

Abstracts

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Biochemistry and nutrition

METABOLISM OF LINOLEIC ACID IN THE CAT. A.J. Sinclair, J.G. McLean and E.A. Monger (Dept. of Agri., Vet. Res. Inst., and Schl. of Vet. Sci., Univ. of Melbourne, Parkville, Victoria 3052, Australia) *Lipids* 14(11), 932-6 (1979). Cats fed a diet containing linoleate as the only polyunsaturated fatty acid showed extremely low levels of arachidonate in the plasma lipids, as well as an increase in linoleate, eicosadienoate and an unknown fatty acid. Administration of [$1-^{14}\text{C}$] linoleic acid and [$2-^{14}\text{C}$] eicosa-8,11,14-trienoic acid to cats showed that in the liver there was no conversion of the [$1-^{14}\text{C}$] 18:2 to arachidonate, whereas there was significant metabolism of [$2-^{14}\text{C}$] 20:3 to arachidonate. It was found when methyl- γ -linolenate was fed to cats that the level of 20:3 ω 6 and 20:4 ω 6 in the erythrocytes increased significantly. These results show that there is no significant Δ 6 desaturase activity in the cat, whereas chain elongation and Δ 5 desaturase enzymes are operative. The unknown fatty acid was isolated from the liver lipids and shown to be a 20-carbon fatty acid with 3 double bonds and which by gas liquid chromatography could be separated from 20:3 ω 9 and 20:3 ω 6. The presence of the Δ 5-desaturase activity and the results of the ozonolysis studies indicated that this unknown fatty acid was eicosa-5,11,14-trienoic acid.

ENHANCED LIPID PEROXIDATION IN LIVER MICROSOMES OF ZINC-DEFICIENT RATS. J.F. Sullivan, M.M. Jetton, H.K.J. Hahn and R.E. Burch (Medical Service, VA Medical Center, 4101 Woolworth Ave., Omaha, Nebraska 68105) *Am. J. Clin. Nutr.* 33(1), 51-6 (1980). The clinical association of decreased serum and hepatic zinc in patients with cirrhosis of the liver presumably arising from excess ethanol ingestion prompted a study of the activities of zinc and alcohol in experimental animals. The purpose of this study was to determine the effect of zinc deficiency upon lipid peroxidation in the liver. The effect of ethanol and zinc deficiency on lipid peroxidation was also evaluated. Rats were used in the experimental design, one group received a control diet, and one was maintained on a zinc-deficient diet. One-half of each group also received 3.85 g ethanol per kilogram body weight daily. Lipid peroxidation *in vivo* was determined by estimation of diene conjugation of microsomal lipids. The *in vitro* lipid peroxidation potential was measured by the generation of malonic dialdehyde by enzymatic as well as nonenzymatic reactions. Analysis of this data indicated that increased hepatic microsomal lipid peroxidation was associated with zinc deficiency whether using *in vivo* or *in vitro* indices of measurement. Review of data from individual animals indicated that the lowest levels of serum zinc were associated with increased hepatic content of phospholipids. The degree of lipid peroxidation in the zinc deficient animals was not increased by ingestion of alcohol.

ON THE MEMBRANE PHOSPHOLIPIDS AND THEIR ACYL GROUP PROFILES OF ADRENAL GLAND. G.Y. Sun (Sinclair Comparative Med. Res. Farm and Biochem. Dept., Univ. of Missouri, Columbia, MO 65211) *Lipids* 14(11), 918-24 (1979). The phospholipid composition and their acyl group profiles from subcellular fractions of guinea pig adrenal gland and the same fractions from the cortex and medulla of the bovine gland were compared. The phospholipids of guinea pig adrenal were enriched in diacylglycerophosphocholines (GPC) which comprised over 50% of the total phospholipids, but the proportions of ethanolamine and choline plasmalogens, sphingomyelin and diacylglycerophosphoserine were lower in guinea pig adrenals as compared to the bovine adrenals. In the bovine adrenal, sphingomyelin and diacyl-GPS were enriched in the medulla, whereas diacylglycerophosphoinositol were enriched in the cortex. Characteristic acyl group profiles were found associated with each type of the phosphoglycerides in adrenal membranes. However, acyl group profiles of the phosphoglycerides were not greatly different either between the bovine and guinea pig adrenal or with respect to the type of subcellular membranes isolated. The lysolecithin from bovine adrenal membranes contained mainly 16:0, 18:0 and 18:1 with only trace amounts of the polyunsaturated fatty acids.

KRABBE DISEASE: A GALACTOSYLSPHINGOSINE (PSYCHOSINE) LIPIDOSIS. L. Svennerholm, M.-T. Vanier, and J.E. Månsson (Dept. of Neurochem., Psychiatric Res. Centre, St. Jörgen Hospital, Univ. of Gothenburg, S-422 03 Hisings-Backa, Sweden) *J. Lipid Res.* 21(1), 53-64 (1980). The primary genetic defect underlying Krabbe disease or globoid cell leukodystrophy is considered to be a deficiency of galactosylceramide- β -galactosidase. In the present study of the brains from 18 patients who had died from Krabbe disease at 7-37 months of age, the concentration of galactosylceramide of cerebral and cerebellar white matter was severely reduced to 10-20% of that in age-matched controls. The lowest values were found in the most long-standing cases. The enzyme has a broad specificity and it normally also degrades galactosylsphingosine. Because of competitive inhibition by the accumulated galactosylceramide its lysosomal hydrolysis will be blocked. The concentration of psychosine will steadily increase and reach toxic levels and kill the oligodendroglial cells. This results in an arrest of the galactosylceramide biosynthesis. Therefore, we feel that galactosylsphingosine and not galactosylceramide is the primary storage substance in the brain in Krabbe disease and that the disease is a psychosine lipidosi.

