ABSTRACTS

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

• Oils and Fats

OIL STAIN ON MILD STEEL INDUCED BY RUST PREVENTIVE OIL. Tamotsu Naruse and Yoichi Kato (Ind. Research Inst., Aichi Pref.). *Yukagaku* 17, 512–16 (1968). In the mixed system of liquid paraffin + rust inhibitor + unsaturated fatty acid, the effect of several additives such as antioxidants or radical scavengers was investigated. The formation of oil stain was retarded by adding these agents, the ability of retardation of radical scavengers being stronger than that of antioxidants.

STUDIES ON TRIGLYCERIDE STRUCTURE OF HYDROGENATED FATS. I. SYNTHESES OF STANDARD TRIGLYCERIDES AND VON RUDLOFF OXIDATION OF THEIR MIXTURES. Yoshikazu Takahashi (Miyoshi Oil & Fat Co., Tokyo). *Yukagaku* 17, 492–505 (1968). Purities of the tri and partial glycerides were checked by melting point, infrared spectrometry and thin-layer chromatography, and a comparison of their polymorphic forms with those reported in the literature was made. The standard triglyceride mixtures were oxidized according to Young's modification and esterified with diazomethane in the presence of methanol. After removal of volatile esters, the oxidized methylated glycerides were fractionated into four glyceride types, S_3, S_2U, SU_2, and U_3 by preparative thin-layer chromatography. The fractions were recovered quantitatively with ether, weighed and the resultant methyl ethers were analyzed by gas-liquid chromatography.

TREND OF IMPORTED PETROLEUM, OILS AND FATS AND THEIR ANALYSIS. Teruo Tenma and Yukio Ono. *Yukagaku* 17, 481–8 (1968).

PAPER CHROMATOGRAPHIC SEPARATION AND QUANTITATIVE DETERMINATION OF METHYL KETONES. Cl. Franzke, J. Strobach and W. Geib (Inst. for Nutr. Chem. and Tech., Humboldt Univ., Berlin, W. Ger.). *Fette Seifen Anstrichmittel* 70, 633–638 (1968). A procedure is described by which methyl ketones in natural fats are separated along with the unsaponifiables and paper chromatographically identified as their dinitrophenyl hydrazones folowed by their elution from the paper chromatogram for quantitative determination by photometry.

GAS CHROMATOGRAPHIC INVESTIGATION OF NATURAL WAXES. K. H. Miltenberger (Farbwerke Hoechst AG, Frankfurt (M)-Hochst, Ger.). *Fette Seifen Anstrichmittel* 70, 736–742 (1968). A rapid gas chromatographic method for the identification of natural waxes is described. The wax acids and the unsaponifiables are investigated after saponification. The methyl esters of the wax acids as well as the acetates and oxidation products from the unsaponifiables showed a characteristic pattern of distribution among the individual waxes. In the characterization of natural waxes it is desirable to investigate the methyl esters of the wax acids, because after saponification the wax acids are obtained as a more uniform class of substances than the unsaponifiables.

INVESTIGATION AND EVALUATION OF SUNFLOWER OILS OF NATURAL PURITY. J. Wurziger and L. Schumann (Chem. and Agr. Inves. Hygiene Inst., Hamburg, Ger.). *Fette Seifen Anstrichmittel* 70, 729–733 (1968). Color reactions for differentiating unrefined from refined sunflower oils are reported. The qualitative detection of the substances responsible for the color reaction can be carried out with unsaponifiables; however, it is also possible to use the oil directly after a certain enrichment. Under the experimental conditions chosen and using antimony trichloride spray the pressed oils gave two blue spots with similar R_f values on the thin-layer chromatogram. The substances which give the blue color were found to be keto-steroids. Keto-steroids were not found in sunflower oils.

HYDROGENATION OF PEANUT AND SESAME OILS IN HEXANE. M. M. Chakrabarty and D. Bhattacharyya (Dept. Applied Chem., Calcutta Univ., Calcutta 9, India). *Fette Seifen Anstrichmittel* 70, 714–719 (1968). The various parameters of hydrogenation of peanut and sesame oils in hexane were studied. At 140C, 100 psig pressure and 200 shakes per minute agitation, the rate and selectivity of the hydrogenation of peanut oil in hexane increased as the catalyst concentration increased from 0.1 to 0.3% nickel. Above 0.5% catalyst the rate remained practically constant and selectivity decreased. The rate and selectivity decreased as the total pressure decreased from 100 to 85 to 70 psig at 140C and 0.3% nickel. *Trans* isomer formation was almost similar to that observed at 100 psig. The reaction appeared to be of the first order for a certain period of hydrogenation at all catalyst concentrations. The temperature was the most important variable in controlling rate and selectivity. Both selectivity and *trans* isomer formation increased from 115C to 140C. These experiments demonstrated the commercial feasibility of the solvent (miscella) hydrogenation process.

INVESTIGATION OF STEROID MIXTURES BY THIN-LAYER CHROMATOGRAPHY. A. Seher and E. Homberg (Fed. Inst. for Fat Res., Munster (Westf.), Ger.). *Fette Seifen Anstrichmittel* 70, 481–485 (1968). A new method of thin-layer chromatography on MgO-Al_2O_3-$CaSO_4$ plates was developed for the separation of natural mixtures of steroids in the form of their acetates. By this method, the formation of critical pairs could be avoided and considerably better separation could be achieved. Application of the method to various vegetable oils showed in all cases the occurrence of a steroid with a R_f value corresponding to cholesterol. This steroid, occurring in considerable amounts especially in palm oil, was isolated from palm oil in centigram amounts by preparative thin-layer chromatography. Melting point, X-ray structural analysis, infrared and mass spectra, optical rotation and rotational dispersion of this substance were identical with the corresponding data of cholesterol. It is thus proven that cholesterol occurs in plants.

PHOSPHOLIPIDS FROM *Bacillus stearothermophilus*. G. L. Card, C. E. Georgi and W. E. Militzer (Dept. Microbiol., Univ. of Montana, Missoula, Mont. 59801). *J. Bacteriol.* 97, 186–192 (1969). The extractable lipid (8% of the cell dry weight) consisted of 30–40% neutral lipid and 60–70% phospholipid from *Bacillus stearothermophilus*. The phospholipids were identified as diphosphatidyl glycerol (23–42%), phosphatidylglycerol (22–39%) and phosphatidyl ethanolamine (21–32%). The lipids of cell fractions were: whole protoplasts 10% of the fraction dry weight; membrane 18%; 30,000 x g particulate fraction 22%; and 105,000 x g particulate fraction 26%.

FATTY ACIDS OF LISTERIA MONOCYTOGENES. L. J. Raines, C. W. Moss, D. Farshtchi and B. Pittman (Bur. Dis. Prevent. and Environ. Contro, National Comm. Dis. Center, Atlanta, Ga. 30333). *J. Bacteriol.* 96, 2175–2177 (1968). The fatty acid composition of thirty-three strains of *Listeria monocytogenes* were studied. Most strains contained C15 branched acids (*i* 15 and *a* 15) in greatest concentration (31–50%). The next most abundant acids were *n* 14:0 (2–25%), *n* 16:0 (6–24%) and 17:0 branched (4–30%). Other fatty acids present in less than 10% amounts were *n* 12:0, 15:0, 17:0, 22:0 and 16:1.

CYCLOPROPANE FATTY ACIDS OF RUGOSE *Vibrio cholerae*. B. L. Brian and E. W. Gardner (Dept. Biol., Texas Christian Univ., Fort Worth, Tex. 76129). *J. Bacteriol.* 96, 2181–2182 (1968). Unusual fatty acid composition was noted in the rugose variant of *Vibrio cholerae* compared to the parent strain. The 16:0 and 16:1 fatty acid were lower in the rugose variant 27% versus 37% and 17% versus 37% respectively. The 18:1 acid was greatly increased in the rugose strain, 40% versus 14%. In addition small amounts of C17 and C19 cyclopropane fatty acids not detected in the parent strain were found in the rugose strain.

A POLAROGRAPHIC PROCEDURE FOR THE DETERMINATION OF PHENOLIC ANTIOXIDANTS IN EDIBLE FATS USING A ROTATING GRAPHITE ELECTRODE. Cl. Franke, F. Kretzschmann and K. Beining (Inst. for Nut. Res. and Tech., Humboldt Univ., Berlin, W. Ger.). *Fette Seifen Anstrichmittel* 70, 472–476 (1968). A description of a polarographic method to determine antioxidants in edible fats is given. The rotating graphite electrode used for this purpose is easily constructed. The method is suitable for the quantitative determination of antioxidants in fats in the range of 0.01–0.02%. Qualitative assessments can be made only to a limited extent. 2,6-Di-tert.-butyl-p-cresol cannot be determined.

AUTOXIDATION OF SATURATED FATTY ACIDS. IV. OXYGEN CONSUMPTION AND THE FORMATION OF PEROXIDES IN THE OXIDATION

OF LAURIC AND STEARIC ACID AND THEIR METHYL ESTERS. H. Thaler and H.-J. Kleinau (Inst. for Nutr., Tech. Sch. Braunschweig, Ger.). *Fette Seifen Anstrichmittel* 70, 465–469 (1968). The investigation demonstrated a reproducible direction of oxygen uptake and peroxide formation. In the oxidation of free acids the oxygen uptake was considerably higher than the corresponding peroxide content. The amounts of peroxides determined are so small that secondary reactions may be assumed to occur easily under the conditions of the experiment. The higher content of peroxide compounds for the oxidation of methyl esters compared to that in the oxidation of free acids indicated a hindrance of the subsequent reactions which are associated with the decomposition of the peroxides. The experiments showed that initially an unusually large part of the absorbed oxygen is consumed in the formation of peroxides. The formation of split products can therefore occur here only with great difficulty in contrast to the case of free acids.

HYDROCARBONS OF BLUE-GREEN ALGAE: GEOCHEMICAL SIGNIFICANCE. K. Winters, P. L. Parker and C. Van Baalen (Marine Science Inst., Univ. of Texas, Port Arkansas 78373). *Science* 163, 467–68 (1969). The hydrocarbon compositions of 11 species of blue-green algae are simple and qualitatively similar. Three marine coccoids contain only monoenoic and dienoic C_{19} hydrocarbons. Hydrocarbons of the remaining eight species are C_{15} to C_{18}. Hydrocarbons of higher molecular weight (C_{20} or more) were not detected. Blue-green algae do not appear to be the source material for the long-chain (greater than 20 carbons) hydrocarbons found in ancient sediments.

PREVENTION OF HYDROGENATION FLAVOR DURING TRACE HYDROGENATION OF BUTTEROIL. A. K. Vasishtha, J. G. Leeder and S. S. Chang (Dept. of Food Science, Rutgers State Univ., New Brunswick, N.J. 08903). *Food Technol.* 23, 110–13 (1969). A previous paper from this laboratory reported that the flavor stability of butteroil may be significantly improved by slight selective catalytic hydrogenation. However, an off-flavor, "hydrogenation flavor," developed during the hydrogenation process which had to be removed by vacuum steam distillation. The latter also eliminated the desirable flavor components of the butteroil. In the present investigation a statistically designed low temperature hydrogenation process has been studied which resulted in a process that can hydrogenate the butteroil without the formation of hydrogenation flavor. The butteroil thus hydrogenated requires no vacuum steam distillation and has an improved flavor stability.

DIRECT GAS CHROMATOGRAPHIC ANALYSIS OF RAPESEED OIL STEROLS. F. Mordret (Ecole Superieure d'Application des Corps Gras, Paris). *Rev. Franc. Corps Gras* 15, 675–681 (1968). The oil is first esterified with methanol and then injected into the gas chromatograph. Operating conditions are: 3% SE-30 on Gas Chrom Q, 80/100 mesh, 2 m. × 4 mm. column at 240C or 3 m. × 4 mm. at 260C, hydrogen flame detector at 245 or 265C, respectively. Under either of these sets of conditions, the methyl esters of the fatty acids emerge within the first ten minutes, and the sterols emerge after twenty to forty minutes. The longer column gave better resolution and permitted good separation of β-sitosterol, campesterol, brassicasterol, and cholesterol, which was used as an internal standard. Results from the direct method correlated well with those obtained from the standard method. The direct method can be used in other situations, such as, for determining possible contamination of olive oil with soybean oil.

CONJUGATED HYDROGENATION OF MAHUA OIL (MADHUCA LATIFOLIA). M. M. Chakrabarty, D. Bhattacharyya, and A. Basu (Dept. of Applied Chem., Calcutta Univ., Calcutta-9, India). *Ind. Chim. Belge* 33, 883–886 (1968). (In English) By the process of conjugated hydrogenation with either the theoretical amount or a 100% excess of ethanol at 225C with 0.5–1.0% nickel catalyst, equilibrium iodine values of 40–41 were obtained after three hours. The initial I.V. of the mahua oil was 60.0. With the theoretical amount of isopropanol, the equilibrium I.V. of 17 was reached in 2–3 hours, while with a 100% excess of isopropanol, equilibrium iodine values of 4–6 were obtained after the same length of time. Thus the rate of hydrogenation and the equilibrium iodine values depend on the type of alcohol, and for secondary alcohols, also on the amount. The reaction with the primary alcohol was highly selective.

EXTRACTION OF OLEAGINOUS RAW MATERIALS: A STUDY OF THE TRANSFER MECHANISM OF FATS. PARTS C, DISCUSSION, AND D, CONCLUSIONS. O. M. Angelidis (Phrynis 7, Athens (503), Greece). *Oleagineux* 23, 667–672 (1968). The first two parts of this article appear in *Oleagineux* 23, 535–540 and 587–595 (1968). Extraction of oleaginous raw materials involves three processes: dissolution of the oil in the solvent, diffusion of oil into the bulk of the solvent, and viscous flow of the solution in the capillaries of the tissue. All three occur simultaneously but at different rates depending on the material being extracted. Capillary flow, which determines the overall rate, is governed by the equation $E = \alpha t^{\beta}$, where E is the residual quantity of oil, α is a constant for any material at a given temperature and moisture, t is time, and β depends on the moisture content, temperature and pretreatment of the material. Equations are given to show the effects of crushing and quantity of residual oil on the rate of extraction. A method is described for calculating extraction capacity in commercial installations.

AUTOXIDATION OF LINOLEIC ACID AT 20–40C. II. THEORY. M. Loury and M. Forney (Institute des Corps Gras, Paris). *Rev. Franc. Corps Gras* 15, 663–673 (1968). The authors review the classical theory of lipid autoxidation and decomposition of hydroperoxides and propose modifications to explain the formation of water, low molecular weight alcohols, and formic and acetic acid esters. These alcohols and esters result from secondary autoxidation of C_3 through C_7 aldehydes formed by the initial autoxidation. Reaction of the OH radical during chain propagation leads to the formation of significant amounts of water.

FORMATION OF FREE FATTY ACIDS IN OIL SEEDS DURING STORAGE. M. Naudet (Lab. National des Matieres Grasses, ITERG) and S. Biasini. *Rev. Franc. Corps Gras* 15, 657–662 (1968). During storage of oil seeds, free fatty acids are formed enzymatically as a result of mold growth on the seeds. The fungal lipases show no specificity with respect to position on the glycerol moiety. Any treatment which inhibits mold growth also inhibits free fatty acid formation. The seeds included in the study were: peanuts, rapeseeds, sunflower seeds, soybeans, copra, and palm kernels. In the latter two materials, the free acids are decarboxylated to form volatile methyl ketones, which causes a net loss of oil.

SEPARATION OF MONO- AND DIGLYCERIDES BY GAS-LIQUID CHROMATOGRAPHY. R. Watts and R. Dils (Dept. of Biochem., Univ. of Birmingham, Birmingham, England). *J. Lipid Res.* 10, 33–40 (1969). The parameters affecting the separation and quantification of trimethylsilyl ethers of mono- and diglycerides have been investigated by gas-liquid chromatography with QF-1 and SE-30 as stationary phases and a flame ionization detector. Results have been compared with those obtained earlier for triglycerides. The isothermal characteristics of a range of trimethylsilyl ethers of mono- and diglycerides on both stationary phases showed that log retention volume was directly proportional to carbon number and inversely proportional to absolute temperature. However, glyceride derivatives with lower carbon numbers deviated from these relationships. By using various rates of programmed temperature rise, we have determined the elution temperatures (Kelvin scale) of the mono- and diglyceride trimethylsilyl ethers relative to that of glycerol trilaurate. The "carbon equivalent of a trimethylsilyl group" is defined and shown to be useful in comparing the chromatographic properties of different glyceride classes. Weight and molar correction factors have been obtained and used to analyze diglycerides derived from egg and bovine brain lecithins.

GAS-LIQUID CHROMATOGRAPHY OF DIALKYL, ALKYL ACYL, AND DIACYL DERIVATIVES OF GLYCEROL. R. Wood, W. J. Baumann, F. Snyder and H. K. Mangold (Medical Div., Oak Ridge Associated Univ., Oak Ridge, Tenn. 37830). *J. Lipid Res.* 10, 128–131 (1969). Dialkyl, alkyl acyl, and diacyl glycerols were resolved as trimethylsilyl ethers and as acetates by gas-liquid chromatography on a nonpolar stationary phase (OV-1). The two types of derivatives proved suitable for quantitative gas chromatographic analysis.

OXIDATIVE CLEAVAGE OF LIPIDS WITH SODIUM METAPERIODATE IN PYRIDINE. W. J. Baumann, H. H. O. Schmid and H. K. Mangold (Univ. of Minnesota, Hormel Inst., Austin, Minn. 55912). *J. Lipid Res.* 10, 132–133 (1969). The cleavage of

α-diols and α-amino alcohols with sodium metaperiodate proceeds under mild conditions and in high yields when pyridine is used as reaction medium. The method is suitable for preparative as well as analytical applications.

THIN-LAYER SYSTEMS GIVING MAXIMUM SEPARATION OF α- AND β-TOCOPHEROLS. P. G. Roughan (Plant Physiol. Div., D.S.I.R., Palmerston North, New Zealand). *J. Chromatog.* 29(1), 293–5 (1967). Mixed layers (0.25 mm thick) of aluminium oxide G-basic zinc carbonate (3:1) or Kieselgur G-basic zinc carbonate (2:1), with $CHCl_3$ and benzene-cyclohexane (3:7) as respective solvents, gave the best results. In all the systems tested, α-tocopherol migrated faster than the β-homologue.

ISOLATION AND THIN-LAYER CHROMATOGRAPHY OF FAT-SOLUBLE DYES. W. Reiners (Inst. f. Pharm. u. Lebensmittelchem., Univ. Wurzburg, Germany). *Z. Anal. Chem.* 229(6), 406–9 (1967) (in German). Dyed fat or mineral oil (≃5 g) is well mixed with 1 g of activated ferric hydroxide, made into a slurry with light petroleum, and filtered through a sintered-glass filter. The filter is washed with light petroleum to remove the fat, and the dye is liberated from the residue by dissolving the ferric hydroxide in warm 18% HCl and extracting with ethyl ether. The ether phase is washed with water, dried over Na_2SO_4, concentrated, and submitted to TLC on Kieselgur G-0.5N oxalic acid (2:5), with light petroleum (boiling-range 50C to 70C) or hexane as developing solvent. R_F values are given for 22 fat-soluble dyes. Carotenoids are isolated from the original light petroleum filtrate by adsorption on fuller's earth, from which they are eluted with acetone.

