Harvard Med. Schl., Boston, MA). *Biochim. Biophys. Acta* **487**, 51–60 (1977). Previous studies from this laboratory have demonstrated a striking difference in rat intestinal glycolipids between differentiated villus cells and immature crypt cells. Villus cells contained proportionally greater amounts of glucosylceramide and hematoside while crypt cells were deficient in hematoside, but contained proportionally greater amounts of trihexosylceramide. In order to further elucidate possible differences between villus and crypt cell glycolipids, a study of the sphingosine and fatty acids of rat intestinal glycolipids was conducted. These results suggest that hematoside and trihexosylceramide, respectively abundant in villus and in crypt cells, may be derived from a different lactosylceramide precursor and further underscore differences in villus and crypt cell glycolipid synthesis.

THE POLAR LIPIDS OF GROUP B STREPTOCOCCI II. COMPOSITION AND POSITIONAL DISTRIBUTION OF FATTY ACIDS. W. Fischer (Inst. of Physio. Chem., Univ. Erlangen-Nurnberg, Erlangen, West Germany). *Biochim. Biophys. Acta* **487**, 89–104 (1977). The polar lipids from group B *Streptococci* have been isolated. Unless overlapping fractions are worked up on each step molecular species can be lost resulting in a fatty acid composition different from the original one. The lipids were shown to be 1(3),2-diacyl-3(1)-0-α-D-glucopyranosyl-*sn*-glycerol, 1(3),2-diacyl-3(1)-0-[α-D-glucopyranosyl-(1,2)-0-α-D-glucopyranosyl]-*sn*-glycerol, 1,2-diacyl-*sn*-glycero-3-phospho-1'-*sn*-glycerol, lysylphosphatidylglycerol and 1',3'-bis(1,2-diacyl-*sn*-glycero-3-phospho-glycerol. The stereochemical configuration of the phospholipids was achieved by combinations of chemical degradations. The uniform fatty acid make-up in all polar lipids favours the hypothesis that their diacylglycerol portions are derived from a common phosphatidic acid precursor with negligible postsynthetic rearrangements of the constituent fatty acids.

SUBCELLULAR DISTRIBUTIONS OF LIPIDS IN CULTURED BHK CELLS: EVIDENCE FOR THE ENRICHMENT OF LYSOBISPHOSPHATIDIC ACID AND NEUTRAL LIPIDS IN LYSOSOMES. J. Brotherus and O. Renkonen (Laboratory of Lipid Res., Dept. of Biochem., Univ. of Helsinki, Helsinki, Finland) *J. Lipid Res.* **18**, 191–202 (1977). Homogenates of cultured hamster fibroblasts (BHK 21 cells) were fractionated by differential centrifugation into six main fractions: nuclear, mitochondrial, light mitochondrial, microsomal, soluble, and floating. The contents of several lipids and some marker enzymes were measured. According to the enzyme distributions, lysosomes were enriched in the floating fraction more than tenfold relative to phospholipid. We conclude that lysobisphosphatidic acid was enriched in the lysosomes of the BHK cells and the lysosomes also contained variable amounts of neutral lipids in the form of intralysosomal droplets.

UTILIZATION OF CHOLINE FROM CRUDE SOYBEAN LECITHIN BY CHICKS. 2. ABSORPTION MEASUREMENTS. P. Budowski, I. Kafra, and D. Sklan (Faculty of Agr., Hebrew Univ. of Jerusalem, Revohot 76–1000, Israel) *Poult. Sci.* **56**, 754–7 (1977). Absorption of choline was investigated in 30-day-old chicks fed a casein-glucose diet supplemented with either choline chloride or crude soybean lecithin and containing $^{91}YCl_3$ as a non-absorbable reference substance. The choline concentration, expressed as choline chloride, was 0.101% in both experimental diets. Net rates of absorption or secretion were computed from the amounts of ^{91}Y and choline assayed in the contents of different segments of the small intestine. Net secretion of total choline into the duodenum was over twice the daily choline intake in both treatments. Most of the choline was absorbed in the upper small intestine. Of the small amounts remaining in the lower jejunum, significantly more choline was absorbed by the choline-fed chicks than by the lecithin-treated birds, but the rates of secretion into the duodenum, the rates of absorption from the upper small intestine and the overall apparent absorbability were not significantly different in the two treatments. Absorption measurements thus indicate that crude soybean lecithin may replace choline chloride as a source of choline.