THE EFFECT OF VARIOUS DIETS ON ATHEROGENESIS IN RHESUS MONKEYS. D. Vesselinovitch, R.W. Wissler, T.J. Schaffner and J. Borenstajn (Dept. of Pathology and Specialized Center of Res. on Atherosclerosis, The Univ. of Chicago, Chicago, IL 60637) *Atherosclerosis* 35(2), 189-207 (1980). The present study compares the production of lesions induced by different dietary regimens. Eighteen male rhesus monkeys were divided into three groups and fed according to varying dietary patterns for 12 months. Although a wide spectrum of lesions was observed in each dietary group, plaques with characteristics specific for each diet were predominant. The first group was fed a diet containing 25% fat as a 1:1 mixture of butterfat and coconut oil and 2% cholesterol. For the second group, the same ratio was alternated at 2-month intervals with a cholesterol-free diet containing 25% corn oil. Animals in the third group were fed a diet consisting of 2% cholesterol and 25% peanut oil. The latter group was subdivided into two subgroups of 3 animals each; one subgroup received peanut oil "contaminated" with aflatoxin. All diets produced hyperlipemia and hyperlipoproteinemia. The highest levels of serum cholesterol were recorded in animals fed a diet enriched by a mixture of butterfat, coconut oil and cholesterol continuously.

INCREASED SYNTHESIS OF PHOSPHATIDYLCHOLINE BY RAT LUNG FOLLOWING PREMATURE BIRTH. P.A. Weinholt, M.M. Quade, T.B. Brozowski and D.A. Feldman (Veterans Admin. Med. Center and Dept. of Biol. Chem., Univ. of Michigan, Ann Arbor, MI 48105) *Biochim. Biophys. Acta* 617(1), 76-84 (1980). Pregnant rats were delivered prematurely at 20 days and 21 days gestation. The survival was $52 \pm 3\%$ at 20 days gestation and 100% at 21 days gestation. [$\text{Me-}^{14}\text{C}$] Choline incorporation into phosphatidylcholine increased by 60% during the 3 h of survival after delivery at 20 days gestation. The increase in incorporation occurred during the first hour of survival. [$\text{Me-}^{14}\text{C}$] Choline incorporation also increased to a lesser extent following survival at 21 days gestation. The incorporation after 3 h of survival at 20 days and 21 days gestation was similar to that obtained with adult lung slices. The pattern of D-[$\text{U-}^{14}\text{C}$] glucose incorporation following survival at 20 and 21 days gestation is similar to that obtained with adult lung slices. Dexamethasone treatment of mothers at 17 and 18 days gestation caused [$\text{Me-}^{14}\text{C}$] choline incorporation to increase to adult values at 20 days gestation but not at 19 days or 21 days. We conclude that an adaptive mechanism exists which produces specific biochemical changes in lung metabolism following premature birth. This mechanism is functional in the rat as early as 2 days prior to normal term.

THE EFFECT OF THE BASAL DIET ON THE UTILIZATION OF FAT AS A SOURCE OF TRUE METABOLIZABLE ENERGY, LIPID, AND FATTY ACIDS. I.R. Sibbald and J.K.G. Kramer (Animal Res. Inst., Agric. Canada, Central Exp. Farm, Ottawa, Ontario, Canada K1A 0C6) *Poult. Sci.* 59(2), 316-24 (1980). An experiment was made to investigate the effect of diet composition

on the utilization of beef tallow by adult cockerels. The birds were starved for 24 hr and then force-fed 1 of 3 basal diets (30 g) supplemented with 0, 1.5, 3.0, or 4.5 g of beef tallow. Excreta voided in the subsequent 24 hr was collected. True metabolizable energy (TME) and lipid availability values were measured. The TME value of tallow differed between basal diets and with the level of tallow input. A corn basal permitted greater TME values than a wheat basal but a supplement of soybean lecithins made the wheat basal as effective as the corn basal. Linoleic acid and phospholipid concentrations in the diet appeared to influence tallow utilization. The basic procedures of the TME bioassay permitted measurement of total lipid and fatty acid availability data. Lipid availability paralleled the TME data. Total lipid utilization ranged from 78 to 93% while the utilization of individual fatty acids varied both between acids and within acids between dietary treatments. Unsaturated fatty acids were well utilized (85 to 96%) while saturated acids were less well utilized (57 to 90%), particularly at high levels of input.

THE EFFECT OF DIETARY PROTEIN ON THE BIOLOGICAL ACTIVITY OF CHOLECALCIFEROL AND ITS METABOLITES IN THE RACHITIC RAT. N. Raghuramulu and H.F. DeLuca (Dept. of Biochem., College of Agric. and Life Sciences, Univ. of Wisconsin, Madison, WI 53706) *J. Nutr.* 110(1), 28-34 (1980). Rats maintained on low-protein rachitogenic diets show increased intestinal calcium transport, serum inorganic phosphate and increased endochondral calcification responses to calciferol, 25-hydroxycholecalciferol (25-OH-CC) and 1,25-dihydroxycholecalciferol (1,25-(OH)₂CC). Because enhanced responses are observed with 1,25-dihydroxycholecalciferol as well as its precursors, it is unlikely that the effect of dietary protein is mediated by a change in the calciferol-25-hydroxylase and the 25-hydroxycholecalciferol-1 α -hydroxylase. Instead, it appears that either metabolism of 1,25-dihydroxycholecalciferol or its activity in the target organs is affected.