RAPID METHOD FOR THE DETERMINATION OF THE EGG CONTENT OF MAYONNAISE AND SALAD CREAM. R. E. Fresenius (Fresenius Chem. Lab., Wiesbaden, Germany). *Z. Anal. Chem.* 229(5), 353–5 (1967) (in German). The method is based on the liberation from lecithin of PO_4^{3-}, which is determined spectrophotometrically by the molybdenum-blue method.

CHARACTERIZATION OF 3-OXO-Δ^4-STEROIDS BY GAS CHROMATOGRAPHY OF ENOL HEPTAFLUOROBUTYRATES. J. Chamberlain (Dept. of Obstet. and Gynaecol., Charing Cross Hosp. Med. Sch., London, England). *J. Chromatog.* 28(2), 404–5 (1967). The steroid (≃2 mg) was heated with benzene (0.1 ml) plus heptafluorobutyric anhydride (0.1 ml) at 65C for 30 minutes. After removal of the reagent and solvent *in vacuo* at 100C, the residual heptafluorobutyrate derivative was dissolved in acetone for GLC on 1% of SE-30 at 190C or on 3% of QF-1 at 240C with use of a chromatograph equipped with a flame ionization detector. Single peaks were obtained for the derivatives of 17 steroids.

TECHNIQUES IN GAS CHROMATOGRAPHY. I. CHOICE OF SOLID SUPPORTS. J. F. Palframan and E. A. Walker (Ministry of Technol., Lab. of the Government Chemist, Cornwall House, Stamford Street, London, S.E.1). *Analyst* 92, 71–82 (1967). A review consisting of 66 references of work done on solid supports since the last general review by Rose in 1959 is reported. The authors review the work by types of support such as diatomaceous earths, fluorocarbons, porous polymer beads, etc.

BUILD 'DAIRY' FOODS TO ORDER. D. E. Miller and J. V. Ziemba (Durkee Famous Foods, Div. Glidden Co.). *Food Eng.* 38(8), 97–107 (1966). The trend towards developing and marketing sophisticated substitutes for natural food products is discussed. Typical are the dairy-simulated products formulated with advanced lipid systems (fats and emulsifiers). These dairy substitutes usually cost less than natural dairy products and may have longer shelf-life. Custom-blended fats and new emulsifiers make it possible to extend the product line by special lipid systems.

EMULSIFYING CAPACITIES AND EMULSION STABILITY OF VARIOUS MEAT TRIMMINGS. R. J. Borton, N. B. Webb and L. J. Bratzler (Dept. Food Sci., Mich. State Univ., East Lansing, Mich.). *Food Technol.* 22, 506–8 (1968). The emulsifying capacities of 13 commercial sausage meat trimmings were evaluated. The leaner products (higher percent protein) had higher fat emulsifying capacities per unit weight of sample. However, the fatter products indicated a more efficient emulsification by the protein, as these products had higher emulsifying capacities per unit of protein. Chopping the meat trimmings with salt and water 18–24 hr before incorporation into a sausage item enhanced the emulsifying capacity per unit of protein.

SEPARATION OF CIS- AND TRANS-ISOMERS OF α-UNSATURATED ACIDS BY THIN-LAYER CHROMATOGRAPHY. S. P. Dutta and A. K. Barua (Dept. of Chem., Bose Inst., Calcutta, India).

J. Chromatog. 29(1), 263–4 (1967). The separation of isomers of such acids of plant origin (e.g., angelic and tiglic acids) has been accomplished on 0.35-mm layers of Silica gel G impregnated with ammoniacal $AgNO_3$, with use of $CHCl_3$-methanol (19:1, 9:1, or 2:1) as solvent.

GAS-LIQUID CHROMATOGRAPHY OF STEROID GLUCURONOSIDES. W. J. A. Vanden Heuvel (Merck Inst. for Therapeutic Res., Rahway, N.J., U.S.A.). *J. Chromatog.* 28(2), 406–8 (1967). The β-D-glucopyranosiduronic acids derived from 3α-hydroxyandrostan-17-one and 3β-hydroxyandrost-5-en-17-one were used as test compounds. Methylation of the carboxygroup and trimethylsilylation of the hydroxy-groups of the sugar part of the molecule was carried out, and the reaction products and reference standards of the parent steroids and their trimethylsilyl derivatives were examined by GLC in a glass U-tube (6 ft. × 4mm) containing 1.5% of SE-30 on Gas-Chrom P (100 to 120 mesh) at 250C and in a glass U-tube (3 ft. × 4mm) containing 2% of NGS on Gas-Chrom P (80 to 100 mesh) at 240C.

DETERMINATION OF THE NUMBER OF OXYGEN SUBSTITUENTS OF STEROIDS BY CHROMATOGRAPHY. D. J. H. Trafford and R. W. H. Edwards (Institute of Child Health, Univ. of London, 30 Guilford Street, London, W.C.1). *Analyst* 93, 453–55 (1968). The difference in retention volume values obtained by difference of R_m values in the presence and absence of formaldehyde are shown to group in a manner determined by the number of polar functional groups and, to a lesser degree, by the nature of the steroid skeleton. It is proposed that determination of the R_m value provides a means of characterizing steroids from natural sources.

MELTING POINT OF EDIBLE SOLID FATS. I. COMPARISON OF OPEN TUBE MELTING POINT AND POLYMORPHISM. Masao Imamura, Isao Niiya, Hiroshi Iijima, Masakazu Okada and Taro Matsumoto. *Yukagaku* 17, 610–16 (1968). The correlation between open-tube melting point and polymorphism in these fats were studied by use of seven samples of palm oil, coconut oil, hydrogenated soybean oil, beef tallow, lard, and hydrogenated whale oil after standing at 0, 10, 20, 30 and 40C for 1, 5, 24, 120 and 480 hours. The higher the standing temperature, the greater was the progress of polymorphism, approaching the stable form in the fats of higher melting point. When left at a low temperature, there was no change in polymorphism. Fatty oils and hardened oils of high melting point showed slow polymorphism, and the modification started from the unstable form of lower melting point. Symmetrical glycerides of high melting point showed fast polymorphism but those with complex composition of fatty acids or with a high molecular weight chain showed polymorphism. For this reason, transition occurred in the descending order from lard, to palm oil, to beef tallow, and the difference in the melting point according to the standing temperature was greatest in the case of beef tallow. Scattering of the melting point became smaller as the standing temperature became higher and as the time of standing longer.

RADIO ACTIVATION ANALYSIS OF TRACE ELEMENTS IN SOYBEAN OILS. II. DETERMINATION OF COPPER AND ZINC. Yoshio Shinbori and Toshio Tamachi (Atomic Research Lab., Musashi Inst. of Technol., Kawasaki, Japan). *Yukagaku* 17, 606–10 (1968). Copper and zinc in refined soybean oils were determined by neutron activation analysis followed by radio chemical purification of resulting radioactive copper and zinc and those were separated by the Cu-salicyladoxime and Zn-quinaldate precipitation method. The sensitivity limit of detection was 10^{-9} g for Cu and 10^{-8} g for Zn.

GAS-LIQUID CHROMATOGRAPHY OF WOOD EXTRACTIVES. Akira Sato (Kyoto Univ.). *Yukagaku* 17, 599–605 (1968). A review.

DIET MARGARINES: FAT CONTENT, SERVING PORTION AND ACCEPTANCE. E. R. Monsen, P. B. Crawford and D. W. Lowe (Univ. of Washington, Seattle, Wash.). *J. Am. Dietetic Assoc.* 54, 29–31 (1969). Chemical analysis of the three "diet" margarines available on the market indicate half the fat content and thus half the calories of two standard margarines tested. Less than half the subjects were able to discriminate between the diet and regular margarines. Those who could discriminate preferred the diet margarine on hot string beans and the regular margarine on toast and bread. It is concluded that diet margarines may be an effective, though expensive, way to make a modest decrease in caloric consumption.

ZIRCONIUM AND OTHER METALLIC DRIERS IN POLISHES. P. G. Chantrell, F. Pitts and T. F. Smith (Magnesium Elektron, Ltd.). *Soap Chem. Specialties* 44(10), 50–6, 89–90 (1968). Metallic soaps (especially zirconium) constitute an excellent medium for the provision of metal ions in waxes and polishes since they are compatible with the other formulation ingredients. Metal ions serve the function of a cross-linking agent, acting through the formation of salts and salt bridges with the carboxylic groups of the organic film-former to create a coherent three-dimensional coating. These soaps are also of value in polish formulations because of their surfactant properties.

PROCESS FOR CONTINUOUSLY DEACIDIFYING GLYCERIDE OILS. W. de Man (Lever Bros. Co.). *U.S. 3,419,588.* A process is described for the continuous neutralization of glyceride oils containing free fatty acids and other impurities while avoiding substantial emulsification and saponification. The process comprises flowing a thin layer of the oil upon a horizontal or slightly inclined surface while spraying on it small droplets of an aqueous alkaline liquid, maintaining the co-current flow of the resultant substantially non-turbulent mixture until neutralization occurs and thereafter separating the resultant neutral oil from the resultant soap- and impurity-containing aqueous phase.

• Fatty Acid Derivatives

ANALYSIS OF PLASTICIZERS BY GAS-LIQUID CHROMATOGRAPHY. Yasuo Choshi, Shoji Tani, Giichi Akazome and Koichi Murai (New Japan Chem. Co., Kyoto). *Yukagaku* 17, 461–6 (1968). Various phthalates and aliphatic diesters were analyzed by gas-liquid chromatography quantitatively and qualitatively. Two columns of lower liquid phase content, such as 1.4% neopentylglycol sebacate or 1.2% SE-30 on Diasolid-M, were used to separate the esters having higher boiling point. By using the internal standard method, those esters were separated quantitatively with good accuracy. The vapor pressures of several esters could be estimated from relative retention time against the standard substances such as diethyl phthalate or dioctyl phthalate.

THE HYDROLYSIS OF MONOSTEARIN IN AN ACIDIC MEDIUM. N. A. Armstrong (Welsh Sch. of Pharm., Univ. of Wales Inst. of Sci. and Tech., Cardiff, Wales). *J. Soc. Cosmetic Chemists* 19, 807–711 (1968). The hydrolysis of pure and commercial grades of monostearin in mineral acid has been studied. The reaction has been shown to follow first order kinetics, with an activation energy of 76.49 kJ $mole^{-1}$, and a collision factor of 9.48 min^{-1}. Study of the hydrolysis of pure monostearin in acidic buffer solutions also reveals first order kinetics. The rate of reaction increases as the pH of the medium is decreased.

SYNTHESIS OF CYCLIC N,O- AND N,S-HETEROCYCLIC COMPOUNDS BY CONDENSATION OF FATTY ACID ALKYLOAMIDES WITH CARBONYL COMPOUNDS AND PHOSPHOROUS PENTASULFIDE RESPECTIVELY. K. Thewalt and G. Renckhoff (Sci. Sec. of Dynamit Nobel Co., Werk Witten). *Fette Seifen Anstrichmittel* 70, 648–653 (1968). Fatty acid alkylolamides of the type $RCONHCHR_1(CH_2)_nCHR_2OH$ (R: alkyl residue with 2 to 17 carbon atoms, cycloalkyl or aryl residue; R_1 and R_2: alkyl residue with 1 to 3 carbon atoms or hydrogen; and n = 0–2) are converted by acidic condensation with lower aliphatic or aromatic aldehydes and ketones respectively to mono- and disubstituted N-acyloxazolidines, N-acyl-1,3-tetrahydrooxazines and N-acyl-1,3-hexahydrooxazepines. Furthermore, new types of 2-alkyl-4,5-dihydro-6H-1,3-thiazine (n = 1) besides monoalkyl substituted delta-2-thiazolines (n = 0) were obtained from fatty acid alkylolamides by condensation with P_2S_5. A large number of the compounds prepared show excellent bacteriostatic and fungistatic action.

INFLUENCE OF SUBSTITUTION AND THE EFFECT OF CHAIN LENGTH ON FATTY ACID–THIONYL CHLORIDE REACTION. H. N. Singh and M. A. Bey (Dept. Chem., Aligarh Muslim Univ., Aligarh, India). *Fette Seifen Anstrichmittel* 70, 640–642 (1968). Kinetic data are reported for the halogenation of the first

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five members of the fatty acid series. The influence of substitution of electron withdrawing and electron releasing group in the methyl radical of acetic acid was studied in solution. The mechanism of the reaction is discussed.

QUANTITATIVE CONVERSION OF BETA-KETO-FATTY ACID ESTERS TO METHYL KETONES. J. Strobach and W. Geib (Inst. for Nutr. Chem. and Tech., Humboldt Univ., Berlin, W. Ger.). *Fette Seifen Anstrichmittel* 70, 638–640 (1968). A method by which beta-keto-fatty acids can be converted to corresponding methyl ketones and quantitatively determined is described.

SEPARATION AND IDENTIFICATION OF 2,4-DINITROPHENYLHYDRAZONES OF UNSATURATED ALDEHYDES AND METHYLKETONES BY THIN-LAYER CHROMATOGRAPHY. P. W. Meijboon (Unilever Res. Lab., Vlaardingen, Netherlands). *Fette Seifen Anstrichmittel* 70, 477–481 (1968). The saturated aldehydes and ketones formed in small amounts by autoxidation of oils and fats were separated as their 2,4-dinitrophenylhydrazones by a combination of partition, adsorption and silver nitrate-thin layer chromatography.

KINETIC STUDIES ON THE ESTERIFICATION OF ACID AND ALCOHOL IN THE PRESENCE OF DOWEX 50 W AND OF SULFURIC ACID. Ting-Chia Huang, J. Wan and Lung-Hai Tsai (Dept. of Chem. Eng., Taiwan Provincial Cheng Kung Univ., Tainan). *J. Chinese Chem. Soc.* 14, 72–87 (1967). Several kinds of alcohols have been chosen to react with succinic acid, and several organic acids have been chosen to esterify with n-butyl alcohol using either Dowex 50 W or sulfuric acid as catalyst. The rate constants are influenced by the temperature, the structure of reactants, and kinds of catalyst; the activation energies are also varied with different reactants and catalysts. The catalytic effect of hydrogen ion is not simple, and the possibility of exciting either the acid or the alcohol to its activated state was affected by their molecular structuress.

GAMMA-RAY INDUCED ESTERIFICATION OF SUCROSE AND FATTY ACIDS. Ung-Ping Wang (Rad. Lab., Union Ind. Res. Inst., Ministry of Economic Affairs). *J. Chinese Chem. Soc.* 2, 70–8 (1968). Gamma-ray induced esterification of sucrose with equimolar amounts of various fatty acids has been investigated at the room temperature. Esterification of sucrose was easily carried out with fatty acids of lower molecular weight and the reaction was retarded severely as the molecular weight of fatty acid becomes larger. When a mixture of 100 g sucrose and 140 g glacial acetic acid was irradiated with the total gamma-ray dose of 4.75×10^6 röntgen with 0.5 ml 98% H_2SO_4 catalyst, a yellowish-brown, water-soluble viscous liquid (86 g) was produced, giving a high yield of 98.1% (based on reacted sucrose). The product is a superior foaming agent in water solution, although the foams fade easily. The mechanism of esterification of sucrose with glacial acetic acid has been investigated. The rate of reaction was found to follow the 2nd-order equation.

KINETIC STUDIES ON THIONYL CHLORIDE FATTY ACID SYSTEM: PART III. HALOGENATION OF DIFFERENT FATTY ACIDS IN DIFFERENT SOLVENTS. H. N. Singh and M. A. Beg (Chem. Labs., Aligarh Muslim Univ., Aligarh, India). *J. Indian Chem. Soc.* 45, 237–42 (1968). Kinetic data are reported for the halogenation of acetic, propionic, butyric and valeric acids, in chloro, bromo, and nitrobenzene between 20 and 40C. The behaviour of solvent and mechanism of the reaction has been discussed in terms of parameters of absolute reaction rate equation.

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EFFECT OF LIPIDS, IN PARTICULAR CHOLESTERYL 14-METHYLHEXADECANOATE, ON THE INCORPORATION OF LABELLED AMINO ACIDS INTO TRANSFER RIBONUCLEIC ACID IN VITRO. J. Hradec and Z. Dusek (Dept. of Biochem., Oncological Inst., Prague 8, Czechoslovakia). *Biochem. J.* 110, 1–8 (1968). Rat liver pH 5 enzymes and cell sap extracted with various organic solvents showed a variable decreased incorporation of labelled amino acids into S-RNA ('soluble' or transfer RNA) *in vitro*. The original enzymic activity could be fully restored, though at different rates, by the addition of lipid extracts in quantities corresponding to those originally present. Of the main lipid groups separated from the extract, only free cholesterol and cholesteryl esters were able to reactivate the extracted pH 5 enzymes in the same way as the whole lipid extract. Addition of pure cholesteryl 14-methylhexadecanoate also fully restored the enzymic activity. There was no energy dependent incorporation of labelled amino acids into ribosomal protein in the presence of extracted cell sap. Addition of cholesteryl 14-methylhexadecanoate fully restored the activity of the cell sap to incorporate labelled leucine and lysine into ribosomal protein and enhanced the incorporation of labelled protein hydrolysate and phenylalanine over the level found with the corresponding non-extracted preparations. It is concluded that lipids play an important role in the synthesis of aminoacyl-S-RNA complexes and that cholesteryl 14-methylhexadecanoate may be the active lipid in this respect.

EFFECTS OF MAGNESIUM IONS, ADENOSINE TRIPHOSPHATE, PALMITOYLCARNITINE AND PALMITOYL COENZYME A ON ACETYL COENZYME A CARBOXYLASE. M. D. Greenspan and J. M. Lowenstein (Grad. Dept. of Biochem., Brandeis Univ., Waltham, Mass. 02154). *J. Biol. Chem.* 243, 6273–80 (1968). Acetyl coenzyme A carboxylase is activated strongly by incubation with magnesium ions in the absence of citrate and by incubation with citrate in the absence of magnesium ions. The activation by magnesium ions is concentration and time dependent. Under optimum conditions the effects of citrate and magnesium ions are not additive. Although the activation by magnesium ions occurs in the absence of citrate, the enzyme requires citrate for the complete reaction. Addition of adenosine triphosphate to the activation medium containing either magnesium ions or citrate or both strongly inhibits the activation of acetyl coenzyme A carboxylase. The inhibition is not due to removal of magnesium ions by chelation with adenosine triphosphate. Enzyme which has been activated with magnesium ions sediments at about the same rate as the inactive enzyme. Palmitoyl carnitine and cetyl trimethylammonium ions also stimulate the activity of the enzyme. This type of activation occurs rapidly, unlike the activation of the rat liver enzyme by magnesium ions and citrate. The maximum degree of stimulation by palmitoyl carnitine is observed when acetyl coenzyme A carboxylase has not been fully activated with other activators, but a 2- to 3-fold stimulation is observed even with enzyme which has been activated optimally with magnesium ions and citrate.