RESPONSE OF ADIPOSE TISSUE LIPOPROTEIN LIPASE TO FASTING IN THE CHICKEN AND THE RAT—A SPECIES DIFFERENCE. J.D. Benson and A. Bensadoun (Div. of Nutr. Sci. and Div. of Biol. Sci., Cornell Univ., Ithaca, N.Y.). *J. Nutr.* **107**, 990–7 (1977). Studies were conducted to assess the influence of extraction procedures on the response of adipose tissue lipoprotein lipase (LPL) to fasting in two species, the rat and the chicken. In both cases, lipoprotein lipase was extracted more efficiently (twofold increase) from acetone powders of adipose tissue by heparin (50 units/ml) than by 25 mM NH_4OH–NH_4Cl. With chicken acetone powders, buffered saline solutions (1.2 M NaCl, 0.005 M Na barbital buffer, pH 6.5) were as effective as heparin solutions. Chickens fasted for 48 hours exhibited higher plasma triglyceride hydrolase, lower plasma triglycerides and higher concentrations of plasma free

fatty acids than either fed or refed chickens. Total lipoprotein lipase activity of extracts of adipose tissue was not significantly affected by the nutritional state of the chicken. However, the heparin-stimulated release of LPL was significantly higher for adipose tissue slices of fed chickens than for those obtained from fasted birds. Rat serum triglyceride hydrolase activities did not differ in fed, fasted or refed groups, as it did in the chicken. Comparison of adipose tissue LPL levels in the three groups showed marked reduction of LPL in the fasted group. The method of extraction, although affecting the absolute quantities of enzyme extracted, did not affect the relative differences due to fasting or feeding.

ON THE RISE IN LOW DENSITY AND HIGH DENSITY LIPOPROTEINS IN RESPONSE TO THE TREATMENT OF HYPERTRIGLYCERIDAEMIA IN TYPE IV AND TYPE V HYPERLIPOPROTEINAEMIAS. L.A. Carlson, A.G. Olsson and D. Ballantyne (King Gustaf V Res. Inst. and the Dept. of Internal Med., Karolinska Inst. and Hosp., Stockholm, Sweden). *J. Atherosclerosis* 26, 603–9 (1977). In Type V hyperlipoproteinaemia the concentration of LDL and HDL cholesterol is low. When the hypertriglyceridaemia is normalized, either by diet or nicotinic acid, both LDL and HDL increase. While both the fall in VLDL and the rise in HDL may be beneficial from the point of view of atherosclerosis the rise in LDL may be harmful. There is at present no way to evaluate the effect of these complex changes. However, these findings stress the importance of considering changes in lipoprotein levels and not only in total serum triglycerides and cholesterol during treatment of hyperlipoproteinaemia.

PRE- AND POST-NATAL DEVELOPMENT OF LIPOPROTEIN LIPASE AND HEPATIC TRIGLYCERIDE HYDROLASE ACTIVITY IN RAT TISSUES. T. Chajek, O. Stein and Y. Stein (Lipid Res. Lab., Dept. of Med. B, Hadassah Univ., Hos., and Dept. of Experimental Med. and Cancer Res., Hebrew Univ.-Hadassah Med. Schl., Jerusalem, Israel) *Atherosclerosis* 26, 549–61 (1977). The ontogenic development of lipoprotein lipase and liver triglyceride hydrolase was studied in the rat. The enzyme activity measured in extrahepatic tissues fulfilled the criteria of lipoprotein lipase from the onset of measurable activity, i.e. it was inhibited by protamine and 1 *M* NaCl and showed requirement for serum and heparin for optimal activity. In the liver, measurable amounts of triglyceride hydrolase, active at pH 8.6 were detected 6 days prior to birth. However, till the fourth postnatal day about 50% of this activity was inhibited by NaCl and its sensitivity towards protamine was also higher than that of the enzyme in adult liver. Three patterns of development of enzymic activity were observed in extrahepatic tissues. These findings indicate that the regulation of the development of lipoprotein lipase activity in extrahepatic tissues is governed by local factors, which can differ even in the same type of tissue, as exemplified by the difference between inguinal and epididymal fat.