EFFECTS OF SATURATED AND POLYUNSATURATED FAT DIETS ON THE CHEMICAL COMPOSITION AND METABOLISM OF LOW DENSITY LIPOPROTEINS IN MAN. J. Shepherd, C.J. Packard, S.M. Grundy, D. Yeshurun, A.M. Gotto, Jr., and O.D. Taunton (Dept. of Med., Baylor College of Med., and the Methodist Hospital, Houston, TX 77030) *J. Lipid Res.* 21(1), 91-9 (1980). This study examined the effects of dietary saturated and polyunsaturated fat on the chemical composition and metabolism of low density lipoproteins (LDL) in eight normal male subjects. The influence of these diets on fecal sterol excretion was also measured in four of the subjects. Overall, polyunsaturated fat feeding produced an enrichment in linoleate with reciprocal changes in palmitate, stearate, and oleate which affected triglycerides more than cholesteryl esters and phospholipids. The above changes in LDL composition were associated with alterations in the metabolism of LDL apoprotein (apoLDL). The polyunsaturated fat diet did not cause a consistent change in fecal neutral or acidic steroid excretion. We conclude that the hypocholesterolemic action of polyunsaturated fat diets is affected by multiple mechanisms whose expression may vary from patient to patient.

THE ACTION OF LIPOXYGENASE-1 ON FURAN DERIVATIVES. R.F. Boyer, D. Litts, J. Kostishak, R.C. Wijesundera and F.D. Gunstone (Dept. of Chem., Hope College, Holland, MI 49423) *Chem. Phys. Lipids* 25(3), 237-46 (1979). Several 2,5-disubstituted furans, which are known to react with peroxyacids, singlet oxygen and other active forms of oxygen were tested as potential inhibitors, co-oxidants, or substrates for soybean lipoxygenase. The furan, 10,13-epoxy-octadeca-10,12-dienoic acid, methyl ester (IV) was converted by lipoxygenase or singlet oxygen or peroxyacid to the acyclic product, methyl 10,13-dioxo-octadec-11-enoate. Apparently furan IV is able to interact with an active site of lipoxygenase ($K_m=220 \mu\text{M}$). 2,5-Dimethylfuran (I), 2,5-diphenylfuran (II) and 3-(5'-methyl-2'-furyl)propenoic acid (III) were neither substrates nor inhibitors of lipoxygenase activity. Lipoxygenase-catalyzed oxidation of furan (IV), which is inhibited by hydroquinone, is explained by a mechanism involving lipoxygenase-superoxide complex and furan-radical intermediates. Also described is the selective cleavage of furan rings by *m*-chloroperoxybenzoic acid to yield the 1,4-diketoethylene functional system.

CHLORINATED HYDROCARBON-CELL MEMBRANE INTERACTIONS STUDIED BY THE FLUORESCENCE QUENCHING OF CARBAZOLE-LABELED PHOSPHOLIPIDS: PROBE SYNTHESIS AND CHARACTERIZATION OF THE QUENCHING METHODOLOGY. J.R. Lakowicz and D. Hogen (Gray Freshwater Bio. Inst., Dept. of Biochem., Univ. of Minn., P.O. Box 100, Navarre, MN 55392, U.S.A.) *Chem. Phys. Lipids* 26(1), 1-40 (1980). In recognition of the need to understand better the interactions of the chlorinated hydrocarbon insecticides with cell membranes we investigated the use of fluorescence quenching of membrane-bound

fluorophores by these chlorinated hydrocarbons. An extensive survey of potential fluorophores identified the N-alkyl derivatives of carbazole as being especially suitable fluorophores. The fluorescence emission of these derivatives is quenched by a wide variety of commonly-used chlorinated hydrocarbons. This quenching is collisional and does not result in significant photodecomposition. When incorporated into lipid bilayers, the fluorescence lifetime of these carbazole-labeled phospholipids reveals the collisional frequency between the fluorophore and the chlorinated hydrocarbon. As a result quenching of membrane-bound fluorophores may be used to measure: (1) the diffusional rate of the chlorinated hydrocarbon in the bilayer; (2) the lipid-water partition coefficient; (3) the maximum binding capacity of the membrane for the chlorinated hydrocarbon. Examples of all these measurements are given, and the fluorometric results are confirmed by direct chemical analysis.

MECHANISM OF THE SPONTANEOUS TRANSFER OF PHOSPHOLIPIDS BETWEEN BILAYERS. M.A. Roseman and T.E. Thompson (Dept. of Biochem., Uniformed Services Univ. of Health Sci., Bethesda, MD 20014) *Biochemistry* 19(3), 439-44 (1980). A fluorescent phospholipid, 1-palmitoyl-2-pyrenedecanoylphosphatidylcholine, was used to study the mechanism of spontaneous phospholipid transfer between single-walled phospholipid vesicles. The half-time for transfer of this molecule between vesicles of dimyristoylphosphatidylcholine at 36°C is 13 h if flip-flop is negligible or 24 h if flip-flop is faster than intervesicle exchange. The half-time is unaffected by the concentration of acceptor vesicles, which indicates that transfer of label takes place by diffusion of monomers or micelles through the aqueous phase rather than by collision of vesicles. These results are compared with previous studies of spontaneous lipid transfer.

INFLUENCE OF MONOVALENT AND DIVALENT CATIONS ON THE SURFACE AREA OF PHOSPHATIDYL-GLYCEROL MONOLAYERS. K. Toko and K. Yamafuji (Dept. of Electronics, Kyushu Univ., Fukuoka 812, Japan) *Chem. Phys. Lipids* 26(1), 79-99 (1980). A theory is presented on the electrostatic properties of the surface area of phosphatidylglycerol monolayers spreading at an air-water interface in the presence of monovalent and divalent cations. In the present theory, the adsorption of monovalent and divalent cations to the membranes is taken into account, besides the dissociation of protons, as a possible cause of the change of surface charge density with the variation of pH or ion concentrations. It is also pointed out that, in the presence of structure-making ions such as Li⁺ and Na⁺, the nearest-neighbour interactions between proton dissociation sites become important for the monolayers in the gel state to yield a sharp expansion of the surface area with the increase of pH. The present theory shows quantitative agreements with previously-observed data.