STEROID-PROTEIN INTERACTIONS. XVII. INFLUENCE OF SOLVENT ENVIRONMENT ON INTERACTION BETWEEN HUMAN α_1-ACID GLYCOPROTEIN AND PROGESTERONE. M. Ganguly and U. Westphal (Biochem. Dept., Univ. of Louisville, School of Med., Louisville, Ky. 40208). *J. Biol. Chem.* 243, 6130–9 (1968). The apparent binding affinity between progesterone and α_1-acid glycoprotein (AAG) is increased by neutral salts, such as Na_2SO_4, $(NH_4)_2SO_4$, or NaCl, which generally stabilize the conformational structure of globular proteins; other perturbants which are known to destabilize globular proteins, e.g. LiBr, $CaCl_2$ or urea decrease the apparent stability of the complex. The intrinsic viscosity of AAG in 3 M $CaCl_2$ and in 4 M NaCl is greater and smaller, respectively, than in water; this is in agreement with the assumption of a conformational change toward a random coil and a more compact structure, respectively, in the two salt solutions. The order of efficiency of the perturbants in increasing or decreasing the apparent progesterone-AAG affinity is similar to the Hofmeister series. The progesterone-binding affinity is independent of the presence of sialic acid in the AAG molecule.

INTERACTIONS OF OBESITY, AND GLUCOSE AND INSULIN LEVELS IN HYPERTRIGLYCERIDEMIA. S. Ford, Jr., R. C. Bozian, and H. C. Knowles, Jr. (Dept. Med., Univ. of Cincinnati, Coll. of Med., Cincinnati, Ohio). *Am. J. Clin. Nutr.* 21, 904–10 (1968). Patients who have high plasma triglyceride levels will usually be overweight. Albrink and her associates have suggested that hypertriglyceridemia is associated with acquired rather than constitutional obesity. They found that weight gained after age 25, or truncal obesity, correlated well with triglyceride levels, whereas forearm fatness reflected constitutional obesity and was not correlated with triglyceride levels. Similarly, Harlan *et al.* found that triglyceride levels and concentrations of the triglyceride-rich very low density lipoproteins (S_f 20–400) were significantly correlated positively with weight gain after age 24 and measurements of truncal obesity tended to have a negative correlation with upper arm fatness. Both of these population studies indicate that acquired adiposity is associated with elevation of triglyceride levels. Abnormal glucose tolerance also is commonly associated with hypertriglyceridemia. Among cases of familiar hypertriglyceridemia more than 90% were found to have abnormal glucose tolerance. In some reports elevated triglyceride levels were not associated with abnormal oral glucose tolerance but were associated with impaired intravenous glucose tolerance or abnormal tolbutamide tolerance tests. Several investigations have revealed high insulin levels in hypertriglyceridemic patients.

FAILURE OF PLASMA PROTEIN LEVEL TO INDICATE DEVELOPING FATTY LIVER IN CHICKENS. M. J. Duke, R. K. Ringer and J. H. Wolford (Dept. of Poultry Sci., Michigan State Univ., East Lansing, Mich. 48823). *Poultry Sci.* 47, 1098–100 (1968). Plasma protein level was not a satisfactory index of developing fatty liver condition. There was no significant difference in plasma protein level between two dietary groups until a fatty liver condition was well developed in one group.

EFFECT OF PYRIDOXINE DEFICIENCY OF LIPID METABOLISM IN THE CHICK. N. J. Daghir and J. M. Porooshani (Faculty of Agricultural Sci., American Univ. of Beirut, Lebanon). *Poultry Sci.* 47, 1094–98 (1968). Three dietary variables, fat (4% corn oil), pyridoxine HCl (8 mg/kg.) and cholesterol (1%) were incorporated into eight different treatments using a basal low-fat, low pyridoxine semi-purified diet. Ten, day-old male chicks were used per treatment and kept on their respective diets for a period of three weeks. Criteria studied were body weight, serum cholesterol, liver fat and cholesterol, and fatty acid composition of abdominal fat. Dietary cholesterol significantly increased serum and liver cholesterol. Pyridoxine deficient birds had higher serum cholesterol values than pyridoxine adequate birds except when both fat and cholesterol were added. Pyridoxine deficient birds had higher stearic acid than pyridoxine adequate ones.

EFFECTS OF METHYLPREDNISOLONE ON PLASMA LIPIDS AND AORTIC MUCOPOLYSACCHARIDES OF NORMAL AND CHOLESTEROL-FED RABBITS. R. Alper, A. Rubulis, J. T. Prior and W. R. Ruegamer (Depts. of Biochem., Med. and Path., State Univ. of New York, Upstate Medical Center, Syracuse, N.Y. 13210). *Proc. Soc. Exp. Biol. Med.* 129, 623–7 (1968). Methylprednisolone (Medrol) was administered to normal and cholesterol-fed rabbits for a period of 2 months and its effects upon plasma cholesterol and triglyceride concentrations and upon the content of aortic acid mucopolysaccharides (AMPS) were studied. Rabbits fed a chow diet developed both a hypercholesterolemia and a hypertriglyceridemia in response to Medrol, and the already elevated cholesterol and triglyceride levels induced by cholesterol feeding were further elevated by Medrol. Despite the hyperlipemia, none of the Medrol-treated animals developed aortic atherosclerosis. Medrol had no significant effect upon the total aortic AMPS content in control animals but it did alter the aortic AMPS content and pattern of cholesterol-fed rabbits. The most significant effect was an increase in an AMPS of low sulfate content. Since all animals fed cholesterol developed some degree of pulmonary atherosclerosis, it is postulated that the anti-atherogenic action of the hormone on the aorta is more closely related to alterations in the AMPS pattern than to changes in the pattern of circulating lipids.

EFFECTS OF A SKIMMED MILK AND CHOCOLATE DIET ON SERUM AND SKIN LIPIDS. I. MacDonald (Guy's Hospital Med. School, London, England). *J. Sci. Food Agr.* 19, 270–2 (1968). A diet containing 28 g. chocolate per kg body weight together with skimmed milk powder was given each 24 hrs. to 14 men and 15 women aged 19–21 years. After five days on the diet the fasting serum cholesterol and triglyceride values were compared with values obtained before the diet commenced. Similar comparisons were made for the cholesterol and triglyceride levels on the skin of the face. There was no overall change in serum triglyceride level in the males and a small but significant rise (5.3 mg/100 ml) in the females. A significant correlation was found between control triglyceride level of the male subjects and the amount of change measured in the experiment. Subjects with a low initial level of serum triglycerides experienced an increase, while the level actually decreased in subjects with a high initial value. The same correlation was not found to apply to the female subjects. The serum cholesterol level decreased significantly (by 14 mg/100 ml) in the males, but was unchanged in the females.

PATHWAYS FOR ANDROGEN BIOSYNTHESIS FROM (7α-^{3}H)PREGNENOLONE AND (4-^{14}C)PROGESTERONE BY RAT TESTIS INTERSTITIUM IN VITRO. Janet Bell, G. P. Vinson, D. J. Hopkin and D. Lacy (Dept. of Zool. and Compar. Anatomy, St. Bartholomew's Hosp. Medical College, Charterhouse Square, London, E.C. 1). *Biochim. Biophys. Acta* 164, 412–20 (1968). Rat testis interstitial tissue, freed from tubule structures, and whole testis tissue were incubated with (7-α-^{3}H)pregnenolone and (4-^{14}C)progesterone. Interstitial tissue was also incubated with (7-α-^{3}H)testosterone and (4-^{14}C)androstenedione. Samples of the incubation media were withdrawn after various intervals of time, and the products isolated and identified. Their yields from the precursors were calculated and plotted against time. The yield vs. time curves were interpreted on the basis that sequential maxima in isotope incorporation in the different products reflected the order in which the compounds occurred in the biosynthetic pathway. In this way it was deduced that under the conditions chosen, the preferred pathway for the formation of testosterone from pregnenolone was pregnenolone . . . progesterone . . . 17α-hydroxyprogesterone . . . androstenedione . . . testosterome. 17α-Hydroxypregnenolone was not formed in significant amount and other possible pathways appeared to be unimportant. Testosterone was invariably metabolized to further products, as yet unidentified, and therefore did not reach equilibrium with androstenedione. Possible explanations are offered for apparent discrepancies in the literature.

STUDIES ON ESTROGEN REGULATION OF CHOLESTEROL BIOSYNTHESIS IN RAT LIVER MICROSOMES. S. Mukherjee and A. Bhose (Labs. of Lipid Res., Dept. of Applied Chem., Univ. of Calcutta, Calcutta, India). *Biochim. Biophys. Acta* 164, 357–368 (1968). An inhibition of cholesterol biosynthesis from acetate has been demonstrated in the liver of rats previously administered 100 μg (per 100 g rat) of 17β-estradiol for periods varying from 4 to 56 days. The site of estrogen control of hepatic sterol synthesis has been investigated in microsomal preparations. The inhibitory effect of exogenous estrogen is observed mostly at premevalonate stages of biosynthesis; the conversion of mevalonate to squalene or of squalene to cholesterol is not significantly altered by hormonal administration. The first estrogen-sensitive step in the pathway of biosynthesis appears to be the condensation of acetoacetyl-CoA and acetyl-CoA, since markedly reduced activity of the hydroxymethylglutaryl-CoA-condensing enzyme has been demonstrated in liver microsomes isolated from treated animals. A decrease in the microsomal hydroxymethylglytaryl-CoA reductase activity also results from the hormone treatment. The overall depression in the rate of cholesterol synthesis is determined by the hormonal inhibition of mevalonate synthesis in microsomes. Estrogen has very little influence on hydroxymethylglutaryl-CoA-cleavage enzyme activity.

MICROBIAL HYDROXYLATIONS. III. 11α-HYDROXYLATION OF SOME 3α,5α-CYCLOPREGNANE DERIVATIVES. L. Tan and L. L. Smith (Dept. of Biochem., Univ. of Texas Med. Branch, Galveston, Texas 77550). *Biochim. Biophys. Acta* 164, 389–95 (1968). The influence of subtle structural changes in the A/B ring system of pregnane-20-ketones on the course of microbial 11α-hydroxylation by *Aspergillus ochraceus* was examined by fermentation of the cyclopregnanes, 6β-hydroxy-3α,5α-cyclopregnan-20-one and 3α,5α-cyclopregnan-6,20-dione, and of pregnenolone, pregnenolone cyclic ethylene ketal, and progesterone bis ethylene ketal. The cyclopregnanes were hydroxylated in the 11α-position; the ethylene ketals were not transformed; pregnenolone was dehydrogenated to progesterone prior to further hydroxylation.

GLYCOSYL DIGLYCERIDES FROM PSEUDOMONAS RUBESCENS. S. G. Wilkinson (Dept. of Chem., The Univ., Hull, Yorkshire G. B.). *Biochim. Biophys. Acta* 164, 148–156 (1968). Glycolipids containing D-glucose and D-glucuronic acid have been isolated from *Pseudomonas rubescens*. The lipids belong to the class of 1–0 monoglycosyl diglycerides. In both lipids the glycosidic linkage had the β configuration. Component fatty acids, which were the same for both lipids, ranged from C_{13} to C_{19} and included saturated, monoenoic, and branched-chain acids.

CHEMICAL SYNTHESIS OF 1-O-(D)- AND 3-O-(L)-GLYCERYL MONOETHERS, DIETHERS AND DERIVATIVES: GLYCERIDES, MONOESTER PHOSPHOLIPIDS AND DIETHER PHOSPHOLIPIDS. G. K. Chacko and D. J. Hanahan (Dept. of Biochem., Univ. of Washington, Seattle, Wash. and Dept. of Biochem., College of Med. Univ. of Arizona, Tucson, Ariz.). *Biochim. Biophys. Acta* 164, 252–271 (1968). A convenient route to the synthesis of 1-O-glyceryl ether from 3-O-glyceryl ether is reported. This technique, which is based on the procedure of Lands and Zschocke for conversion of 3-O-benzylglycerol to 1-O-benzylglycerol, utilized either 3-O-hexadecylglycerol or 3-O-octadecylglycerol as the starting material. The ether was converted to its ditosylate and this latter group was displaced with freshly fused 1-O-alkyl glycerol, with an equal but opposite optical rotation to the starting material, in good yields. 1-O-cis-9′-Octadecenyl-2-O-octadecanoylglycerol was synthesized by borate replacement of 1-O-cis-9′-octadecenyl-2-O-octadecanoyl-3-O-tritylglycerol in triethyl borate and boric acid, followed by column chromatography on neutral silicic acid. Catalytic amounts of $HClO_4$ isomerizes 2-stearoyl selachyl alcohol completely to its positional isomer 3-stearoyl selachyl alcohol Acylation of 1-O-cis-9′-octadecenylglycerol in the presence of up to 2 M excess of acyl chloride was found to occur preferentially at the primary hydroxyl group forming 3-stearoyl selachyl alcohol. The diacyl derivatives formed

in good yield only when a 4–6M excess of acyl chloride was used. The properties of the synthetic diacyl glyceryl derivatives correlated closely with the natural diacyl glyceryl monoether isolated from dog fish liver oil.

INHIBITION OF PHOSPHOLIPASE C BY PHOSPHONATE ANALOGS OF GLYCEROPHOSPHATIDES. A. F. Rosenthal and M. Pousada (Dept. of Labs., Long Island Jewish Hosp., New Hyde Park, N.Y. 11040). *Biochim. Biophys. Acta* 164, 226–237 (1968). The inhibition of *Clostridium perfringens* phospholipase C (phosphatidylcholine cholinephosphohydrolase, EC 3.1.4.3) hydrolysis of lecithin by eight synthetic phosphonate-containing analogs of lecithin, cephalin, and phosphatidic acid was studied. There was a wide variation among the inhibitory activities; the lecithin analogs as a group were the most active. The two most effective inhibitors found were 3,4-dioctadecoxybutylphosphonylcholine and 2-hexadecoxy-3-octadecoxypropylphosphonylcholine. Except for a lag phase in the inhibited reactions, both of these substances gave relatively simple kinetics, including ordinary competitive inhibition of the enzyme as the inhibition mode. These two lecithin analogs appear to exert little effect on the ζ-potential of lecithin particles. Interpretations of these studies and potential utility of the inhibitors are discussed.

PHOSPHOLIPASE IN ARTERIAL TISSUE. II. PHOSPHATIDE ACYL-HYDROLASE AND LYSOPHOSPHATIDE ACYL-HYDROLASE ACTIVITY IN HUMAN AND RAT ARTERIES. S. Eisenberg, Y. Stein and O. Stein (Lipid Res. Lab., Hadassah Univ. Hosp. and Dept. of Experimental Med. and Cancer Res., Hebrew Univ. Hadassah Med. School, Jerusalem, Israel). *Biochim. Biophys. Acta* 164, 205–214 (1968). Homogenates of human and rat aortae were investigated for phosphatide acyl-hydrolase activity, using biosynthetically labeled rat liver lecithin. The products of hydrolysis were identified as lysolecithin and free fatty acid. The enzymic activity was stimulated by Ca^{2+} and by sodium deoxycholate and was inhibited by Triton X-100. The pH optimum ranged from 7.9–8.6 and the enzymic preparation hydrolyzed preferentially the 2 position of lecithin. The arterial enzyme hydrolyzed both endogenous and exogenous lecithin under the same conditions. Lysolecithinase activity in aortae and umbilical arteries could be demonstrated only in the absence of sodium deoxycholate. A significant increase in the lecithinase activity of aortic preparations from older rats was encountered. The findings are discussed with regard to the role of phospholipase in the regulation of arterial phospholipid composition.

ON THE SPECIFICITY OF RAT LIVER LYSOPHOSPHOLIPASE. V. Van Den Bosch, A. J. Aarsman, A. J. Slotboom and L. L. M. Van Deenen (Lab. of Biochem., State Univ. Utrecht, Utrecht, Netherlands). *Biochim. Biophys. Acta* 164, 215–225 (1968). A study on the specificity of rat liver lysophospholipase activity (EC 3.1.15) revealed that both 1-acyl-*sn*-glycero-3-phosphorylcholine and 2-acyl-*sn*-glycero-3-phosphorylcholine are deacylated. From both positional isomers the unsaturated analogs appeared to be degraded at higher rates. Circumstantial evidence is presented indicating that 2-acyl-*sn*-glycero-3-phosphorylcholine can be attacked directly by this lysophospholipase without a prior migration of the fatty acyl constituent. Compounds lacking the free hydroxyl group present in lysophosphatidylcholines, *e.g.* acyl-ethylene glycolphosphorylcholine and 1-acylpropane diol-3-phosphorylcholine, also fall in the enzyme's range of specificity. Monoacyl derivatives of *sn*-glycero-1-phosphorylcholine, *sn*-glycero-2-phosphorylcholine, as well as *sn*-glycero-3-phosphorylcholine, were found to be degraded. Inhibition of lysophospholipase activity by various agents exhibited the same effect on the deacylation of both 1-acyl- and 2-acyl-*sn*-glycero-3-phosphorylcholine. The degradation of monoacyl-phosphatidylcholine appeared to be strongly inhibited in the presence of phosphatidylcholine.

RAT KIDNEY GLYCERYLPHOSPHORYLCHOLINE DIESTERASE. J. J. Baldwin and W. E. Cornatzer (Guy and Bertha Ireland Res. Lab., Dept. Biochem., Univ. of N. Dakota Sch. Med., Grand Forks, N. D. 58201). *Biochim. Biophys. Acta* 164, 195–204 (1968). The characterization of rat kidney glycerylphosphorylcholine diesterase (L-3-glycerylphosphorylcholine glycerophosphohydrolase, EC 3.1.4.2) is described. Of 8 tissues tested, rat kidney shows the greatest activity but substantial amounts are found in spleen and lungs. Considerably lower levels of activity are present in heart, skeletal muscle, liver, brain and intestine. Among subcellular components, the microsomal fraction contained the greatest amount of diesterase. Optimal activity occurs at pH 9.2. The enzyme preparation shows a narrow substrate specificity. Of 6 phosphodiesters tested, only glycerylphosphorylcholine and glycerolphosphorylethanolamine showed any activity. Michaelis constants for these compounds are $2.2 \cdot 10^{-3}$M and $11.5 \cdot 10^{-3}$M, respectively. Identical resistance toward heat inactivation substantiates the common identity of the two activities. The enzyme is inhibited by ethanolamine, *N*-methylethanolamine, *N,N*-dimethylethanolamine and choline. The selective inhibitors of the cholinesterases, prostigmine and physostigmine, do not significantly affect the activity of the diesterase. EDTA inhibits the enzyme. However, it has not been possible to demonstrate the participation of a metal cofactor.

A FURTHER CHARACTERIZATION OF LIPOPROTEIN LIPASE. H. Greten, R. I. Levy and D. S. Fredrickson (Lab. Molecular Diseases, Nat. Heart Inst., Nat. Institutes of Health, Bethesda, Md. 20014). *Biochim. Biophys. Acta* 164, 185–194 (1968). The isolation of hydrolysis products from glyceryl (1-^{14}C) trioleate provided a sensitive and reproducible assay system to measure plasma post-heparin lipolytic activity and adipose tissue lipoprotein lipase (glycerolester hydrolase, EC 31.1.3). Essentially no lipolytic activity was detected in pre-heparin plasma. With this system temperatures lower than 30C were required for linear hydrolysis rates. At 27C zero order kinetics could be obtained for more than 2 hours. Hydrolysis of triglyceride by adipose tissue lipoprotein lipase was also greater at 27C than at 37C. Up to 50% of the original post-heparin lipolytic activity was retained after lyophilization and extraction of plasma with organic solvents, permitting measurement of enzyme activity after nearly complete removal of endogenous substrate, even in patients with gross hypertriglyceridemia. These studies thus suggest two ways that assay of post-heparin lipolytic activity may be improved: the use of lower temperatures to achieve greater stability of the enzymatic activity and delipidation of post-heparin plasma to remove competition from endogenous glycerides.