METABOLISM OF LIPID-LINKED *N*-ACETYLGLUCOSAMINE INTERMEDIATES. W.W. Chen and W.J. Lennarz (Dept. of Physiological Chem., The Johns Hopkins Univ. Schl. of Med., Baltimore, Md.) *J. Biol. Chem.* 252, 3473–9 (1977). The synthesis and metabolism of dolichol-linked *N*-acetylglucosamine intermediates have been studied using a membrane preparation from hen oviduct. An acetone powder preparation of membranes was found to catalyze synthesis of *N*-acetylglucosaminyl-(pyro)phosphoryldolichol in the presence of UDP-*N*-acetylglucosamine and dolichol phosphate. Incubation of purified *N*-acetylglucosaminyl(pyro)phosphoryldolichol with oviduct membranes and UDP-*N*-acetylglucosamine resulted in its conversion to *N,N'*-diacetylchitobiosyl(pyro)phosphoryldolichol. Upon incubation with oviduct membranes, purified *N,N'*-diacetylchitobiosyl(pyro)phosphoryldolichol was found to serve as a donor of its disaccharide unit to an endogenous protein with an apparent molecular weight of 25,000. These findings indicate that the saccharide moiety of *N,N'*-diacetylchitobiosyl-(pyro)phosphoryldolichol can be transferred to protein either directly, or after elongation to oligosaccharide-(pyro)phosphoryldolichol.

PREVALENCE OF CORONARY ARTERY DISEASE AND PERIPHERAL ARTERY DISEASE IN PATIENTS WITH DIFFERENT TYPES OF PRIMARY HYPERLIPIDEMIA. G. Crepaldi, R. Fellin, G. Briani, G. Baggio, E. Manzato and R. Veronese (Dept. of Internal Med., Div. of Gerontology and Metabolic Diseases, Univ. of Padua, I-35100 Padua, Italy) *Atherosclerosis* 26, 593–602 (1977). The prevalence of coronary artery disease (CAD) and peripheral artery disease (PAD) was studied in 280 (203 males, 77 females) patients with different types of primary hyperlipoproteinemia. In primary hyperbetalipoproteinemia the prevalence of CAD (45% for Type IIa and 47% for Type IIb) is significantly higher than that in the other types of hyperlipoproteinemia (38% for Type IV and 17% for Type V). On the other hand, PAD prevalence is much higher in hypertriglyceridemia (21% in Type IIb and 20% in Type V) than in hypercholesterolemia alone (9% in Type IIa). These results suggest that patients with Type IIb are exposed to the greatest risk as far as atherosclerotic complications are concerned. Moreover, the high frequency of PAD found in hypertriglyceridemia can be related to the high occurrence of diabetes in these patients. The effects of other major risk factors of atherosclerosis (smoking and hypertension) were also evaluated. Our results indicate that the association of hypercholestolemia and hypertension is more dangerous than the co-occurrence of hypercholesterolemia and smoking.

LOCALIZATION OF LIPASE-LIKE IMMUNOREACTIVITY IN PORCINE ADIPOSE, AORTIC AND MYOCARDIAL TISSUE. W. Nieuwenhuizen, J.J. Emeis and C.M. Van Sabben (Gaubius Inst., Health Res. Organization TNO, Herenstraat 5d, Leiden, The Netherlands) *Atherosclerosis* 27, 97–106 (1977). Lipase has been purified from pig adipose tissue and antibodies have been produced in rabbit. By indirect immunofluorescence and immunoenzyme techniques lipase-like immunoreactivity was demonstrated in the intima of pig aorta, in the endothelial cells of the myocardium and in plasma membranes of adipocytes and skeletal muscle cells. Lipase activity in extracts of some of these tissues was inhibited by the addition of anti-lipase antibodies. At least part of the immunoreactivity in the examined tissues is due to active lipases. ●