THE RELATIVE RESPONSE OF HEPATIC LIPIDS IN THE RAT TO GRADED LEVELS OF DIETARY MYO-INOSITOL AND OTHER LIPOTROPES. D.B. Andersen and B.J. Holub (Dept. of Nutr., College of Bio. Sciences, Univ. of Guelph, Guelph, Ontario, Canada) *J. Nutr.* 110(3), 496-504 (1980). Hepatic triglyceride (TG) accumulation appeared to vary inversely up to a point with the level of dietary myo-inositol in the absence of dietary choline, and each of the major fatty acids of liver TG was similarly affected. Although the comparison of the major fatty acids changed little, 16:1 was generally higher and 18:0 lower in the liver TG of myo-inositol-deficient rats. Furthermore, both the level and the fatty acid composition of non-esterified fatty acids (NEFA) in liver changed dramatically. Supplementation of diets containing myo-inositol or choline with DL-methionine (0.2% by weight) did not significantly influence hepatic TG concentrations. Even when choline was present in the diet at the National Research Council (NRC) requirement (5.4 mmol/kg diet), supplementary myo-inositol decreased further liver TG levels below the levels achievable with choline alone. Dietary myo-inositol was as efficient as choline on a molar basis in preventing the accumulation of TG in liver when compared at 5.4 mmol/kg diet.

MYO-INOSITOL-RESPONSIVE LIVER LIPID ACCUMULATION IN THE RAT. D.B. Andersen and B.J. Holub (Dept. of Nutr., College of Bio. Science, Univ. of Guelph, Guelph, Ontario, Canada) *J. Nutr.* 110(3), 488-95 (1980). A nutritionally acceptable diet, with all nutrients at or above their known requirements, was found useful for studying the myo-inositol-responsive lipid accumulation in young rats. Succinyl sulfathiazole supplementation of the diet was judged unnecessary to produce increases in the concentration of hepatic triglycerides (TG). The TG accumulation appeared to decrease somewhat after reaching maximal levels under conditions of myo-inositol deficiency in young rats, whereas the accumulation was not observed in older animals. Both young male and female rats responded to dietary myo-inositol when supplemented at a level similar to the level of myo-inositol in the human diet.

EFFECTS OF CHOLIC ACID ON THE METABOLISM OF ENDOGENOUS PLASMA TRIGLYCERIDE AND ON BILIARY LIPID COMPOSITION IN HYPERLIPIDPROTEINEMIA. B. Angelin and B. Leijd (Dept. of Med., Karolinska Institutet at Serafimerlasarettet, Stockholm, Sweden) *J. Lipid Res.* 21(1), 1-9 (1980). Plasma lipids, endogenous triglyceride kinetics, and biliary lipid composition were determined in 13 patients with primary hyperlipoproteinemia (HLP) before and during treatment with cholic acid (15 mg/kg body weight/day for 3 months). In patients with type IIa HLP (n = 5), no consistent effects were seen on fasting plasma lipids or triglyceride turnover determined over a 10-hour period. Plasma triglyceride concentration was decreased in six of the eight patients with type IV HLP. Apparent triglyceride production rate was not significantly changed during treatment, but a decrease was seen in the five patients with initially elevated triglyceride synthesis. Treatment with cholic acid resulted in an increased proportion of bile acids and a decreased proportion of cholesterol and phospholipids in fasting duodenal bile; bile saturation with cholesterol was not significantly reduced. The results are discussed in relation to previous studies on the integrated regulation of bile acid and triglyceride metabolism, and it is concluded that cholic acid and chenodeoxycholic acid exert different effects on plasma triglyceride metabolism and on biliary lipid composition in HLP.

POSSIBLE INVOLVEMENT OF FATTY ACID BINDING PROTEIN IN PEROXISOMAL β -OXIDATION OF FATTY ACIDS. E.L. Appelkvist and G. Dallner (Dept. of Biochem., Arrhenius Lab., Univ. of Stockholm and Dept. of Pathology at Huddinge Hosp., Karolinska Institutet, Stockholm, Sweden) *Biochim. Biophys. Acta* 617(1), 156-60 (1980). The localization of β -oxidation of fatty acids in isolated peroxisomes from rat liver was investigated. The enzyme system is soluble in the luminal compartment and carnitine does not appear to be involved in the transfer of the CoA derivatives through the peroxisomal membrane. Experiments involving proteolysis, inhibitors and competitive inhibition suggest that a fatty acid binding protein is responsible for the carrier process. This carrier protein seems to be present in increased amounts both in the supernatant and in the peroxisomes after clofibrate induction.

EFFECT OF TYPE OF DIETARY FAT ON PLASMA AND TISSUE CHOLESTEROL OF CALVES. K.K. Barrows, T.R. Heeg, A.D. McGilliard, M.J. Richard and N.L. Jacobson (Dept. of Animal Sci., Iowa State Univ., Ames, IA 50011) *J. Nutr.* 110(2), 335-42 (1980). Two experiments were conducted to examine the effects of liquid milk diets with varying proportions of beef tallow (T) and soybean oil (SBO), with and without dry feed, on cholesterol concentrations in blood and other tissues of nonruminating calves. In experiment 1, 4-10-day-old male Holstein calves were fed for 15 weeks a reconstituted milk containing 9% or 12% dried skim milk and 2% SBO, 2% T or 3.5% T. Plasma cholesterol and cholesterol content (% of dry matter) of muscle, liver, omental fat and perirenal fat were greater ($P < 0.05$) for calves fed the 2% SBO diet. No significant differences were observed in cholesterol concentrations of the aorta, coronary arteries, brain or spleen. Experiment 2 used 4-10-day-old male Holstein calves to study the effect of 2% fat diets formulated in ratios of SBO 2/3:T 1/3 and SBO 1/3:T 2/3 and to observe the effect of dry feed added to 2% T and 2% SBO diets. Tallow and SBO fed in weighted combinations affected plasma and tissue cholesterol concentrations like the predominant fat fed alone. The greatest increases in plasma and tissue cholesterol concentrations occurred in calves fed SBO diets. Dry feed supplementation decreased body cholesterol concentrations of calves fed either fat. Body weight gain of calves fed SBO was less in both experiments.