PLASMA POST-HEPARIN LIPOLYTIC ACTIVITY IN HYPERCHYLOMICRONEMIA (FAT INDUCED LIPEMIA). R. H. Bradford and R. H. Furman (Cardiovascular Section, Oklahoma Med. Res. Found. and Dept. of Biochem. and Med., Univ. of Oklahoma City, Okla.). *Biochim. Biophys. Acta* 164, 172–184 (1968). Low levels of plasma post-heparin lipolytic activity, assayed *in vitro*, were observed in three siblings with hyperchylomicronemia, a heritable disorder characterized by markedly increased levels of serum triglyceride transported as chylomicroms (S_f 5000). Although no evidence of inhibitors was observed in pre- or post-heparin plasma from these hyperchylomicronemic siblings, *in vitro* studies with esterase and lipase inhibitors demonstrated that plasma post heparin lipolytic activity in these siblings was relatively less inhibited by NaCl, sodium deoxycholate, Triton, or sodium dodecyl sulfate than was the post-heparin lipolytic activity in plasma from subjects with other types of hyperglyceridemia or from subjects with normal serum triglyceride concentrations.

EFFECT OF PROSTAGLANDIN AND DIETARY FATS ON LIPOLYSIS AND ESTERIFICATION IN RAT ADIPOSE TISSUE IN VITRO. S. S. Pawar and H. C. Tidwell (Biochem. Dept. Univ. of Texas Southwestern Med. School, Dallas, Texas). *Biochim. Biophys. Acta* 164, 167–171 (1968). A possible role of prostaglandin (PEG_1) in lipid metabolism was investigated. It was found to inhibit the release of free fatty acids and glycerol from adipose tissue of fat-fed rats, the amount depending upon the nature of the fat ingested. The release of less glycerol and free fatty acids from tissue of rats fed a polyunsaturated fat (corn oil) as compared to those fed lard was in effect similar to that in the presence of prostaglandin. Preincubation of tissue with prostaglandin produced a similar response suggesting that prostaglandin may have entered the adipose tissue cells during preincubation causing an inhibition of lipolysis. There was also a greater incorporation of palmitic acid-1-^{14}C into the lipids and triglycerides of this tissue in the corn oil-fed rat and in prostaglandin-treated tissues. An inhibition of lipolysis and increased rate of removal of fatty acids from the medium by adipose tissue in the presence of prostaglandin or by this tissue of corn oil-fed rats, both associated with an increase esterification of fatty acids in the tissue, suggests the possibility that the effect of the latter may be the promotion of the formation by these rats of greater amounts of prostaglandin from the unsaturated fatty acids ingested.

SHORT-CHAIN FATTY ACID ACTIVATION IN RAT LIVER; A NEW ASSAY PROCEDURE FOR THE ENZYMES AND STUDIES ON THEIR INTRACELLULAR LOCALIZATION. Magne Aas and J. Bremer (Inst. of Clin. Biochem., Univ. of Oslo, Rikshospitalet, Oslo). *Biochim. Biophys. Acta* 164, 157–166 (1968). Acetyl-CoA synthetase (acetate:CoA ligase (AMP), EC 6.2.1.1) and medium chain acyl-CoA synthetase (acid:CoA ligase AMP, EC 6.2.1.2) have been studied in rat-liver homogenate and cellular subfractions. A new assay procedure for the enzymes

has been worked out. The acyl-CoA formed is transformed into acylcarnitine by excess amounts of acetyl-CoA:carnitine acetyl transferase (EC 2.3.1.7) in the presence of radioactive carnitine. With differential centrifugation of rat-liver homogenate, it has been shown that the medium-chain acyl-CoA synthetase is exclusively localized in mitochondria, and by subfractionation of the mitochondria we found that the enzyme is confined to the matrix. The acetyl-CoA synthetase apparently has a bimodal intracellular localization, the enzyme being localized both in mitochondria and in the particle-free supernatant. The mitochondrial part is confined to the matrix.

THE ALK-1-ENYL ETHER AND ALKYL ETHER LIPIDS OF BOVINE HEART MUSCLE. H. H. O. Schmid and T. Takahashi (Univ. of Minnesota, The Hormel Inst., Austin, Minn. 55912). *Biochim. Biophys. Acta* 164, 141–147 (1968). Choline phosphatides and ethanolamine phosphatides as well as neutral alkoxylipids and triglycerides were isolated from a lipid extract of bovine heart muscle. The constitutent alk-1-enyl glycerol ethers, alkyl glycerol ethers and fatty acids of these lipid classes were analyzed after selective acidic or enzymatic hydrolysis. Comparative analyses of the aliphatic moieties at the 1 and 2 positions of the phosphatides revealed a pronounced similarity of the three classes of choline phosphatides, whereas significant differences between the various ethanolamine phosphatides were encountered. Alk-1-enyl diglycerides and alkyl diglycerides differed in the composition of their aliphatic moieties from each other, from the triglycerides and from the phosphatides.

THE STRUCTURES OF ENZYMICALLY PRODUCED DIPHOSPHOINOSITIDE AND TRIPHOSPHOINOSITIDE. C. Prottey, J. G. Salway and J. N. Hawthorne (Dept. Med. Biochem. and Pharmacol., Univ. Birmingham, Birmingham, G. B.). *Biochim. Biophys. Acta* 164, 238–251 (1968). The structures of enzymically produced diphosphoinositide and triphosphoinositide have been determined. Phosphatidylinositol kinase of rat brain produces diphosphoinositide having the inositol-1,4-diphosphate structure. Diphosphoinositide kinase of rat brain produces triphosphoinositide having the 1,4,5-triphosphate structure. Rat-brain triphosphoinositide phosphomonoesterase forms the 1,4-diphosphoinositide. Wheat phytase (EC 3.1.3.8) also hydrolyzes triphosphoinositide to the 1,4-diophosphoinositide. The alkaline phosphatase (EC 3.1.3.1) of calf intestine hydrolyzes triphosphoinositide to the 1,5-diphosphoinositide, a compound not previously described. The natural diphosphoinositide of ox brain has the 1,4 structure.

HYDROLYSIS OF SYNTHETIC CHOLESTEROL ESTERS CONTAINING TRANS FATTY ACIDS. D. Sgoutas (Burnsides Res. Lab., Urbana, Illinois 61801). *Biochim. Biophys. Acta* 164, 317–326 (1968). The aim of this work was to test the specificity of rat liver cholesterol esterase(sterol-ester hydrolase, EC 3.1.1.13) with regard to the hydrolysis of *cis* and *trans* unsaturated cholesterol esters. Several synthetic cholesterol esters were employed as model substrates for the soluble fraction and microsomes from rat liver homogenates. In all cases, *trans* fatty acid cholesterol esters were hydrolyzed to a lesser degree than *cis* unsaturated esters. Furthermore, a preference for the 9-*cis*-unsaturated octadecanoates was indicated. The ability of cholesterol esterase to recognize the shape of the acyl moiety of cholesterol esters is discussed.

THE BIOSYNTHESIS OF CYCLIC CAROTENOIDS IN RHODOTORULA MUCILAGINOSA AND RHODOTORULA AURANTIACA. R. Bonaly and J. R. Malenge (Lab. de Chimie Biologique, Faculte de Phar. de Nancy et Centre de Cinetique Physique et Chimique de C.N.R.S., Nancy, France). *Biochim. Biophys. Acta* 164, 306–316 (1968). The carotenoid compositions of two yeasts *Rhodotorula mucilaginosa* and *R. aurantiaca* have been examined; acyclic and cyclic pigments were found. Kinetic experiments with resting cell suspensions were performed to examine the pathway of biosynthesis of the cyclic carotenoids. The results allow one to exclude the alternative hypothesis of the independent synthesis of carotenoids and support the idea that phytoene is the precursor of the cyclic pigments. The transformation of the acyclic into cyclic compounds occurs by the following two routes: neurosporene → β-zeacartene → γ-carotene and neurosporene → lycopene → γ-carotene. γ-Carotene is converted both to β-carotene and to torulene and torularhodin. 3′,4′-Dehydro-18′-oxo-γ-carotene and 3′,4′-dehydro-18′-hydroxy-γ-carotene have been isolated and may be considered as the intermediate compounds in the conversion of torulene to torularhodin.

THE TURNOVER OF MYELIN IN THE ADULT RAT. M. E. Smith (Neurology Service, Veterans Admin. Hosp., Dept. of Med. (Neurology), Stanford Univ. Sch. of Med., Palo Alto, Calif.).

Biochim. Biophys. Acta 164, 285–293 (1968). The rate of loss of radioactivity of lipids of brain subcellular fractions was followed for 56 days after injection of adult rats with uniformly labeled glucose-^{14}C. Myelin did not reach its maximum specific activity until 5 days after injection, then showed a slow rate of loss of radioactivity. When the turnover of the lipid components of purified myelin was measured, inositol phosphatide and lecithin were found to have a more active metabolism than the other myelin lipids which include serine phosphatide, sphingomyelin, ethanolamine phosphatide, cerebroside, cerebroside sulfate, and cholesterol. These results, which agree with our previous findings, indicate that the metabolism of myelin on the outer layers is identical to that laid down early in the life of the animal. Proteolipid protein showed a half-life of about 35 days, thus was one of the more active myelin components. The pattern of metabolism, though on a longer time scale in myelin, is similar to that of the most active brain membrane fraction. These results are discussed in relation to the possible structure of myelin membranes.

BILE ACIDS AND LIPID METABOLISM. III. INFLUENCE OF BILE ACIDS ON PHOSPHOLIPIDS IN LIVER AND BILE OF THE ISOLATED PERFUSED DOG LIVER. L. Swell, C. C. Bell and C. Entenman (Veterans Admin. Hosp. and Dept. of Surgery, Med. College of Virginia, Richmond, Va. and Inst. for Lipid Res., Berkeley, Calif.). *Biochim. Biophys. Acta* 164, 278–284 (1968). The isolated perfused dog liver was used to study the effect of bile acids on the excretion of biliary phospholipids and the ^{32}P-labeling of the individual plasma, liver and biliary phospholipids. Although high amounts of phospholipid-^{32}P were found in the liver, none was excreted in the bile unless taurocholate was infused. Biliary phospholipid excretion continued to increase throughout the taurocholate infusion period. The liver was the source of the biliary phospholipids. However, during the taurocholate infusion the specific activity of the biliary phospholipids greatly exceeded the liver phospholipids which suggest that a specific phospholipid is secreted into the bile by the liver or that biliary phospholipids are derived from a specific pool in the liver. There was a marked heterogeneity in the labeling pattern of the plasma, liver and biliary phospholipids. The bile phosphatidyl-^{32}P choline fraction accounted for over 95% of the total phospholipid-^{32}P. Marked differences between the composition of the liver and plasma phospholipids were noted. The data support the view that the stimulatory effect of bile acids on biliary phospholipid excretion is related to the formation of a biliary micellar complex and that highly selective mechanisms are involved in the formation of plasma and biliary phospholipids.

FRACTIONATION OF UNALTERED PHOSPHATIDYL (1,2-^{14}C) ETHANOLAMINES ACCORDING TO THE DEGREE OF UNSATURATION OF THEIR PREDOMINANT FATTY ACIDS. S. M. Hopkins, G. Sheehan and R. L. Lyman (Dept. Nutritional Sciences, Univ. of Calif., Berkeley, Calif.). *Biochim. Biophys. Acta* **164**, 272–277 (1968). Female rats were injected intraperitoneally with 1.5 μC of ethanolamine-^{14}C per 100 g of body weight 1 hour before they were killed; and the phospholipids were separated by thin-layer chromatography, and the radio-activity of the fractions determined. Phosphatidyl ethanolamine, isolated by thin-layer chromatography, was fractionated according to the degree of unsaturation on $AgNO_3$-treated silica gel layers into fractions containing dienes, tetraenes and hexaenes (mostly 22:6 with smaller amounts of the 22:5 fatty acids). The phosphatidyl ethanolamine was eluted with choline chloride in methanol and extracted into light petroleum. The radio-activity and fatty acids were determined, and the yields of each were 75% or greater. The specific activities of the fractions varied greatly. The dienes had the highest specific activity with the tetraenes having the lowest activity. Hydrolysis of the phosphatidyl ethanolamine with phospholipase A, showed that in addition to palmitic and stearic acids, the α-position contained 68% of the oleic acid and about 9% of the arachidonic acid. The relative simplicity of the procedure makes it a useful method for studying the metabolism of the major species of phosphatidyl ethanolamine *in vivo*.

CHOLESTEROL UPTAKE BY $L_{5178}Y$ TISSUE CULTURE CELLS: STUDIES WITH DELIPIDIZED SERUM. G. H. Rothblat, M. K. Buchko and D. Kritchevsky (Wistar Inst. of Anatomy and Biol., Philadelphia, Pa. 19104). *Biochim. Biophys. Acta* **164**, 327–338 (1968). The uptake of free cholesterol by tissue culture cells ($L_{5178}Y$) has been studied in a system using delipidized rabbit serum protein as the carrier of free cholesterol. The addition of phospholipids together with the free sterol reduces the adsorption of free cholesterol during a 5 hour incubation period. Various purified phospholipids differed in effectiveness, sphingomyelin being most effective in reducing free cholesterol uptake, followed by lecithin > phosphatidyl ethanolamine > phosphatidyl serine. Lysolecithin, at low concentrations, stimulated uptake slightly; at higher concentrations there was inhibition. In additional experiments, the addition of cholesterol plus various free fatty acids or glycerides to the delipidized protein affected free cholesterol adsorption by the cells. The influence of these compounds was not as pronounced as that of phospholipid. The uptake of cholesterol by $L_{5178}Y$ cells was influenced by the concentration of free cholesterol, phospholipids and protein in the culture medium.

FURTHER STUDIES ON CORTICOSTEROIDOGENESIS. V. 11β-HYDROXYLATION OF DEOXYCORTICOSTERONE BY MITOCHONDRIA INCUBATED WITH MALATE, SUPERNATANT FRACTION AND SUPERNATANT FRACTION + PYRUVATE + CO_2. F. G. Peron and B. V. Caldwell (The Worcester Found. Expt. Biol., Shrewsbury, Mass.). *Biochim. Biophys. Acta* **164**, 396–411 (1968). Rat adrenal gland mitochondria (P_2) utilize malate for generating intramitochondrial NADPH required for 11β-hydroxylation of deoxycorticosterone. High speed supernatant fractions (Sup.) of rat adrenal gland homogenates which are devoid of mitochondria and microsomes can replace malate when incubated with the rat adrenal gland mitochondria for the aforementioned steroid hydroxylation. The substance(s) in the Sup. which are responsible for intramitochondrial NADPH generation were found to be heat-labile and dialyzable so that small as well as large molecular weight substances appear to be implicated in the phenomenon of pyridine nucleotide reduction. P_2 alone cannot utilize pyruvate + CO_2 to form malate although the presence of the malate dehydrogenase (decarboxylating:NADP) (EC 1.1.1.40) in the P_2 is strongly indicated. Thus, malate in P_2 as a result of its oxidation via the intramitochondrial NADP-linked malate dehydrogenase (decarboxylating:NADP), generates NADPH and pyruvate + CO_2. Sup. on the other hand, in the presence of NADPH + CO_2 converts pyruvate into malate. This malate in the Sup. + P_2 combinations can be utilized by the P_2 for intramitochondrial NADPH production necessary for corticosterone production from deoxycorticosterone.

TURNOVER, BY SYNTHESIS AND BY TRANSFER, OF FREE AND ESTERIFIED CHOLESTEROL. MAGNITUDE OF THESE COMPARTMENTS IN ADULT RATS AND THEIR DISTRIBUTION IN TISSUES. C. Chevallier, R. D'Hollander and F. Simonnet (Dept. of Biologie, Centre D'Etudes Nucleares, Saclay, France). *Biochim. Biophys. Acta* **164**, 339–356 (1968). The isotopic equilibrium method has been applied to the study of the biodynamics of cholesterol in the adult rat. The magnitudes of the "labile cholesterol transfer space" and the "cholesterol compartment turned over by synthesis" were studied in groups fed diets containing cholesterol-4-^{14}C, acetate-1-^{14}C, mevalonate-2-^{14}C palmitate-1-^{14}C for periods of 2 and 4 months. In some studies, the specific activities of tissue cholesterol were measured on fractions purified by preparative gas-liquid chromatography, as well as the more classical methods. About 25% of the total sterols, including cholesterol, were esterified in the rat. Two-thirds of the esterified fraction were sterols other than cholesterol, whereas 96% of the free sterols were cholesterol. Of the total sterols, 14–19% appeared to be precursor sterols, and more than three-quarters of these were esterified. The "labile transfer space for free cholesterol" accounts for 70–75% of the total free cholesterol in the adult animal. The "free cholesterol compartment being turned over by synthesis" represents no more than 2–3% of the total free cholesterol.

STUDIES ON THE REACTION OF CYTOCHROME C WITH CORTICOSTEROIDS. C. Monder (Res. Inst. for Skeletomuscular Diseases, Hosp. for Joint Diseases and Med. Center, New York, N.Y. 10035). *Biochim. Biophys. Acta* **164**, 369–80 (1968). The present experiments were designed to demonstrate a model system which illustrates the possibility that intracellular oxidation-reduction reactions may occur between corticosteroids and cellular components. Cortisol reduced cytochrome c in buffered aqueous solution. The rate depends on buffer used and showed an optimum at pH 8.4 in Tris and pH 9.3 in carbonate. Reduction in glycine buffer showed no pH optimum. Reaction rate was first order with respect to cortisol concentration, but was more complex with respect to cytochrome concentration. High concentrations of cytochrome c inhibited the reduction. The ketol side chain on the steroid was essential for reduction. One product resulting from the oxidation of cortisol by cytochrome c was 21-dehydrocortisol. 17α-Hydroxycorticosteroids reduced cytochrome c faster than did 17-deoxy derivatives. Cytochrome c was also reduced by 21-dehydrocortisol, with a first order rate with respect to steriod. In bicarbonate buffer no pH optimum was obtained, and reduction rate was about 0.6 that of cortisol, though total reduction was greater than with cortisol. Several lines of evidence showed that cortisol reduced cytochrome c directly without prior oxidation to 21-dehydrocortisol. Cortisol or a derivative was bound to cytochrome c and the binding increased with incubation time.