FAILURE OF A LARGE DOSE OF VITAMIN A TO ENHANCE THE ANTIBODY RESPONSE TO TETANUS TOXOID IN CHILDREN. K.H. Brown, M.M. Rajan, J. Chakraborty and K.M.A. Aziz (Cholera Res. Lab., Dacca, Bangladesh and Div. of Geographic Med., Johns Hopkins Univ., Baltimore, MD) *Am. J. Clin. Nutr.* 33(2), 212-7 (1980). Field studies to determine the effects of a large dose of water miscible vitamin A on selected parameters of children's immunological function were completed in rural Bangladesh. There was no difference between vitamin A treated or control groups in tetanus antitoxin responses after tetanus toxoid immunization or in skin test reactivity to common antigens. Subsequent studies with mice demonstrated vitamin A dose-related antitoxin responses, but the animals required amounts of vitamin that would be likely to cause undesirable side effects if administered in similar doses to children.

THE EFFECTS OF VITAMIN E DEPLETION AND REPLETION ON PROSTAGLANDIN SYNTHESIS IN SEMITENDINOSUS MUSCLE OF YOUNG RABBITS. A.C. Chan, C.E. Allen and P.V.J. Hegarty (Dept. of Food Sci. and Nutr. and Dept. of Animal Sci., Univ. of Minnesota, St. Paul, MN 55108) *J. Nutr.* 110(1), 66-73 (1980). The effects of dietary vitamin E-depletion and repletion on

the cyclooxygenase activity was studied in the semitendinosus muscle of rabbits. The prostaglandin (PG) cyclooxygenase system in rabbit semitendinosus muscle was characterized and found to depend on reduced glutathione and 1-epinephrine as cofactors. Skeletal muscle cyclooxygenase generates PGE₂ and PGF₂ α at a ratio approximately equal to one. Weanling New Zealand white rabbits were fed a vitamin E-deficient diet for 4 to 5 weeks. Controls received 50 mg dl- α -tocopherol acetate twice weekly. Vitamin E deficiency caused a significant reduction in cyclooxygenase activity but did not change the PGE₂/PGF₂ α ratio. Oral supplementation of tocopherol acetate promptly returned the cyclooxygenase activity back to the control values within 48 hours. The decreased cyclooxygenase activity explains in part the increased level of arachidonic acid in skeletal muscle phospholipid previously reported in this laboratory. The possible involvement of decreased prostaglandin endoperoxides with platelet aggregation in vitamin E deficiency is discussed.

STUDIES ON THE EFFECT OF HEPATECTOMY ON PIG POST-HEPARIN PLASMA LIPASES. C. Ehnholm, T. Schröder, T. Kuusi, B. Bång, P. Kinnunen, K. Kahma and M. Lempiinen (Central Public Health Lab., Third Dept. of Med. and Second Dept. of Surgery, Univ. of Helsinki, Helsinki, Finland) *Biochim. Biophys. Acta* 617(1), 141-9 (1980). The effect of different amounts of heparin injected intravenously in swine on lipoprotein lipase and hepatic lipase activities in post-heparin plasma was studied using an immunochemical method. It is concluded that the higher the heparin dose injected the longer can lipolytic activities be measured in plasma. After hepatectomy no immunoreactive hepatic lipase activity could be demonstrated in post-heparin plasma confirming our previous findings that the liver is the only source of hepatic lipase. To study the role of the liver in the clearance of plasma lipoprotein lipase activity after the administration of heparin normal and hepatectomized pigs were given 200 I.U./kg body weight followed by a heparin infusion of 100 I.U./h per kg body weight. In the control pigs the heparin injection caused a rapid release of lipoprotein lipase and hepatic lipase activities. From these experiments we conclude that after heparin injection the liver is involved in the clearance of post-heparin plasma lipolytic activity.

THE MECHANISM OF ETHER BOND FORMATION IN O-ALKYL LIPID SYNTHESIS. S.J. Friedberg, D.M. Gomillion, and P.L. Storer (Univ. of Texas Health Sci. Center at San Antonio, Dept. of Med., San Antonio, TX 78284) *J. Biol. Chem.* 255(3), 1074-9 (1980). O-Alkyl dihydroxyacetone phosphate is formed enzymatically from acyl dihydroxyacetone phosphate and a long chain fatty alcohol. This reaction is accompanied by stereospecific exchange of the *pro-R* hydrogen of carbon 1 (carbon 1 of all compounds corresponds to carbon 1 of *sn*-glycerol) of the dihydroxyacetone phosphate moiety with retention of configuration. In the present investigation, data are provided to show that the initial loss of hydrogen from carbon 1 of acyl dihydroxyacetone phosphate does not depend on the presence of the fatty alcohol. In addition, the occurrence of a Schiff base between enzyme and acyl dihydroxyacetone phosphate, comparable to the fructose-1,6-diphosphate aldolase reaction, could not be demonstrated. It is concluded that the formation of 1-O-alkyl dihydroxyacetone phosphate via the formation of intermediate 1-O-acyl endiol and 1-O-alkyl endiol is a likely mechanism.