OXIDATION OF 5β-CHOLESTANE-3α,7α,12α-TRIOL BY RAT-LIVER MITOCHONDRIA. K. Okuda and N. Hoshita (Dept. Biochem., Hiroshima Univ. School of Dentistry, Hiroshima, Japan). *Biochim. Biophys. Acta* **164**, 381–88 (1968). The oxidation of 5β-cholestane-3α,7α,12α-triol (an intermediate in the conversion of cholesterol to cholic acid) was studied. Experimental conditions for the study of the oxidation of 5β-cholestane-3α,7α,12α-triol were established. When 5β-cholestane-3α,7α,12α-triol was incubated with rat-liver particulate fractions, the formation of 5β-cholestane-3α,7α,12α,26-tetrol and 5β-cholestane-3α,7α,12α-triol-26-oic acid was observed only in the incubation mixture containing mitochondria. The addition of boiled rat-liver extract to the incubation mixture greatly stimulated the production of 5β-cholestane-3α,7α,12α,26-tetrol, while 100,000 × g supernatant did not show such a stimulative effect. Among the various animals studied, rats and mice showed the greatest activity for the oxidation of 5β-cholestane-3α,7α,12α-triol. Among the cofactors tested, only NADPH (an excess amount compared to that contained in boiled extract) could substitute for the boiled rat-liver extract in the oxidation of 5β-cholestane-3α,7α,12α-triol.

CELLULAR FATTY ACID COMPOSITION AND IDENTIFICATION OF RUMEN BACTERIA. R. W. Ifkovits and H. S. Ragleb (Dept. Biochem. Purdue Univ., Lafayette, Ind. 47907). *J. Appl. Microbiol.* **16**, 1406–1413 (1968). The fatty acid compositions of 21 pure cultures of rumen bacteria (12 genera, 14 species) were compared. Consistent and reproducible fatty acid profiles were obtained, however, overlapping similarities did not allow differentiation between families and genera.

AFLATOXIN IN FOOD AND FORAGE PRODUCTS. T. Jakubczyk. *Przemysl Spozywczy* 22(5), 205–210 (1968). Mycotoxins in general, are discussed. The characteristic chemical structure of aflatoxins is given and their formation by molds of the group *Aspergillus flavus-oryzae*, outlined. The occurrence of the aflatoxins, their toxicity and mode of action of the organisms and methods of determination and of elimination or destruction in food and forage products are reviewed. (Rev. Franc. Corps Gras)

INACTIVATION OF LIPASES DURING EXTRACTION OF RAPE SEEDS AND THE SEEDS OF CRAMBE ABYSSINICA. L. Jiraskova *et al.* *Sb. Vys. Sk. Chem.-Technol. Praze, Potraviny* E20, 79–84

(1968). The lipases of Crambe are more active and more stable than those of rape seeds. However, the increase in acidity during storage of the grains or of the press cake is not caused solely by lipases, but also by other esterases. In the case of industrial extraction, the residual lipolytic activity depends inversely on the temperature of heating. In dry press cakes, the lipases are relatively thermostable: the lipases of rape are more stable than pancreatic lipase. (Rev. Franc. Corps Gras)

RAPESEED. X. COUNTERCURRENT EXTRACTION OF THE PROTEIN FROM RAPESEED PRESS CAKE. J. Pokorny *et al. Sb. Vys. Sk. Chem.-Technol. Praze, Potraviny* E22, 113–119 (1968). The countercurrent (continuous) extraction of rapeseed press cake in a column with solutions of sodium hydroxide or sodium carbonate resulted in an extract containing a greater percentage of dry matter, total nitrogen, and precipitatable protein than obtained by batch extraction. These improvements did not counterbalance the disadvantages of the more complicated continuous procedure. (Rev. Franc. Corps Gras)

STUDIES ON THE REPLACEMENT OF PEANUT CAKE AND SOYBEAN CAKE BY RAPE CAKE IN THE FATTENING RATIONS FOR SWINE. S. Berthold. *Tluszcze Jadalne* 12(3), 97–103 (1968). Nondebittered rape cake should not be used in the diet of swine because it lowers growth rate and increases the consumption of feed. In contrast, completely or partially debittered rape cake is a valuable feed for fattening and can be used successfully in place of soya and peanut cakes in the "Bekon" diet at a level of about 15%. (Rev. Franc. Corps Gras)

PURIFICATION OF RAT PANCREATIC LIPASE. L. I. Gidez (Inst. de Chimie Biol., Faculte des Sciences, Marseille, France, and Depts. of Biochem. and Med., Albert Einstein College of Med., Yeshiva Univ., Bronx, New York 10461). *J. Lipid Res.* 9, 794–8 (1968). A procedure for the isolation of lipase (glycerol-ester hydrolase, EC 3.1.1.3) from rat pancreas is described. The purification scheme includes homogenization of the pancreas, centrifugation at 3,000 rpm, centrifugation at 40,000 rpm, DEAE-cellulose chromatography, precipitation of amylase as the amylase-glycogen complex, gel filtration of the amylase-free proteins on Sephadex G-100, and chromatography on carboxymethyl-Sephadex C-50. The enzyme showed only one band on polyacrylamide gel electrophoresis and had a specific activity of 5330 ± 80 units/mg of protein.

METABOLISM OF GLYCEROL ETHER-CONTAINING LIPIDS IN DOGFISH (SQUALUS ACANTHIAS). D. C. Malins (U.S. Bureau of Commercial Fisheries, Food Science Pioneer Res. Lab., Seattle, Washington 98102). *J. Lipid Res.* 9, 687–92 (1968). Dogfish *(Squalus acanthias)* received intrahepatic injections of either palmitic acid-1-^{14}C or chimyl alcohol-1-^{14}C. The lipids of the liver were then analyzed for incorporated radioactivity. The experiments with labeled palmitic acid demonstrated that fatty acids are reductively incorporated into the alkyl and alkenyl ether chains of glycerolipids. Significantly lower specific activities were found for the diacyl alk-1′-enyl ethers and diacyl glycerol ethers than for other glycerol ether-containing lipids. These compounds may therefore represent terminal points in ether-lipid metabolism. The studies with labeled chimyl alcohol indicate that dogfish liver contains enzymes that have a high capacity for oxidatively cleaving alkyl ether linkages. Furthermore, it is probable that alkyl ethers are converted directly to alkenyl ethers, possibly via a biodehydrogenation reaction.

DETERMINATION OF THE MOLECULAR WEIGHT OF APOPROTEIN SUBUNITS FROM LOW DENSITY LIPOPROTEIN BY GEL FILTRATION. C. E. Day and R. S. Levy (Dept. of Biochem., Univ. of Louisville School of Med., Louisville, Kentucky 40202). *J. Lipid Res.* 9, 789–93 (1968). Another method has been developed for obtaining a soluble apoprotein from the low density lipoprotein (LDL) of human plasma in the density class $1.019 < d < 1.063$. The approximate molecular weight of the apoprotein subunit from this lipoprotein density class was determined by gel filtration on Sephadex G-200 to be about 80,000. Both on gel filtration and analytical ultracentrifugation the soluble apoprotein showed one peak, but on cellulose acetate electrophoresis it showed two bands, which suggests two differently charged components. Because of the nature of the determination, the value of 80,000 probably represents an upper limit to the molecular weight of the LDL subunits.

EFFECT OF DIETHYLSTILBESTROL ON SKIN STEROLS OF THE MALE RAT. L. Horlick (Dept. of Med., Univ. of Saskatchewan, Univ. Hosp., Saskatoon, Saskatchewan, Canada). *J. Lipid Res.* 9, 773–81 (1968). Diethylstilbestrol (DES) was injected in doses ranging from 600 μg to 0.4 μg/kg body weight into mature male rats over a 3 week period. Profound effects on skin morphology and on sterol content of skin were noted. The sebaceous glands atrophied and the epidermis lost granularity. The concentrations of all skin sterols, with the exception of cholesterol, were reduced. At a dose level of DES of 4 μg/kg there was still a perceptible reduction in the concentration of Δ^7-cholestenol. Incubation of skin fragments with acetate-2-^{14}C for 2 hours demonstrated a reduced uptake of ^{14}C into the nonsaponifiable fraction of skin lipids at all dose levels studied. Preliminary thin-layer chromatography of the nonsaponifiable fraction revealed that the uptake of ^{14}C into cholesterol was only slightly decreased; uptake into cholesterol precursors was decreased somewhat more. The epidermis and dermis were separated by incubation of skin with elastase and hyaluronidase. The epidermis contained at least three times as much sterol per mg dry weight as did the dermis. Unesterified cholesterol was the major sterol present in both layers; the other sterols were present mainly as esters. DES injection resulted in no change in the free sterol content but markedly reduced the ester content of the epidermis and dermis.

SERUM LIPIDS AND DEHYDROEPIANDROSTERONE EXCRETION IN NORMAL SUBJECTS. J. Sonka, M. Fassati, P. Fassati, I. Gregorova and K. Picek (Lab. of Endocrim. and Met. and Dept. of Epidemiology, Faculty of Med., Charles Univ., Prague 2, U nemocnice 1, Czechoslovakia). *J. Lipid Res.* 9, 769–72 (1968). Serum levels of cholesterol, phospholipids, β-lipoproteins and free fatty acids were correlated with urinary dehydroepiandrosterone (DHEA) excretion in healthy blood donors. An indirect dependence was found for cholesterol and phospholipids that was more important in persons with a low DHEA excretion. The correlation seems to be more a function of the dependence of both DHEA excretion and serum lipid levels on age than a direct relationship between these factors.

ISOLATION AND IDENTIFICATION OF CHOLESTERYL ALKYL ETHERS FROM BOVINE CARDIAC MUSCLE. H. Funasaki and J. R. Gilbertson (Dept. of Biochem. and Nutr., Graduate School of Public Health, Univ. of Pittsburgh, Pittsburgh, Pa. 15213). *J. Lipid Res.* 9, 766–68 (1968). Cholesteryl alkyl ethers have been isolated from bovine cardiac muscle and characterized by thin-layer and gas-liquid chromatography. The fraction contained at least three homologues. Cholesteryl hexadecyl ether, which accounted for over 90% of the total components observed on gas chromatography, was identified by mass spectrometry.

GAS-LIQUID CHROMATOGRAPHY ANALYSES OF 1,2-ETHANEDIOL MONOETHERS AND MONOESTERS. R. Wood and W. J. Baumann (Med. Div., Oak Ridge Associated Univs., Oak Ridge, Tenn. 37830, and The Univ. of Minn., The Hormel Inst., Austin, Minn. 55912). *J. Lipid Res.* 9, 733–38 (1968). Synthetic mixtures of saturated and unsaturated monoethers and monoesters of 1,2-ethanediol, ranging in chain length from 12 to 20, were analyzed as acetates, trifluoroacetates (TFA), and trimethylsilyl (TMS) ethers by gas chromatography on polar and nonpolar liquid phases. Acetates, TFA derivatives, and TMS derivatives of the glycol ethers were eluted ahead of the corresponding glycol ester derivatives on both liquid phases. The elution order of derivatives of the same compound was found to be TMS derivative before TFA derivative before acetate on the polar liquid phase, and TFA derivative before TMS derivative before acetate on the nonpolar liquid phase. Elution orders relative to methyl stearate were also determined. With one exception, all of the derivatives, and both liquid phases, were found suitable for the quantitative analysis of diol monoethers and monoesters.

SELENIC LIPIDS IN GAUCHER'S DISEASE. N. G. Kennaway and L. I. Woolf (External Staff of the Med. Res. Council, Dept. of the Regius Professor of Med., Univ. of Oxford, Oxford, England). *J. Lipid Res.* 9, 755–65 (1968). Column chromatography (on cellulose, silicic acid, and Florisil) and thin-layer chromatography were employed for the separation and purification of lipid fractions from normal and Gaucher spleens. A new hydrolysis procedure, followed by paper chromatography, was used for identification of sugar moieties. A nonhydrolytic combined colorimetric procedure, with anthrone and orcinol, was used for the estimation of glucose and galactose separately in glycolipids. The limitations of this method were examined. Spleens from two control subjects and three patients with Gaucher's disease have been examined in detail. In all Gaucher spleens, the predominant feature was the massive accumulation of glucocerebroside: neutral ceramide oligohexoside levels were probably within the normal range, as were other neutral lipids and phospholipids. In one case examined for gangliosides, these were increased twentyfold. One Gaucher spleen, in which others had reported that the stored "cerebroside" contained predominantly lactose as the saccharide moiety, has been examined in detail and it has been established that the stored

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material was, in fact, glucocerebroside, ceramide lactoside levels not being significantly elevated. In a further nine cases glucose was the major sugar detected in the splenic lipids.

EFFECTS OF EPINEPHRINE ON GLUCOSE TRANSPORT AND METABOLISM IN ADIPOSE TISSUE OF NORMAL AND HYPOTHYROID RATS. G. A. Bray and H. M. Goodman (New England Med. Center Hospitals and Dept. of Med., Tufts Univ. Schol of Med., and the Dept. of Physiol. Harvard Med. School, Boston, Mass. 02111). *J. Lipid Res.* 9, 714–19 (1968). Epinephrine increases the oxidation of glucose in adipose tissue even when its lipolytic effects are markedly reduced or abolished by propranolol, nicotinic acid, ouabain or thyroidectomy. In order to locate the site(s) at which epinephrine stimulates glucose utilization, we studied the effects of epinephrine on the oxidation of various metabolites of glucose. Epinephrine neither increased the production of $^{14}CO_2$ from 1- or 3-^{14}C- pyruvate nor affected pyruvate conversions to glyceride-glycerol. To assess the possibility that epinephrine might accelerate the entry of glucose into adipocytes, the accumulation of the nonmetabolized sugar L-arabinose in the intracellular water of adipose tissue were studied. Epinephrine increased arabinose penetration into adipocytes to a degree comparable with that caused by 0.1 mU/ml of insulin. Virtually identical results were obtained in tissues from thyroidectomized rats in which the lipolytic effects of epinephrine were significantly reduced. It is concluded that epinephrine increases glucose oxidation by promoting its entry into adipose tissue and that the effect is independent of lipolysis.

RAPID PREPARATION OF TRITIUM-LABELED BILE ACIDS BY ENOLIC EXCHANGE ON BASIC ALUMINA CONTAINING TRITIATED WATER. A. F. Hofmann, Patricia A. Szczepanik and P. D. Klein (Gastroenterology Unit, Mayo Clinic and Mayo Found., Rochester, Minn. 55901 and Div. of Biol. and Med. Res., Argonne National Lab., Argonne, Ill. 60439). *J. Lipid Res.* 9, 707–13 (1968). When a 3-keto bile acid methyl ester was chromatographed on basic alumina inactivated with tritiated water, the enolic hydrogen atoms at C-2 and C-4 exchanged with tritium atoms. The ^{3}H-labeled keto ester was reduced with borohydride, and the resultant mixture of 3α- and 3α-hydroxy epimers was resolved by preparative thin-layer chromatography to yield a pure 2,4-^{3}H-labeled bile acid ester. Lithocholic, chenodeoxycholic, deoxycholic and cholic acids having a specific activity of 1–10 μc/μmole were prepared from their 3-keto derivatives. The tritium label remained intact during alkaline saponification *in vitro* and enterohepatic cycling *in vivo* in human subjects.

METABOLISM OF SEX HORMONES IN THE AORTIC WALL. A. V. Chobanian, P. I. Brecher, R. D. Lille and H. H. Wotiz (Boston City Hospital, and the Depts. of Med. and Biochem., Boston Univ. School of Med., Boston, Mass. 02118). *J. Lipid Res.* 9, 701–06 (1968). The metabolism of labeled sex hormones was examined in human, canine and rat aortas. Isolated arterial tissue converted estrone to estradiol, estradiol to estrone, and estrone sulfate to estrone and estradiol. The arterial wall also appeared to metabolize testosterone to androstenedione and an unidentified, relatively nonpolar derivative. Both estrogens and testosterone appeared to enter the arterial wall rapidly. No competition in arterial uptake between the two hormones was apparent. No specific arterial binding of estradiol could be demonstrated. The concentration of estradiol-^{3}H in the canine aorta exceeded that in the plasma 1–6 hours after estradiol-^{3}H administration. The uptake and disappearance of estradiol-^{3}H in the aorta generally resembled the patterns observed in body tissues other than the adrenal gland and uterus. The uptake of estradiol-^{3}H was greatest in the adrenal gland while its retention was maximum in the uterus.

ELECTROPHORETIC SEPARATION OF PLASMA LIPOPROTEINS IN AGAROSE GEL. R. P. Noble (The Sharon Res. Inst., The Sharon Hosp., Sharon, Connecticut 06069). *J. Lipid Res.* 9, 693–700 (1968). A method has been developed for the separation of serum or plasma lipoproteins by electrophoresis in an agarose-agar gel mixture. The gel is applied to the surface of a thin polyester photographic film strip. With minor alterations in technique either single samples on individual strips or many samples on one large sheet may be processed. After fixation and dehydration the transparent film is stained with Sudan Black B and washed with water. The finished electrophoretogram can be obtained in 5 hours and consists of widely separated bands of lipoprotein fractions on a colorless transparent background, ideally suited for scanning with a densitometer. Plasma samples from different subjects show pre-β lipoproteins of different mobilities. An effect of gel concentration on the extent of lipoprotein migration is demonstrated. The clearcut separation of lipoproteins by this method will facilitate the classification of hyperlipoproteinemias and improve quantitative estimates of lipoprotein distribution.

MODIFICATION OF LOFLAND'S COLORIMETRIC SEMIAUTOMATED SERUM TRIGLYCERIDE DETERMINATION, ASSESSED BY AN ENZYMATIC GLYCEROL DETERMINATION. A. R. Timms, L. A. Kelly, Judith A. Spirito and R. G. Engstrom (Biochem. Sect., Res. Dept., Sandoz, Inc., Hanover, New Jersey 07936). *J. Lipid Res.* 9, 675–80 (1968). Erroneously high values for serum triglyceride levels obtained with the semiautomated method of Lofland were found to be due to contamination of the isopropanol extracts with glucose or other carbohydrate. Treatment of the isopropanol extracts with a mixture of copper sulfate and calcium hydroxide removed the contaminating glucose. Analysis of the glucose-free extracts by either the semiautomated or manual colorimetric method gave values in good agreement with each other and with those obtained by a new specific enzymatic method. The simple modification described in this paper obviates the necessity for the more expensive automated fluorometric apparatus.

LIPID COMPOSITION AND SECRETORY ACTIVITY OF BOVINE MAMMARY CELLS IN VITRO. J. E. Kinsella and R. D. McCarthy (Lipids Lab., Pa. State Univ., Univ. Park, Pa. 16802). *Biochim. Biophys. Acta* 164, 530–39 (1968). The lipid content and composition of collagenase dispersed mammary cells was determined. Individual cells contained approximately 300 pg lipid per cell which decreased as the cells aged. The neutral lipid fraction contained triglycerides (56%), sterols (22%), and free fatty acids (13%) as major components. Phosphatidyl choline was the major polar lipid fraction (52%). A trace quantity of phosphatidic acid was tentatively detected in the cellular lipids. Active secretion of fat droplets was observed by phase microscopy. The cells secreted from 0.6 to 7.5 pg lipid per cell per hour during the initial 3 days in culture after which secretion diminished. The secreted lipid was composed predominantly of triglyceride whose fatty acid composition was similar to that of bovine milk fat.

STUDY OF ENDOGENOUS FATTY ACIDS OF THE LYMPHATIC CHYLOMICRONS TRIGLYCERIDES IN THE RAT AFTER ADMINISTRATION OF VARIOUS MIXED LABELED TRIGLYCERIDES. P. Boucrot and J. Clément (Lab. de Physiologie Animale et de la Nutr., Faculte des Sciences, Dyon 21, France). *Biochim. Biophys. Acta* 164, 558–65 (1968). Rats with cannulated thoracic ducts received one or several meals containing various quantities of mixed triglycerides with 2 or 4 different labeled fatty acids. The composition of endogenous and exogenous fatty acids of the lymphatic chylomicron triglycerides was determined by gas-liquid chromatography and by radioactivity determinations. Whatever the nature and the amounts of the triglycerides fed, the quantities of endogenous fatty acids and the proportions of each of them vary little for lymph collection time of 14–16 hours. The results offer an explanation for the variations, observed by different authors, in the extent of the dilution by endogenous acids in the lymph; their meaning as to the origin of these endogenous lipids was also discussed.

THE INCORPORATION OF GLYCEROL-3-^{14}C INTO LIPIDS BY DISPERSED BOVINE MAMMARY CELLS. J. E. Kinsella (Lipids Lab., Pa. State Univ. Park, Pa. 16802). *Biochim. Biophys. Acta* 164, 540–49 (1968). Freshly dispersed cells obtained from lactating bovine mammary gland actively absorbed and metabolized glycerol-3-^{14}C present in the culture medium. About 19% of the available free glycerol was incorporated into lipids within 18 hours. Activation of glycerol and lipid biosynthesis apparently occurred intracellularly. Labeled lipids, mostly triglycerides, accumulated in the media during the experimental period. Most of the radioactivity of the cellular lipids was associated with the triglycerides, 1,2-diglycerides and phosphatidylcholine. Significantly, both ceramidemonohexose and ceramidedihexose contained radioactivity. The specific activity-time curves tentatively indicated that the triglycerides were derived from the 1,2-diglycerides. Phosphatidylinositol and cardiolipins initially had very high specific activities which declined rapidly. The specific activities of phosphatidylcholine and phosphatidylethanolamine suggested that they may have been derived from the 1,2-diglycerides. The monoglycerides showed extremely high specific activity at all stages of the experiment whereas that of the phosphatidic acid was negligible. Evidence was adduced for the functioning of the α-glycerophosphate pathway in bovine mammary tissue. It was concluded that bovine mammary cells can utilize exogenous glycerol and that the secretory cells can synthesize the normal glycerolipids and the cerebrosides found in milk.

METABOLISM OF SPHINGOMYELIN IN THE INTESTINAL TRACT OF THE RAT. A. Nilsson (Div. of Physiolog. Chem., Chem. Centre,

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AOCS Honored Student Program

The American Oil Chemists' Society is quite pleased with the success of the AOCS Honor Student Program which has been established for seven years. The objective of this program is to stimulate the teaching and research in the chemistry and technology of fats and oils in colleges and universities. Fifteen graduate students are selected each year to receive the AOCS Honor Student Award. They are invited to attend the National Meetings of the AOCS with all their expenses paid. Each of them also receives a handsome certificate of the award. This program was originally sponsored financially by AOCS. However, during the past two years, the expenses of this award have been taken over by contributions from companies in the fats and oils industry. Those who have contributed to this program to encourage future oil chemists are: Baker Castor Oil Company; Cargill, Inc.; Central Soya Company; Corn Products Company; Corn Products Company, Best Foods Division; Emery Industries, Inc.; Fatty Acid Producers' Council; Hunt Foods & Industries, Inc.; National Dairy Products Corporation; Swift & Company.

Professor Stephen S. Chang of the Food Science Department of Rutgers, The State University, who has served as Chairman of this program since its initiation has announced the following fifteen students as recipients of the Honor Student Award of 1969. Z. L. Bandi, Biochemistry Department, University of Georgia; K. E. Beery, Food Science & Industry, Penn State University; J. D. Castell, Food Science & Tech., Oregon State University; W. J. Esselman, Biochemistry Department, Penn State University; R. M. Gould, Department Physiol Chem., John Hopkins University; E. A. Grellert, Biophysics & Nucl. Medicine, UCLA Medical School; N. Heidelbaugh, Food Engineering Department, Massachusetts Institute of Technology; C. Hirschberg, Biochemistry Department, University of Illinois; W. A. May, Food Science Department, Rutgers University; A. L. Ochs, Pharmacology Department, Washington University; C. J. Rudolph, Biochemistry Department, Oklahoma State University; T. Santosusso, Chemistry Department, Temple University; C. Siegfried, Biochemistry Department, St. Louis University; R. Smallidge, Biochemistry Department, Purdue University; A. Waggoner, Chemistry Department, University of Oregon. They will attend either the AOCS National Meeting in San Francisco in the spring or the fall meeting in Minneapolis as guests of the Society.

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Univ. of Lund, Lund, Sweden). *Biochim. Biophys. Acta* 164, 575–84 (1968). Labeled sphingomyelins were fed to rats having a lymph fistula and to intact rats whose small intestinal content and intestinal wall or faeces were analyzed. The same types of experiments were performed with labeled palmitoylsphingosin, sphingosin and dihydrosphingosin as substrates. Most of the fed sphingomyelin was metabolized in the small intestinal tract. From 36–60% of the fatty acid was found in the lymph triglycerides and lecithin and 10–17% of the choline moiety appeared in the lymph lecithin. No evidence for absorption and transport by the lymph of intact dietary sphingomyelin, ceramide or sphingosyl-phosphoryl-choline was obtained. Dihydrosphingosin was well absorbed and metabolized in the mucosal cells to C_{16} fatty acid which was incorporated mainly into chylomicron triglycerides. Indirect evidence was obtained that much of the sphingosin portion of dietary sphingomyelin is absorbed and metabolized in a similar way. A part of the sphingosin bases was incorporated into ceramide and sphingomyelin in the mucosal cells. The hydrolysis of sphingomyelin is initiated in the small intestinal lumen.

INCORPORATION OF MOLECULAR OXYGEN INTO TESTOSTERONE ACETATE DIRECTLY DERIVED FROM PROGESTERONE BY CLADOSPORIUM RESINAE. H. Nakano, H. Sato and B. Tamaoki (Nat. Inst. of Radiological Sciences, Anagawa-4-chome, Chiba-shi 280, Japan). *Biochim. Biophys. Acta* 164, 585–95 (1968). Progesterone was adaptively converted under aerobic conditions by *Cladosporium resinae* to testosterone acetate and subsequently to testosterone and androstenedione. Under partially anaerobic conditions, production of testosterone acetate was limited, and the substrate was converted mainly to 20α-hydroxypregn-4-en-3-one. In order to clarify the role of molecular oxygen in the course of side-chain cleavage of progesterone by *C. resinae*, a double tracer method was applied using $^{18}O_2$ as a tracer of molecular oxygen and ^{14}C as a tracer of the steroidal substrate. After the microbiological transformation of progesterone under $^{18}O_2$ atmosphere, testosterone acetate, along with the testosterone derived from the testosterone actate by microbial esterase during the culture, and the testosterone which was chemically produced by saponification of the testosterone acetate, were obtained and subjected to mass-spectrometric analysis. Fragment analysis and chemical degradation of the products revealed that an atom of molecular oxygen was directly inserted into the C_{17}–C_{20} bond of one molecule of progesterone, probably by a reaction mechanism similar to that of the Baeyer-Villiger reaction.

LIPOGENESIS FROM GLUCOSE AND PYRUVATE IN FAT CELLS FROM GENETICALLY OBESE RATS. G. A. Bray (New England Med. Center Hosp. and Tufts Univ. School of Med., Boston, Mass. 02111). *J. Lipid Res.* 9, 681–86 (1968). Subcutaneous fat cells were isolated from genetically obese rats and from rats with obesity produced by hypothalamic lesions. Insulin did not augment the oxidation of fatty acids or their synthesis from glucose-1-^{14}C or glucose-1-^{3}H by fat cells from either group. Radioactivity from pyruvate-3-^{14}C was incorporated into fatty acids to the same degree by fat cells from these two groups. The presence of 5 mM glucose in the incubation medium containing fatty cells and pyruvate-3-^{14}C or aspartate-3-^{14}C stimulated the synthesis of fatty acids to a greater extent in cells of genetically obese rats. Fasting, in contrast, reduced the incorporation of radioactivity from pyruvate and glucose into fatty acids by fat cells from the genetically obese animals. In all experiments the fat cells from genetically obese rats converted more radioactivity into glyceride-glycerol relative to CO_2 than did fat cells from hypothalamic obese fats.

COMPOSITION AND SYNTHESIS OF FATTY ACIDS IN ATHEROSCLEROTIC AORTAS OF THE PIGEON. R. W. St. Clair, H. B. Lofland, Jr. and T. B. Clarkson (Depts. of Path. and Lab. Animal Med., The Bowman Gray School of Med. of Wake Forest Univ., Winston-Salem, N. Carolina 27103). *J. Lipid Res.* 9, 739–47 (1968). The composition, synthesis and esterification of fatty acids were studied in aortas of White Carneau and Show Racer pigeons after perfusion of the aortas with a medium containing acetate-1-^{14}C. For both breeds of pigeons the principal change in aortic fatty acids, in response to an atherogenic diet, was a marked increase in the percentage of oleic acid in the cholesteryl ester fraction. In atherosclerotic aortas incorporation of acetate-1-^{14}C into the phospholipid and glyceride fractions increased 2-fold, while a much greater increase (up to 10-fold) was seen in incorporation into cholesteryl esters. In those birds receiving the atherogenic diet, palmitic acid accounted for approximately 50% of the fatty acid radioactivity, compared with approximately 25% from control aortas. Calculation of fatty acid synthesis showed the major newly synthesized fatty acid to be stearic acid in the phospholipid fraction; stearic, palmitic, and oleic acids in the glycerides; and oleic acid in the cholesteryl esters. The pattern of fatty acid synthesis was closely similar to the actual fatty acid composition of the aorta. In atherosclerotic aortas an increased synthesis of all fatty acids was seen, but the greatest increase was seen in the synthesis of oleic acid and its esterification to cholesterol.

STATISTICAL ANALYSIS OF THE MAIN EFFECTS AND INTERACTIONS OF pH, TAUROCHOLATE AND CALCIUM ON PANCREATIC LIPASE ACTIVITY. C. N. Ansted and I. A. Hansen (Dept. of Biochem., Univ. of Western Australia, Nedlands). *Chem. Phys. Lipids* 2, 343–60 (1968). A factorial experiment was used to determine the nature of the effects of pH, taurocholate and Ca^{2+} on the rate of hydrolysis of long-chain triglycerides by pig pancreatic lipase. A stable emulsion of triolein was used as substrate. Statistical analysis showed that there were significant interactions between the three factors. Under the particular conditions of substrate and emulsion used, the following effects were found: In the presence of taurocholate and calcium ions the rate of hydrolysis increased in a non-linear fashion from pH 7.5 to 9.5. The overall effect of taurocholate in the presence of calcium ions showed an optimum at about 6.9 mM (0.37%, w/v). Ca^{2+} showed a similar optimum at about 40 mM. Although there was interaction between pH and taurocholate concentration, the plots of reaction rate vs. pH had the same general shape and position on the pH axis for all values of taurocholate. There was pronounced interaction between pH and Ca^{2+}; optimum Ca^{2+} was highly dependent on pH, and the reaction rate at higher pH values was highly dependent on Ca^{2+}. The interaction between Ca^{2+} and taurocholate involved an exponential function of both.

ELECTRON MICROSCOPY OF HUMAN SERUM LIPOPROTEINS USING NEGATIVE STAINING. G. M. Forte, A. V. Nichols and R. M. Glaeser (Donner Lab., Lawrence Radiation Lab., Univ. of California, Berkeley). *Chem. Phys. Lipids* 2, 396–408 (1968). Human serum lipoproteins were separated by ultracentrifugation into 3 fractions: high density lipoproteins, low density lipoproteins, and very low density lipoproteins including chylomicrons. The structures of the various fractions were studied with the electron microscope after negative staining with sodium phosphotungstate. The high density lipoproteins appear to be structures 86 A in average diameter possessing an electron-dense central region. The lipoproteins are composed of subunits which in some cases appear to be elongated with an estimated short axis of 36 A and long axis of 64 A. The high density lipoproteins may further be fractionated into their subclasses, HDL_2 and HDL_3. The HDL_3 molecules are considerably smaller in average diameter (65 A) than the HDL_2 (95 A). The low density lipoproteins appear spherical with a mean diameter of 216 A and they display a high degree of deformability, but they have no visible subunit structures. Bridgelike connections between adjacent lipoproteins also appear to be present. The very low density lipoproteins plus chylomicrons are extremely heterogeneous in size and shape and range from 230–5000 A in diameter. The smaller molecules seem to be spherical while the extremely large ones are flattened and irregular in shape.

LIPID COMPOSITION AND TURNOVER OF ROUGH AND SMOOTH MICROSOMAL MEMBRANES IN RAT LIVER. H. Glaumann and G. Dallner (Dept. of Pathol., Sabbatsberg Hosp., Karolinska Inst., and the Inst. of Biochem., Univ. of Stockholm, Stockholm, Sweden). *J. Lipid Res.* 9, 720–29 (1968). Subfractions of rat liver microsomes (rough, smooth I, and smooth II), isolated in a cation-containing sucrose gradient system, were analyzed. After removal of absorbed and luminal protein these subfractions had the same phospholipid/protein ratio, about 0.40. Both the classes and the relative amounts of phospholipids were similar in the three subfractions, but the relative amounts of neutral lipids (predominantly free cholesterol and triglycerides) were higher in smooth I and especially in smooth II than in rough microsomes. Various pieces of evidence indicate that the neutral lipids are tightly bound to the membranes. Glycerol-^{3}H was incorporated into the phospholipids of the rough and smooth II microscomes significantly faster than into those of the smooth II membranes; ^{32}P incorporation followed a similar but less pronounced pattern. Acetate-^{3}H was incorporated into the free cholesterol of smooth I microsomes only half as fast as into the other two subfractions. Injection of phenobarbital increased the cellular phospholipid and neutral lipid content in the rough and smooth I, but not in the smooth II microsomes. Consequently, the neutral lipid/phospholipid ratio of all three subfractions remained unchanged after phenobarbital treatment.

ON THE MECHANISM OF MALONYL-CoA-INDEPENDENT FATTY ACID SYNTHESIS. I. THE MECHANISM OF ELONGATION OF LONG-CHAIN

administration of a meal, containing 56 g of fat, serum alkaline phosphatase of intestinal origin was demonstrated electrophoretically in 14 subjects lacking it in the fasting state. Other subjects ingesting skim milk or glucose did not develop the intestinal component. A dilution technique indicating the sensitivity of the electrophoretic method is presented and shows that 2–3 mU/ml of phosphatase activity can be detected. Because of this sensitivity, intestinal phosphatase could be demonstrated in types of subjects in whom it had not been found by other authors. The importance of diet in studies of intestinal phosphatase is emphasized.

APPARENT CAROTENOID INCREASES IN THE DIGESTIVE TRACT OF BEEF CATTLE. R. Almendinger and F. C. Hinds (Ruminant Div., Dept. of Animal Sci., Univ. of Illinois, Urbana, Ill.). *J. Nutr.* 97, 13–18 (1969). Carotenoid determinations were made on haylage, hay and the feces of cattle fed these roughages. Excretion of β-carotene and other carotenoids was much greater than the amount contained in the haylage and was about equal to that contained in the hay. Apparent increases in β-carotene and total carotenoid were observed in haylage samples incubated with hydrochloric acid when concentrations approximated the conditions found in the abomasum, but this did not account for the total amount of pigment in the feces. This apparent increase was probably due to a releasing of residual carotenoids not extracted by the usual procedures.

COCARCINOGENIC PRINCIPLES FROM THE SEED OIL OF CROTON TIGLIUM AND FROM OTHER EUPHORBIACEAE. E. Hecker (Biochemisches Inst. am Deutschen Krebsforschungszentrum, Heidelberg, Germany). *Cancer Res.* 28, 2338–49 (1968). The spurge family or *Euphorbiacea* includes some 280 genera and 8000 species which occur in tropical and in temperate regions all over the world. These succulent or nonsucculent plants range from herbs and shrubs to tree and cactus types. Many of them contain a milky juice which is more or less toxic, especially for cold-blooded animals, and can produce a dermatitis similar to that from poison ivy. The fruits are usually three-celled capsules, each cell containing a single seed from which in some species toxic, vesicating and irritant seed oils may be obtained. The largest genera of the spurge family are those of Croton, with about 700 species, and of spurge of Euphorbia, with about 1600 species.

TUMOR-PROMOTING AGENTS FROM CROTON TIGLIUM L. AND THEIR MODE OF ACTION. B. L. Van Duuren and A. Sivak (Lab. of Organic Chem., and Carcinogenesis, Inst. of Env. Med., New York Univ. Med. Center, New York, N. Y.). *Cancer Res.* 28, 2349–56 (1968). The isolation of chemically pure tumor-promoting agents from croton oil in this laboratory facilitated the exploration of a number of areas concerning their mode of action and the role of tumor-promoting agents in chemical carcinogenesis. Our recent studies have centered on the mode of action of these phorbol esters on cellular and intracellular membranes. The use of tumor-promoting agents in two areas of carcinogenesis studies are also described in this report. They are: inhibition of tumorigenesis in two-stage carcinogenesis and the relationship between initiating agents, mutagens, and carcinogens.

INTERRELATIONSHIPS OF HYPERINSULINISM AND HYPERTRIGLYCERIDEMIA IN YOUNG PATIENTS WITH CORONARY HEART DISEASE. M. Tzagournis, R. Chiles, J. M. Ryan and T. G. Skillman (Div. of Endocrin. and Metabo., and Cardiol., Dept. of Med., The Ohio State Univ. College of Med., Columbus, Ohio). *Circulation* 38, 1156–63 (1968). Fasting serum lipid levels, glucose tolerance, and immunoreactive insulin concentrations of 50 young patients with coronary heart disease (CHD) and 30 control subjects were evaluated to study the interrelationships of these metabolic factors. Abnormalities in one or more of these factors could be shown in 90% of the patients and 20% of the control subjects. Thirty-four of the 50 patients had elevated cholesterol or triglyceride levels, or both, 30 had abnormally elevated or delayed insulin responses after glucose, and 17 had abnormal glucose tolerance. A significant correlation existed between serum triglyceride and insulin concentrations. When insulin levels were reduced by phenformin, triglyceride concentrations fell toward normal. These findings indicate that carbohydrate, insulin, and lipid abnormalities are rather prevalent in patients with CHD. Excessive insulin secretion secondary to mild glucose intolerance probably induces hepatic synthesis of triglycerides and hypertriglyceridemia. Dietary alterations or pharmacological agents may help to control some of the metabolic abnormalities associated with premature CHD.