DIFFERENT METABOLISM OF SATURATED AND UNSATURATED LONG CHAIN PLASMA FREE FATTY ACIDS BY INTESTINAL MUCOSA OF RATS. A. Gangl, W. Kornauth, J. Mlczech, O. Sulm, and B. Klose (Universitätsklinik für Gastroenterologie und Hepatologie, Allgemeines Krankenhaus, Garnisonsgasse 13, A - 1090 Vienna, Austria) *Lipids* 15(2), 75-9 (1980). During fat absorption, unsaturated long chain fatty acids are esterified at a higher rate than saturated fatty acids of similar chain length. This phenomenon has been attributed to differences in the binding affinity of fatty acids to a cytosolic fatty acid-binding protein. As intestinal mucosa utilizes plasma free fatty acids as well, we investigated whether long chain plasma free fatty acids of different degree of saturation are metabolized also at different rates. The data show that despite equal initial uptake by intestinal mucosa unsaturated long chain fatty acids taken up from plasma are esterified to a higher and oxidized to a lower extent than saturated plasma free fatty acids. Unsaturated plasma free fatty acids, therefore, may provide a more important source of fatty acids for endogenous intestinal lipoprotein lipids than saturated plasma free fatty acids. It is speculated that the fatty acid binding protein might be operative not only in the intracellular transport and metabolism of liminal fatty acids but of plasma free fatty acids as well.

COMPARATIVE EFFECTS OF ALL-TRANS AND 13-CIS RETINOIC ACID ADMINISTRATION ON SERUM AND LIVER LIPIDS IN RATS. L.E. Gerber and J.W. Erdman Jr. (Dept. of Food

Sci., Univ. of Illinois, Urbana, IL 61801) *J. Nutr.* 110(2), 343-51 (1980). The effects of all-*trans* and 13-*cis* retinoic acid upon serum and liver lipids were investigated in Sprague-Dawley rats. Groups of rats were fed daily with 105, 210 and 315 $\mu\text{g/g}$ diet of one of the retinoids for periods of up to 8 days. Other groups were injected intraperitoneally (I.P.) daily with retinoids at levels equivalent to the daily intake of rats receiving 105 or 210 μg of retinoid/g diet. All dietary concentrations of all-*trans* retinoic acid induced hypertriglyceridemia, however, only the highest dietary concentration of the 13-*cis* form caused this response. Injection of the all-*trans* form consistently increased serum triglycerides, while 13-*cis* retinoic acid did so in only one case. Retinoid-fed rats fasted for 6 hours before blood sampling demonstrated similar increases in serum triglycerides compared to their respective controls. Also, retinoid administration reduced serum retinol at all levels tested with the all-*trans* form appearing to be more potent. Growth and feed intake was somewhat reduced in rats receiving the highest level of all-*trans* retinoic acid. Liver analysis did not reveal fatty liver or alterations in phospholipid, cholesterol or vitamin A content in any groups monitored. Our previous studies have shown induction of hypertriglyceridemia when rats were fed as low as 26 $\mu\text{g/g}$ diet of all-*trans* retinoic acid. The current studies would indicate that feeding 315 $\mu\text{g/g}$ diet of the 13-*cis* isomer was required to elicit a similar response.

Fats and oils

STUDY OF THE FRACTIONATION OF PALM OIL. OBTAINMENT OF A FLUID OIL WITH AN IODINE VALUE ABOVE 75, AND CHARACTERISTICS OF THE RESULTING PRODUCTS. J.M. Klein. *Oleagineux* 34,(11) 531-6 (1979). The advantage of fractionating palm oil to extract its fluid fraction is underlined. To enable this fluid fraction to be used in temperate climates without a deposit forming at 15°C, it is essential that the maximum content in trisaturated triglycerides and disaturated-monounsaturated triglycerides should not be exceeded. The different criteria to be respected in order to obtain a fluid fraction comparable to groundnut oil as regards stability when cold are reviewed. The triglyceride composition of the fractions recovered is also examined.

INFLUENCE OF TECHNIQUES ON THE QUALITY OF FOOD PRODUCTS IN THE FAT AND OIL INDUSTRY. J.P. Helme. *Rev. Fr. Corps Gras* 27(3), 121-30 (1980). Traditional refining removes substances which hinder the hot or cold performance of fats: phospholipids, fatty acids, metallic traces, waxes and contaminants: mycotoxins and pesticides. However, artifacts and isomers can form during refining, mainly during bleaching and deodorization, but the oxidation products are not fully removed by refining. Concerning the fat processing (law of February 12, 1973) the problem depends on the kind of process: fractionation, hydrogenation or interesterification.

MARGARINE: SPECIFIC ASPECTS IN SOME PARTICULAR NUTRITIONAL SITUATIONS. H. Bour. *Rev. Fr. Corps Gras*, 27,(2), 55-60 (1980). Food products answering definite nutritional needs can be obtained by modifying the components of the fatty or aqueous phase, the proportions and kind of raw materials. The margarines containing short and medium chain fatty acids give good results in the case of fat malabsorption. The low-calorie margarines are able to reduce the lipids of the diet. The high in polyunsaturated fatty acid margarines are interesting to protect the vascular system.