EFFECTS OF SODIUM BICARBONATE, MAGNESIUM OXIDE, AND CALCIUM HYDROXIDE ON MILK FAT SECRETION. J. W. Thomas and R. S. Emery (Dept. of Dairy, Michigan State Univ., East Lansing). *J. Dairy Sci.* 52, 60–3 (1969). Addition of 0.70% $NaHCO_3$ plus 0.35% MgO, and twice or three times that level to grain mixtures fed to cows on a high grain-restricted roughage ration, increased milk fat percentage and daily secretion. Increasing mineral levels produced a linear increase in fat percentage. The mineral level fed had the greatest effect on daily secretion of milk fat, probably due to the lower grain consumption and milk production observed at the highest level. No differences in milk fat test, production, or grain consumption were noted when the higher mineral level (272 g $NaHCO_3$ plus 136 g MgO) was administered directly to the rumen via a fistula or mixed with the grain consumed. Evidently, decreased grain intake when these minerals are fed is not due to the taste of the grain mixture. This level of minerals fed to cows on normal rations did not change milk fat percentage, but grain consumption was decreased. Magnesium oxide in three different calcined states and $Ca(OH)_2$ were equally effective in maintaining fat percentage, but the hydroxide was less acceptable to cows.

BROILING, SEX AND INTERRELATIONSHIPS WITH CARCASS AND GROWTH CHARACTERISTICS AND THEIR EFFECT ON THE NEUTRAL AND PHOSPHOLIPID FATTY ACIDS OF THE BOVINE LONGISSIMUS DORSI. R. N. Terrell, G. G. Suess, R. G. Cassens and R. W. Bray (Dept. of Meat and Animals Sci., Univ. of Wisconsin, Madison, Wis. 53706). *J. Food Sci.* 33, 562–5 (1968). Broiling had a greater effect on phospholipid fraction fatty acids than on neutral fraction fatty acids. The percentages of C18:3 (neutral fraction) and C14 and C15 (phospholipid fraction) were significantly smaller (5% level) in the broiled steaks. However, the percentage of C8 (phospholipid fraction) was larger in the broiled than in the raw steaks (5% level). Sex differences, restricted to the neutral fraction acids, were greater than the effect of broiling. Steers had a larger percentage of C16 and C18 and a smaller percentage of C18:1 than heifers (5% level). Neutral fraction acids (C18, C18:1 and C18:2) were significantly correlated with lipid phosphorus, cholesterol, % fat trim (retail), estimated % carcass fat and estimated % carcass lean. Phospholipid fraction acids (C16, C18, C18:3 and C20:4) were associated with average daily gain and days of animal age (5% level). Low nonsignificant correlations were found among individual fatty acids from the neutral and phospholipid fractions of bovine longissimus dorsi muscle with tenderness and juiciness scores.

INFLUENCE OF MICROORGANISMS ON INTESTINAL ABSORPTION: OLEIC ACID ^{131}I AND TRIOLEIN ^{131}I ABSORPTION BY GERMFREE AND CONVENTIONALIZED RATS. B. Tennant, M. Reina-Guerra D. Harrold and M. Goldman (Dept. of Clin. Sci. and the Radiobiol. Lab., School of Vet. Med., Univ. of Calif., Davis, Calif.). *J. Nutr.* 97, 65–69 (1969). Several differences in lipid metabolism have been demonstrated between germfree and conventional rats. To evaluate the role which intestinal absorption might play in determining these differences, we compared the absorption of oleic acid ^{131}I and triolein ^{131}I in germfree and conventionalized rats. Gastric emptying of both compounds appeared to be delayed in germfree rats and correspondingly less radioactivity reached the cecum during the 6-hour period following intragastric administration. When corrections were made for differences in gastric emptying, germfree and conventionalized rats absorbed oleic acid and triolein at similar rates. Under the conditions of our studies, intestinal microorganisms did not appear to influence the rate of either lipolysis or fatty acid absorption directly, but significantly influenced the rate at which fat was transported along the gastrointestinal tract.

PROTECTION BY OROTIC ACID AGAINST THE RENAL NECROSIS AND FATTY LIVER OF CHOLINE DEFICIENCY. J. B. Simon, R. Scheig and G. Klatskin (Dept. of Internal Med., Yale Univ. School of Med., New Haven, Conn. 06510). *Proc. Soc. Exp. Biol. Med.* 129, 874–7 (1968). Addition of 1% orotic acid to a choline-deficient diet lowered the incidence of hemorrhagic renal necrosis in young rats from 86% to 41%, and within 24 hr reduced the accumulation of hepatic triglycerides by almost 50%. Simultaneous supplementation of the diet with 0.25% adenine sulfate did not influence these protective effects of orotic acid in choline deficiency. It is suggested that orotic acid may lower the requirement of the body for choline through a metabolic interaction.

CARCASS QUALITY OF TURKEYS AS AFFECTED BY ESTRADIOL-17-MONOPALMITATE AND VITAMIN E. I. EFFECT ON BREEDER HEN TURKEYS. L. D. Pickett, B. F. Miller and R. E. Moreng (Dept. of Avian Sci., Colorado State Univ., Fort Collins, Colorado 80521). *Poultry Sci.* 47, 1488–92 (1968). One hundred twenty-five Broad Breasted White turkey breeder hens

A, or with 5 mg vitamin A acetate/kg. Some of the vitamin A-supplemented rats were pair-fed with deficient rats to obviate the effect of inanition. After 42 to 60 days, weight loss characteristic of vitamin A deficiency occurred; animals were then killed and liver mitochondrial lipids were analyzed. Cholesterol and triglycerides from vitamin A-deficient animals were significantly higher than those of corresponding pair-fed animals and controls fed *ad libitum*. The deficient rats also showed a significant decrease in liver mitochondrial lipid phosphorus and total lipid. Mitochondrial lipids from the pair-fed controls were significantly higher in arachidonic and linoleic acids when compared with deficient rats or with controls whose food intake was not restricted. Vitamin deficient rats and pair-fed controls had significantly less palmitoleic acid in their mitochondrial lipids than did control rats fed *ad libitum*.

STEROL PRECURSORS OF CHOLESTEROL IN ADULT HUMAN BRAIN. H. A. Lowenstam and D. McConnell (Calif. Inst. of Technol., Pasadena 91109). *Science* 162, 1495–7 (1968). Adult human brain contains cholestanol and two series of cholesterol precursors having 30, 29, 28, and 27 carbon atoms; one has an unsaturated steroid nucleus, and the other is unsaturated in both nucleus and side chain. The ability of preparations of brain to incorporate a specific precursor into cholesterol, as well as into these sterol metabolites *in vitro*, indicates that cholesterol synthesis continues long after brain maturation.

SYNERGISM BETWEEN CYCLOPROPENOID FATTY ACIDS AND CHEMICAL CARCINOGENS IN RAINBOW TROUT (SALMO GAIRDNERI). D. J. Lee, J. H. Wales, J. L. Ayres and R. O. Sinnhuber (Dept. of Food Sci. and Technol., Oregon State Univ., Corvallis, Oregon 97331). *Cancer Res.* 28, 2313–8 (1968). The synergistic activities of cyclopropenoid fatty acids (CPFA), epoxyoleic acid, sesame seed oil and autoxidized salmon oil with aflatoxin B_1 were studied in rainbow trout. *Sterculia foetida* oil (49% sterculic, 7% malvalic acids) and *Hibiscus syriacus* oil (2% sterculic and 19% malvalic acids) were used as sources of CPFA. The combination of both 112 and 56 ppm CPFA procured from *S. foetida* oil and 210 ppm CPFA procured from *H. syriacus* oil promoted early tumor development, increased tumor incidence, and caused a several-fold increase in tumor growth rate over the positive control. Trout fed 100 ppm 2-acetylaminofluorene for 15 months did not develop hepatomas, but the addition of *H. syriacus* oil to the diet induced a 20% incidence of liver tumors. Diets containing 200 ppm epoxyoleic acid (in *Vernonia anthelmintica* oil), 5% sesame seed oil, or 5% autoxidized salmon oil (peroxide value 200–300) did not alter the carcinogenicity of aflatoxin B_1. Some unusual lipid deposits were noted in the livers of fish receiving sesame seed oil.

THE METABOLISM OF GLUCOSE-2-T BY ADIPOSE TISSUE. J. Katz and R. Rognstad (Cedars-Sinai Med. Res. Inst., Cedars-Sinai Med. Center, Los Angeles, Calif. 90029). *J. Biol. Chem.* 244, 99–106 (1969). Rat epididymal fat pad segments were incubated with glucose labeled uniformly with ^{14}C, and with tritium in position 2, and in positions 1, 3, and 6. Under conditions of high lipogenesis, 70 to 80% of tritium (T) from position 2 was recovered in water, with the remainder in lipid glycerol, fatty acids and lactate. The highest tritium retention, relative to carbon, was in glycerol, with $T:^{14}C$ ratios from glucose uniformly labeled with ^{14}C and with tritium in position 2 in the presence of insulin of 0.6 to 0.8. Of this tritium, 12 to 15% was on C-1 of glycerol.

SULFATIDES AND SODIUM ION TRANSPORT, SPHINGOLIPID COMPOSITION OF THE RECTAL GLAND OF SPINY DOGFISH. K. Karlsson, B. E. Samuelsson and C. O. Steen (Inst. of Med. Biochem., Univ. of Goteborg, Fack, 400 33 Goteborg 33, Sweden). *FEBS Letters* 2, 4–6 (1968). It was recently found, that sulfatides are present in relatively high concentrations in the outer part of kidney medulla and in the avian salt gland, tissues known to have a corticosteroid dependent sodium ion transport. Based on these findings, it was proposed that sulfatides are involved as carriers or receptors in this process. To evaluate this hypothesis further, the rectal gland of spiny dogfish was analyzed for the presence of sulfatides. The only known function of this organ, as well as of the salt gland, is excretion of sodium chloride in a concentration higher than that of sea water.

MILK FAT SYNTHESIS ON RESTRICTED-ROUGHAGE RATIONS CONTAINING WHEY, SODIUM BICARBONATE AND MAGNESIUM OXIDE. J. T. Huber, R. S. Emery, J. W. Thomas and I. M. Yousef (Dairy Dept., Michigan State Univ., East Lansing). *J. Dairy Sci.* 52, 54–9 (1969). A study was conducted to elucidate the mechanism through which partially delactosed whey and minerals ($NaHCO_3$ plus MgO) increase milk fat synthesis on restricted-roughage rations and to determine the minimum effective level of the whey. Thirty-six Holstein cows were fed concentrate *ad libitum* and 2.3 kg hay/day for eight weeks. Concentrates contained varying levels of whey (0, 3, 4, and 14%), or minerals (2.5% $NaHCO_3$ and 1% MgO), or both whey (14%) and minerals. Fat depression was least on the mineral rations, but concentrate intake and milk yields were also lowest. As whey in concentrate increased, milk fat also increased, with 14% whey almost as effective as minerals.

EFFECT OF DIETARY PROTEIN SOURCE AND CORN OIL AND CELLULOSE LEVELS ON STRONTIUM-CALCIUM DISCRIMINATION IN GROWING RATS. E. W. Hartsook, R. L. Cowan, P. T. Chandler and J. B. Whelan (Dept. of Animal Sci., Pa. State Univ., Univ. Park, Pa.). *J. Nutr.* 97, 95–103 (1969). The effect of diet composition on strontium-calcium discrimination in the growing rat was investigated using tracer doses of ^{89}Sr and ^{45}Ca administered by stomach tube to both sexes in a $2 \times 2 \times 3$ factorial experiment whose variables in addition to sex (S) were protein (P), casein or zein at a 20% dietary level, cellulose (C), at a 3 or 12% dietary level, and corn oil (CO), at a zero, 4 or 8% dietary level. Significant main effects of treatments were: casein produced 41 and 42% greater absorption and 48 and 59% greater femur content of ^{89}Sr and ^{45}Ca, respectively, than zein, but did not alter either $^{89}Sr/^{45}Ca$ absorbed or $^{89}Sr—^{45}Ca$ in femurs. Casein produced a 69% reduction of serum ^{45}Ca as compared with zein which caused the $^{89}Sr/^{45}Ca$ in serum to be 534% greater than the value for zein. When dietary cellulose increased from 3 to 12%, ^{89}Sr of bone and ^{45}Ca of serum were decreased by 21 and 25%, respectively. The main effect of dietary level of corn oil was nonsignificant. Significant interactions for respective criteria were: absorbed ^{45}CA, C with CO; bone ^{89}Sr, C with CO; bone ^{45}Ca, C with CO; $^{89}Sr/^{45}Ca$ in femur, P with S and P with CO; serum ^{89}Sr, P with C; and serum ^{45}Ca, P with C and S with C.

EFFECT OF DIETARY FAT SOURCE ON THE APPARENT DIGESTIBILITY OF FAT AND THE COMPOSITION OF FECAL LIPIDS OF THE YOUNG PIG. R. M. G. Hamilton and B. E. McDonald (Dept. of Animal Sci., Macdonald College (McGill Univ.), Montreal, Quebec, Canada). *J. Nutr.* 97, 33–41 (1969). Two experiments, each of 5 weeks duration, were conducted with 18-day-old weanling pigs to determine the effect of dietary fat source and age of animal on apparent fat digestion and absorption. In each experiment two males and two females were fed diets that contained 10% coconut oil, rapeseed oil, lard or tallow. Gas-liquid chromatographic analyses indicated that dietary fatty acid composition influenced the fatty acid patterns of the fecal lipids, although there were notable qualitative and quantitative differences. Total saturated fatty acids constituted a higher proportion of the FFA fraction than of the dietary lipid fraction or of the triglyceride fraction for groups fed rapeseed oil, lard or tallow. Apparent digestibilities of palmitic and stearic acids were lower than those of the unsaturated or medium-chain saturated fatty acids.

"SOLUBILIZATION" OF THE STEAROYL-CoA DESATURASE OF RAT LIVER MICROSOMES. M. I. Gurr, K. W. Davey and A. T. James (Unilever Res. Lab., Colworth House, Sharnbrook, Bedford, England). *FEBS Letters* 1, 320–2 (1968). Nearly all the many fatty acid desaturating enzymes which have so far been investigated are firmly bound to cellular particles or membrane systems. As far as we are aware, the stearoyl-ACP desaturase of *Euglena gracilis* is the only well documented example of a "soluble" desaturase. A short report in 1964 indicated that it might be possible to obtain the rat liver microsomal stearoyl-CoA desaturase in a soluble, partially purified form. We are interested in studying the mechanism by which the two hydrogen atoms are removed, in a highly specific way, from a saturated fatty acid chain during the enzymic formation of a double bond. Studies of this kind demand a soluble highly purified enzyme and in this paper we shall describe some initial steps towards this objective.

ENZYMIC CONVERSION OF LINOLEIC ACID TO ETHYLENE BY EXTRACTS OF APPLE FRUITS. T. Galliard, A. C. Hulme, M. J. C. Rhodes and L. S. C. Wooltorton (Agr. Res. Council, Food Res. Inst., Colney Lane, Norwich, England). *FEBS Letters* 1, 283–86 (1968). The origin of ethylene, the ripening hormone of many fruits, has yet to be defined. A recent review described possible sources of ethylene and from recent studies with both plant preparations and model systems, methionine (*via* methional), acetaldehydes, propionaldehydes and linolenic acid have emerged as possible precursors of ethylene. The present paper describes the enzymic conversion of linolenic acid to ethylene by cell-free extracts of apple fruits in the presence of oxygen

(Continued from page 237A)

system used, two or more components (polypeptides) were seen, presumably reflecting apoprotein aggregational differences under the several conditions of study. These observations strongly suggest that apo HDL consists of two different polypeptides: one rich in cystine, and one not.

INFLUENCE OF CARBON TETRACHLORIDE, VITAMIN E AND PROTEIN UPON LIVER SLICE RESPIRATORY ACTIVITY. E. M. Blendermann and L. Friedman (Div. of Nutr., Food and Drug Admin., U.S. Dept. of Health, Education and Welfare, Washington, D.C. 20204). *Proc. Soc. Exp. Biol. Med.* 129, 831–36 (1968). Vitamin E deficiency and low protein intake, factors reported to accentuate the overt toxicity symptoms of CCl_4 in rats, were investigated for their effects on CCl_4-induced alteration of carbohydrate metabolism by rat liver slices. Rats fed for 5 weeks on purified diets containing 18 or 10% casein with or without added vitamin E were fasted 24 hr, intubated orally with 1 ml of CCl_4 in olive oil per kg of body weight, and then fasted during the subsequent 24 hr. Protection against CCl_4-induced lethality by adequate dietary protein and/or vitamin E was observed. However, the following CCl_4 effects were unaffected by lowering dietary casein or vitamin E levels: (i) increased liver size and fat content; (ii) lowered oxygen uptake and increased production of $^{14}CO_2$ from uniformly labeled glucose-^{14}C by liver slices; and (iii) a net production of lactic acid (seen only with liver slices from CCl_4-dosed rats). Thus it is concluded that in rats, the protection against CCl_4 toxicity by dietary vitamin E or adequate protein levels is not related to any effects on these parameters of CCl_4-altered carbohydrate metabolism.

DEDIFFERENTIATION OF PHOSPHOLIPID COMPOSITION IN SUBCELLULAR PARTICLES OF CANCER CELLS. L. D. Bergelson, E. V. Dyatlovitskaya, T. I. Torkhovskaya, I. B. Sorokina and N. P. Gorkova (Inst. of Chem. of Nat. Compounds of the Academy of Sci., of the USSR, Moscow, Russia). *FEBS Letters* 2, 87–90 (1968). It has been shown that specificity of phospholipid distribution no longer occurs in subcellular particles from cancer cells. Thus, the mitochondria, microsomes and nuclei of Zajdela ascites rat hepatoma and the mitochondria and microsomes of solid mouse hepatoma 22 contain all the individual phospholipids present in the whole tissue. This lack of specificity of subcellular phospholipid distribution in tumor cells becomes further evident on examination of the "enrichment factors" obtained by comparing the amount of a given phospholipid component per total phospholipids in each subcellular fraction with the value for the whole tissue homogenate.

PLASMINOGEN ACTIVATOR ACTIVITY OF RAT LYSOSOMES. E. L. Beard, M. H. Montuori, G. J. Danos and R. W. Busuttil (Dept. of Biol. Sciences, Loyola Univ. of the South, New Orleans, Louisiana 70118). *Proc. Soc. Exp. Biol. Med.* 129, 804–08 (1968). Rat tissues have been fractionated in order to assay their plasminogen activator activity. Mitochondria, microsomes and the final supernate separated from rat liver were essentially equal in this activity suggesting that the soluble plasminogen activator in the cell homogenate had equilibrated among them. Lysosomes showed significantly greater plasminogen activator activity than the other fractions. The plasminogen activator release from lysosomes could be increased by sonification and freezing and thawing. When the lysosomes of a series of six tissues were compared, lung lysosomes were richest in plasminogen activator. The activator activity was approximately the same for lysosomes of rat spleen, kidney, liver, brain and lymph node. Proteolytic activity of rat liver cell fractions was less than ¼ that measured in the presence of plasminogen. Fibrinolytic activity of cell fractions measured with heated fibrin clots was negligible.