INVESTIGATIONS ON THE SELECTIVE HYDROGENATION MECHANISMS. I. PRINCIPLE OF VEGETABLE OIL MODELING. G. Mallet and E. Ucciani. *Rev. Fr. Corps Gras*, 27(2), 71-80 (1980). Whatever the catalyst it is always difficult to establish the mechanism of selective hydrogenation in the case of vegetable oils. Kinetic study, structures and isotopic labelling provide necessary but not sufficient information. For instance, it is actually impossible to differentiate dienes coming from linolenic acid, from isomerized linoleic acid. As a matter of fact, nobody knows how linolenic acid is reduced when hydrogenation of oils is in progress. The simulation technique allows to remedy this situation. The principle is founded on a chemical labelling with different alcohols at the carboxyl group of the linolenic, linoleic and oleic chains (for example R₁COOMe, R₂COOPr, R₃COOHx). Different models are discussed. A preparative fractionation procedure is described. At last a new catalyst characterization is proposed on the basis of the chromatographic profile of the dienes coming from the linolenic chain.

SEPARATION OF OXIDIZED AND UNOXIDIZED MOLECULAR SPECIES OF PHOSPHATIDYLCHOLINE BY HIGH PRESSURE LIQUID CHROMATOGRAPHY. C.G. Crawford, R.D. Plattner, D.J. Sessa and J.J. Rackis (Northern Regional Res. Center, ARS, SEA, USDA, Peoria, IL 61604) *Lipids* 15(2), 91-4 (1980). Soy phosphatidylcholine (PC) has been separated into its major molecular species

by reverse-phase high pressure liquid chromatography (HPLC). An aqueous methanol gradient was used that allowed detection of the various species by their absorbance at 206 nm. Oxidized species were detected by their absorbance at 234 nm and were resolved from the unoxidized species. This technique has been used to separate and purify unoxidized dilinoleyl phosphatidylcholine (di 18:2 PC) from other species of soy PC and to monitor the autooxidation of an aqueous suspension of the purified di 18:2 PC. Two oxidized products were formed from di 18:2 PC. Further analysis showed that they were PC, but one of the products contained an oxidized and an unoxidized fatty acid; in the other product, both fatty acids were oxidized.

IDENTIFICATION OF CHONDRILLASTEROL IN TWO CUCURBITACEAE SEED OILS BY PROTON NUCLEAR MAGNETIC RESONANCE SPECTROSCOPY. T. Iida, T.M. Jeong, T. Tamura and T. Matsumoto (Coll. of Engineering, Nihon Univ., Koriyama, Fukushima-ken, 963 Japan) *Lipids* 15(1), 66-8 (1980). The absolute configuration at C-24 of C-24 epimeric 24-ethyl-5 α -cholesta-7, E-22-dien-3 β -ols I-IV previously isolated from tea seed oil, shea fat, and gourd and sponge cucumber seed oils, respectively, was studied by proton nuclear magnetic resonance spectroscopy. The results showed that the sterols I and II are identical with spinasterol (24S/ α -ethyl group), whereas the sterols III and IV are identified as its 24R/ β -epimer, chondrillasterol. This study has thus for the first time properly documented the presence in tracheophytes of a 24 β -ethylsterol in which $\Delta^{25(27)}$ -bond is reduced.

10 α -CUCURBITA-5, 24-DIEN-3 β -OL FROM GOURD SEED OIL. T. Itoh, T. Tamura, T.M. Jeong, T. Tamura and T. Matsumoto (Coll. of Sci. and Technology, Nihon Univ. 1-8, Kanda Surugadai, Chiyoda-ku, Tokyo, 101 Japan) *Lipids* 15(2), 122-3 (1980). 10 α -Cucurbita-5,24-dien-3 β -ol was isolated from the unsaponifiable matter of the seed oil of gourd (*Lagenaria leucantha* var. *Gourda*; Cucurbitaceae); it has been previously synthesized, but never found in the plant kingdom. The triterpene alcohol represents the parent compound of a number of cucurbitacins, highly oxygenated tetracyclic triterpenes found in Cucurbitaceae and some other flowering plants.

CAROTENE IN BOVINE MILK FAT GLOBULES: OBSERVATIONS ON ORIGIN AND HIGH CONTENT IN TISSUE MITOCHONDRIA. S. Patton, J.J. Kelly and T.W. Keenan (Dept. of Food Sci., Pennsylvania State Univ., Univ. Park, PA 16802) *Chem. Phys. Lipids* 15(1), 33-8 (1980). The location and origin of carotenoids in bovine milk fat globules was investigated using spectral absorption of lipid solutions at 461 nm to quantitate carotene. Release of membrane from globules as a result of churning to butter or by freezing and thawing of the globules yielded membrane preparations which were devoid of carotene. Globule cores from these procedures exhibited carotene concentrations comparable to those in total milk lipids. Fractionation of lactating bovine tissue and analysis of lipid extracts revealed that the intracellular fat droplets have carotene concentrations approximating those of secreted globules. However, intracellular membranes of the tissue, particularly the mitochondria, are much richer in carotenoids than formative or secreted fat globules. The evidence indicates that bovine milk fat globules acquire carotene during their formation in the cell, but that some minor fraction of the total carotene may be extracted from the enveloping secretory membrane. Mean carotene values ($\mu\text{g/g}$ of lipid) for fractions from three samples of lactating tissue were: whole tissue 47, mitochondria 461, microsomes 69, cytosol 67, fat droplets 8, milk 9. One tissue analysis indicated that Golgi membranes contain somewhat more carotene than do microsomes.

CHANGES IN STEREOSPECIFIC DISTRIBUTION OF YEAST FATTY ACIDS WITH AGE. Y. Phornpiboonya and R.C. Jack (Dept. of Biological Sci., St. John's Univ., Jamaica, NY 11439, U.S.A.) *Chem. Phys. Lipids* 26(1): 57-66 (1980). Stereospecific analyses of glycerolipids from 7-, 14- and 21-day-old cultures of the yeast *Lipomyces lipoferus* revealed that each position of the glycerolipids had a unique distribution of fatty acids which changed to varying degrees with age, and that, in the triacylglycerols, age had a greater effect on fatty acid content at sn-3 than at sn-1 or sn-2. Age-related changes in unsaturation were, however, greater in the phospholipids than in the triacylglycerols. Among the major phospholipids of *L. lipoferus* (phosphatidylcholine, phosphatidylinositol and phosphatidylethanolamine), changes in the proportion of unsaturated to saturated fatty acids, and in the number of double bonds per mole, were greater at sn-2 than at sn-1, except for phosphatidylinositol between 14 and 21 days of age. The pattern of acylation of phosphatidylinositol between 14 and 21 days was thus different from that of phosphatidylcholine and phosphatidylethanolamine. Furthermore, at the three ages investigated, phosphatidylinositol had low and relatively constant levels of unsaturation compared with phosphatidylcholine and phosphatidylethanolamine. The net decrease in phospholipid double bonds per

mole in aging cells of *L. lipoferus*, and previous data, suggest that aging in this yeast is accompanied by a decrease in membrane fluidity.

ISOLATION OF 1,3-DISTEAROYL-GLYCERO-2-PHOSPHOCHOLINE (β -LECITHIN) FROM COMMERCIAL 1,2-DISTEAROYL-*SN*-GLYCERO-3-PHOSPHOCHOLINE. M.M. Ponpipom and R.L. Bugianesi (Merck Sharp & Dohme Res. Labs., Rahway, NJ 07065) *J. Lipid Res.* 21(1), 136-9 (1980). Different batches of 1,2-distearoyl-*sn*-glycero-3-phosphocholine (DSPC) had varying amounts of contaminants which appeared to affect systematic biological studies. This contaminant was separated by silica gel column chromatography followed by high performance liquid chromatography and identified as 1,3-distearoyl-glycero-2-phosphocholine (β -lecithin).

DETERMINATION OF DOUBLE BOND POSITION IN TRI- TO HEXAENOIC FATTY ACIDS BY MASS SPECTROMETRY. B. Schmitz and H. Egge (Physiologisch Chemisches Institut der Universität Bonn, Nussallee 11, D-5300 Bonn F.R.G.) *Chem. Phys. Lipids* 25(3), 287-98 (1979). Methyl esters of polyenoic fatty acids (FAE) with up to six double bonds can be transformed to polyhydroxy derivatives by treatment with OsO_4 . Under low energy electron impact the trimethylsilyloxy (O-TMS) derivatives of these polyenoic fatty acids exhibit mass spectrometric fragmentation pat-

terns that are straightforward due to the almost exclusive cleavage of the C-C bonds in between vicinal O-TMS groups. Thus it is possible to localise original double bond positions and also to predict fragmentation patterns of hitherto unknown fatty acid species. The method is applicable to polyenoic compounds with isolated as well as conjugated double bonds.

SYNTHETIC STUDIES OF PHOSPHOINOSITIDES. A.E. Stepanov and V.I. Shvets (Lomonosov Inst. of Fine Chem. Tech., Moscow U.S.S.R.) *Chem. Phys. Lipids* 25(3), 247-63 (1979). Intensive study of native lipids, in particular of polar lipids, is justified by their importance for molecular organisation and functioning of biological membranes. One of the major groups of native phospholipids is the phosphoinositides which are actively involved in brain cells, nervous tissue, liver and other vital organs of man and higher animals. A recent review by Michell provides exhaustive data on chemistry and metabolism of this interesting class of phospholipids. One approach to the study of the structure and properties of these compounds is provided by the chemical synthesis of molecular types of phospholipids with native stereochemical configuration. Advances in the synthesis of phosphoinositides open new vistas of synthetic individual phosphoinositides with practically any structure and molecular type.

A METHOD FOR THE QUANTITATIVE ESTIMATION OF CHOLESTEROL α -OXIDE IN EGGS. L.S. Tsai, K. Ijichi, C.A. Hudson and J.J. Meehan (Western Regional Res. Center, SEA, USDA, Berkeley, CA 94710) *Lipids* 15(2), 124-8 (1980). A method for the quantitation of cholesterol α -oxide in egg and egg products is described. Total lipids extracted from dry egg samples were fractionated on a silicic acid column to concentrate cholesterol oxides which were then quantitatively determined by gas liquid chromatography (GLC). Those samples which showed cholesterol oxides by GLC were further analyzed by high pressure liquid chromatography (HPLC) for the ratio of cholesterol α -oxide and cholesterol β -oxide. Cholesterol α -oxide content was calculated from the combined results of GLC and HPLC.

SYNTHESIS OF [11- 2H_2], [8- 2H_2], [7- 2H_2], [6- 2H_2], [5- 2H_2], [4- 2H_2] and [3- 2H_2] *cis*-9-OCTADECENOATES. A.P. Tulloch (National Res. Council of Canada, Prairie Regional Lab., Saskatoon, Saskatchewan S7N 0W9 Canada) *Chem. Phys. Lipids* 25(3), 225-35 (1979). Deuterated oleates have been synthesized by semihydrogenation of acetylenic intermediates. [11- 2H_2] Oleate was prepared by two-carbon chain extension of the C_{16} alcohol obtained from [1- 2H_2] octyl bromide and 7-octyn-1-ol. [8- 2H_2] and [7- 2H_2] oleates were both prepared from dimethyl suberate, tetradeuterio intermediate C_{18} alcohols were synthesized from [1,8- 2H_4] and [2,7- 2H_4] octane diols by monobromination, conversion to deuterated 9-decyn-1-ols and reaction with octyl bromide. Oxidation gave [8- 2H_2]-9-octadecynoate and [2,7- 2H_2]-9-octadecynoate, after semihydrogenation of the latter, deuterons at C-2 were removed by

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