BROWN ADIPOSE CELLS: SPONTANEOUS MOBILIZATION OF ENDOGENOUSLY SYNTHESIZED LIPID. A. Angel (Dept. of Med., Univ. of Med., Univ. of Toronto, Toronto 5, Canada). *Science* 163, 288–90 (1969). Isolated brown adipose cells, devoid of a basement membrane, readily synthesized a variety of lipids from radioactive acetate, a reaction augmented by glucose and insulin. A large proportion of the newly formed fatty acids passed into the incubation medium. In intact brown adipose slices and isolated white adipose cells, most of the synthesized lipid was retained as glyceride esters. The data suggest that the rapid turnover of endogenously synthesized lipid in brown adipose cells is almost totally obscured in studies with intact tissue slices because of interstitial barriers to the egress of fatty acid.

STUDIES ON PHOSPHOMUTASES. III. PHOSPHORYL TRANSFER TO PHOSPHOGLUCOMUTASE FROM 1,3-DIPHOSPHOGLYCERATE. J. B. Alpers and G. K. H. Lam (Dept. of Biol. Chem., Harvard Med. School, Boston, Mass. 02115). *J. Biol. Chem.* 244, 200–04 (1969). Phosphoglucomutase from rabbit skeletal muscle has been labeled with ^{32}P by incubation with 1,3-diphosphoglycerate-1-^{32}P. The product was not retained on columns of carboxymethyl cellulose and could thus be separated from nonphosphorylated species. Incubation with 1,3-diphosphoglycerate caused an increase in the proportion of phosphoenzyme as judged chromatographically. The evidence that the labeling occurred at the enzyme's active site is that (a) the incorporation was inhibited by glucose 1-phosphate; (b) the incorporated label was not removed by boiling in N acid but was labile to weak alkali; (c) incubation of the reisolated enzyme with glucose-1-P released the label as organic phosphate; and (d) in all studies, the labled enzyme behaved the same as that labeled by exchange with glucose phosphates. Phosphoryl transfer from 1,3-diphosphoglycerate to phosphoglyceratemutase, phosphorylase b, or the phosphoenzyme form of phosphoglucomutase was not observed.

THE IN VITRO CATABOLISM OF CHOLESTEROL. A COMPARISON OF THE FORMATION OF 26-HYDROXYCHOLESTEROL AND CHENODEOXYCHOLIC ACID FROM CHOLESTEROL IN RAT LIVER. D. Mendelsohn and L. Mendelsohn (Dept. of Chem. Pathol., Witwatersrand Univ. Med. School, Johannesburg, South Africa). *Biochemistry* 7, 4167–71 (1968). The conversion of cholesterol into 26-hydroxycholesterol and chenodeoxycholic acid has been studied using a rat liver preparation *in vitro*. The results show that cholesterol can be enzymatically converted into chenodeoxycholic acid by this system. However, no definite evidence for the enzymatic formation of 26-hydroxycholesterol from cholesterol was obtained. In all instances where radioactive 26-hydroxycholesterol was isolated, further purification of this compound revealed that most of the radioactivity was due to an autoxidation contaminant of cholesterol, most probably 25-hydroxycholesterol. Since the enzyme system employed in this study is able to transform cholesterol into both cholic and chenodeoxycholic acid, the fact that no enzymatic conversion of the former compound into 26-hydroxycholesterol was observed indicates that 26-hydroxycholesterol is probably not an important intermediate in bile acid genesis.

STEROID-PROTEIN INTERACTIONS. XVIII. ISOLATION AND OBSERVATIONS ON THE POLYMERIC NATURE OF THE CORTICOSTEROID-BINDING GLOBULIN OF THE RAT. G. J. Chader and U. Westphal (Biochem. Dept., Univ. of Louisville, School of Med., Louisville, Ky. 40208). *Biochemistry* 7, 4272–82 (1968). Chromatographic techniques in combination with gel filtration resulted in the isolation of a corticosteroid-binding globulin from pooled rat serum. The isolated corticosteroid-binding globulin-corticosterone complex was homogeneous by sedimentation velocity ($s^0_{20,w} = 3.56$ S), paper electrophoresis, and immunoelectrophoresis (α_1-globulin). A molecular weight of 61,000 ± 1100 was obtained by the approach to sedimentation equilibrium method whereas the corticosterone content indicated a molecular weight of approximately 53,000 for the active steroid-binding species. A carbohydrate content of 27.8% was found. Distinct differences in certain amino acid residues such as half-cystine may account for differences in steroid-binding and polymeric properties between rat corticosteroid-binding globulin and corticosteroid-binding globulin from the human and rabbit.

ORGANOLEPTIC PROPERTIES AND GAS CHROMATOGRAPHY PATTERNS OF STEAM DISTILLATES FROM FRESH AND STALE MILK FAT. A. Tamsma, F. E. Kurtz, A. Kontson and M. J. Pallansch (Dairy Products Lab., Eastern Utilization Res. and Dev. Div., USDA, Washington, D.C.). *J. Dairy Sci.* 52, 152–57 (1969). We have examined the chromatographic patterns as well as the flavor characteristics of steam distillates of milk fat to obtain a more objective procedure than that given solely by taste panel evaluation for folowing the development of stale flavor during the storage of fat-containing dairy products. Both fresh and stale milk fats were steam deodorized at 50 and 75C at 1 mm. Stale milk fat was also steam deodorized at 100C (1 mm) and at 50C (3 mm). One-millimeter pressure is preferable to higher pressures. Distillation at 75C or higher produces artifacts, volatile compounds generated from precursors, undesirable for following the formation of stale-flavor compounds during storage. Volatiles are recovered less completely at 50C than at higher temperatures, but artifact generation is low. Moreover, the flavor of volatiles recovered at 50C is characteristic of stale fat, and the chromatographic patterns of the 11 most prominent peaks correlate well with the flavor intensity and with the expected relative concentrations of the pertinent off-flavor compounds produced by storage of fats at various temperatures.

UPTAKE OF LABELED LONG CHAIN FATTY ACIDS IN VIVO AND IN VITRO BY DIFFERENT PHOSPHOLIPIDS IN MILK. M. M. A. Al-Shabibi, J. Tobias and R. E. Brown (Dept. of Dairy Science, Univ. of Illinois, Urbana). *J. Dairy Science* 52,

and 0.6 mg/10^9 cells of free and esterified cholesterol, respectively. Predominance of free cholesterol in leukocytes (free-ester ratio = 3:1) contrasted with predominance of esterified cholesterol in plasma (1:2.5). Twenty-two rabbits received intragastric administration of cholesterol-4-^{14}C and were killed after 4 hrs, 18 hrs, 7 days and 14 days. The cholesterol fractions of leukocytes and plasma were assayed by liquid scintillation counting. In leukocytes, the free fraction of labeled cholesterol was highest after 7 days, but the esterified fraction remained low throughout the 14-day period. In plasma, the labeled cholesterol was highest also after 7 days, but the esterified fraction was higher than the free throughout the experiment. The results indicate that blood leukocytes play a role in the transport of cholesterol in the circulation.

DIMERS AND TRIMERS OF α-TOCOPHEROL: METABOLIC AND SYNTHETIC STUDIES. B. S. Strauch, H. M. Fales, R. C. Pittman and J. Avigan (Lab. of Metabolism, Nat. Heart Inst., Nat. Inst. of Health, Bethesda, Md.). *J. Nutr.* 97, 194–202 (1969). The metabolism of α-tocopherol was studied in rats and mice. Evidence has been presented for formation of dimers and trimers of α-tocopherol in animals that received parenteral α-tocopherol. These products were shown to be identical with synthetic products of oxidative condensation of α-tocopherol. The structure of these synthetic compounds was extensively studied by gas-liquid chromatography and mass spectrometry providing evidence of the necessity for modification of previously proposed structures. Dimers and trimers were not demonstrable after oral administration of α-tocopherol to rats. It is suggested that oxidative condensation does not represent a quantitatively important metabolic pathway under physiological conditions.

SERUM CHOLESTEROL AND GLUCOSE LEVELS IN RATS FED REFINED AND LESS REFINED SUGARS AND CHROMIUM. H. Schroeder (Dept. of Physio., Dartmouth Med. School, Hanover, N. Hamp.). *J. Nutr.* 97, 237–42 (1969). Refined sugar (sucrose) contains less chromium than partly refined sugar. Because human hypercholesteremia and diabetes mellitus have been believed associated with the consumption of sugar, 200 weanling rats were given a low chromium diet of Torula yeast, lard and sucrose, with essential trace metals in drinking water, to ascertain effects on fasting serum cholesterol and glucose levels. Groups were given white sugar containing 0.002 ppm Cr without and with chromium (III) supplementation (5 ppm in drinking water), less refined "raw" sugar containing 0.06 ppm Cr, and still less refined brown sugar with 0.12 ppm Cr. Serum cholesterol levels were relatively elevated and increased with age in the group receiving white sugar; in that given white sugar plus chromium or brown sugar they were low. Effects were similar in both sexes. Younger rats fed raw sugar had lower levels than those fed white. Fasting serum glucose was relatively low in rats fed brown sugar and in female fed white plus chromium; minimal effects occurred in those given "raw" sugar. These data offer evidence that refined sugar without chromium can relatively elevate serum cholesterol and glucose levels, and that chromium (III) can lower both substances.

CORRELATION BETWEEN CONCENTRATION OF SERUM FREE FATTY ACIDS AND KETONE BODIES IN ALLOXAN DIABETES. B. Rudas and K. Reissert (Inst. for Gen. and Exp. Pathology, Univ. of Vienna, Austria). *Proc. Soc. Exp. Biol. Med.* 130, 243–46 (1969). Lard was given orally to rats 6 days and 6 weeks after alloxan administration and to insulin-treated alloxanized rats 6 days and 6 weeks after insulin withdrawal. Two hours after the fat feeding serum FFA and blood ketone concentrations were higher but serum FFA levels were lower in the acutely than in the chronically insulin deficient rats. The results indicate that in alloxan diabetic rats the degree of ketonemia is not proportional to the level of serum FFA but seems partly to depend on the duration of insulin deprivation.

A COMPARATIVE STUDY OF ALKALINE LIPOLYTIC ACTIVITY IN ADIPOSE TISSUE OF VARIOUS MAMMALS. R. Rivello, J. Cortner and J. Schantz (Depts. of Pediatrica and Med., State Univ. of N.Y. at Buffalo). *Proc. Soc. Exp. Biol. Med.* 130, 232–35 (1969). The alkaline lipolytic activity (ALA) of human adipose tissue has been characterized previously. The present study reports the variations in quantitative activity and starch gel zymograms in adipose tissue of nine other mammals.

EFFECTS OF TWO SYNTHETIC ANTIOXIDANTS, VITAMIN E, AND ASCORBIC ACID ON THE CHOLINE-DEFICIENT RAT. P. M. Newberne, M. R. Bresnahan and N. Kula (Dept. of Nutr., MIT, Cambridge, Mass.). *J. Nutr.* 97, 219–31 (1969). The purpose of this work was to determine the effects of two synthetic antioxidants, butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT), dl-α-tocopherol and ascorbic acid on the fatty liver and on the renal and cardiovascular systems of choline-deficient rats. Weanling and 6-week-old male rats were fed casein-peanut meal diets devoid of choline for 8 and 10 days, respectively. Survivors were fasted and then killed, and tissues collected for serum and liver lipid analyses and for morphologic assessment of tissue damage. Fatty liver, hemorrhagic kidneys and cardiovascular damage characteristic of choline deficiency were observed in deficient animals of both ages. However, BHA and BHT protected rats in both age groups from heart and aorta damage and also prevented renal damage in the week-old group. Mortality was decreased by tocopherol, ascorbate and BHA and BHT, but most notably by the latter two. In weanling rats all additives increased serum lipids and all except ascorbate decreased liver lipids. In 6-week-old rats serum lipids varied, whereas liver lipids were increased by tocopherol and ascorbate and decreased by BHA and BHT. We conclude that tocopherol and BHA and BHT tend to decrease the effects of choline deficiency on the liver, renal and cardiovascular systems whereas ascorbate enhances them. Possible mechanisms for the various effects are discussed.

PHOSPHOLIPID METABOLISM DURING AMINO ACID TRANSPORT IN HAMSTER SMALL INTESTINE. M. McLeod and R. Bressler (Depts. of Med., Physio., and Pharm., Duke Univ., Durham, N. Car.). *Proc. Soc. Exp. Biol. Med.* 130, 268–73 (1969). Previous studies of phospholipid metabolism during active transport have used ^{32}P and ^{14}C glycerol as a measure of phospholipid metabolism. In this investigation linoleic-1-^{14}C-albumin and ^{32}P were used in studying phospholipid metabolism during amino acid transport in the hamster small intestine. The results demonstrate an augmented incorporation of linoleic-1-^{14}C acid into lecithin at the site of maximum amino acid transport. The enhanced incorporation of fatty acid into lecithin may be related to: (a) a diacyl-monacyl phosphoglyceride cycle resulting in a lamellar-micellar shift in this membrane phospholipid associated with transport and/or (b) enhanced phospholipid fatty acid oxidation occurring with transport, or (c) other undefined mechanisms.

A REQUIREMENT FOR LIPIDS BY THE MICROSOMAL STEARYL COENZYME A DESATURASE. P. D. Jones, P. W. Holloway, R. O. Peluffo and S. J. Wakil (Dept. of Biochem., Duke Med. Center, Durham, N.C. 27706). *J. Biol. Chem.* 244, 744–54 (1969). Hen liver microsomes, when extracted with aqueous acetone, lose their ability to desaturate stearyl coenzyme A to oleate. The desaturase activity of these particles is partially restored by addition of lipid micelles prepared from microsomal lipid. Fractionation of the microsomal lipid yields fractions which are tested for their ability to restore the desaturase activity. A mixture of phospholipids, triglycerides, and fatty acids is able to restore the desaturase activity of the acetone-extracted microsomes to the level of the original microsomes. Phosphatidylcholine, isolated from the phospholipid, can partially replace phospholipids in this mixture. The properties of the reconstituted system are similar to those of the original microsomes. There is an absolute requirement for oxygen and a reduced pyridine nucleotide, although DPNH, rather than TPNH, is the preferred electron donor. The similarities between the lipid requirements of the DPNH-cytochrome c reductase and the stearyl-CoA desaturase support the suggestion that the two systems share a common electron transfer pathway or carrier.

BIOSYNTHESIS OF SPHINGOLIPID BASES. III. ISOLATION AND CHARACTERIZATION OF KETONIC INTERMEDIATES IN THE SYNTHESIS OF SPHINGOSINE AND DIHYDROSPHINGOSINE BY CELL-FREE EXTRACTS OF HANSENULA CIFERRI. R. N. Brady, S. Di Mari and E. E. Snell (Dept. of Biochem., Univ. of Calif., Berkeley, Calif. 94720). *J. Biol. Chem.* 244, 491–96 (1969). In the absence of reduced triphosphopyridine nucleotide, two isotopically labeled ketones were found to accumulate when L-serine and ammonium palmitate-^{14}C were incubated with ATP, coenzyme A and a cell-free particulate preparation from the yeast *Hansenula ciferri*. These compounds, in the form of their N-acetyl-, trimethylsilyl-, and 2,4-dinitrophenylhydrazone derivatives, have been identified as 3-ketodihydrosphingosine (1-hydroxy-3-oxo-2-aminooctadecane) and 3-ketosphingosine (1-hydroxy-3-oxo-2-aminooctadec-4-ene) by thin-layer chromatography and gas-liquid chromatography, and by repeated crystallizations of N-acetyl-3-ketodihydrosphingosine to constant specific radioactivity. In the presence of TPNH, these compounds are enzymatically reduced to dihydrosphingosine and sphingosine. These findings indicate that 3-ketodihydrosphingosine and 3-detosphingosine are intermediates in the biosynthesis of dihydrosphingosine and sphingosine, respectively, and that in yeast the biosynthetic pathways leading to saturated and unsaturated sphingolipid bases diverge at some point before reduction of the ketonic intermediates with TPNH.

• Drying Oils and Paints

ADSORPTION OF WATER VAPOUR, STEARIC ACID, STEARYL ALCOHOL AND STEARIC ACID MODIFIED ALKYD RESINS ONTO RUTILE TITANIUM DIOXIDE PIGMENTS. T. Doorgeest (Verfinstituut TNO, Schoemakerstraat 97, Delft, Netherlands). *J. Oil Colour Chem. Assoc.* 50, 1079–1114 (1967). In a systematic investigation of the interaction of pigments and binders, adsorption isotherms of water vapour, stearic acid, stearyl alcohol and stearic acid modified alkyd resins are determined. Rutile titanium dioxide pigments, differing in surface structure, are used as adsorbents. The adsorption of stearic acid, stearyl alcohol and alkyd resin is affected by small percentages of water as well as by the type of solvent used. Water catalyzes chemical reactions between stearic acid and titanium dioxide pigments containing zinc oxide and/or an alumina-silica coating. A correlation exists between the quantity of alkyd resin adsorbed per gram of pigment and the specific surface area, as determined from the adsorption of stearic acid or stearyl alcohol. From this it is concluded that alkyd resin molecules, stearic acid molecules and stearyl alcohol molecules are adsorbed on to the same surface sites.

CRITICAL COMPOSITIONS OF LINSEED OIL DERIVATIVES: WATER SOLUBLE TYPES. A. E. Rheineck and R. A. Heskin (No. Dakota State Univ., Fargo, N.D.). *Paint Technol.* 40, 450–458 (1968). The work described was prompted by the need for a liquid curing membrane for wet concrete. Criteria for such a membrane are water thinnable, must form an essentially water vapor impervious coating, and must dry to a good film under adverse conditions. Furthermore, the composition was to be based upon linseed oil and delivered to the job as a high solids, readily dilutable with water system. Two families of coatings were prepared by reacting linseed oil with maleic anhydride and fumaric acid respectively. After unreacted acidic components were removed, the residual acidity was neutralized with various amines in a water/butyl cellosolve solution. The water tolerances (dilution characteristics), water vapor transmission and dry film characteristics were determined. The most soluble (water) compositions were fumaric acid adducts neutralized with either di-ethyl or tri-ethyl amine. Between 10% and 14% fumaric acid addition products were preferred.

• Detergents

EFFECT OF INITIAL SURFACTANT LOCATIONS ON THE VISCOSITY OF EMULSIONS. T. J. Lin (Max Factor, 1655 N. McCadden Pl., Hollywood, Calif. 90028). *J. Soc. Cosmetic Chemists* 19, 683 (1968). Viscosities of emulsions immediately following homogenization were studied as a function of HLB and the initial surfactant locations. Keeping the total surfactant concentration constant, the ratios of the initial concentration of the hydrophilic surfactant to that of the lipophilic surfactant in each phase were varied. The experimental results indicate that the initial locations of the surfactants not only affect the initial viscosity of the emulsions but also the emulsion stability, particle size distribution, and emulsion type as well.

CHROMATOGRAPHIC RESOLUTION OF ETHYLENE OXIDE ADDUCTS INTO THEIR HOMOLOGUES AND THEIR QUANTITATIVE DETERMINATION. R. Wickbold (Chem. Werke Huls AG, Marl, Ger.). *Fette Seifen Anstrichmittel* 70, 688–692 (1968). The separation of ethylene oxide condensates in their polymeric homologues by gas chromatography is not satisfactory due to the limited volatility of the components having a large number of ethylene oxide groups. Column chromatography with silica gel as absorbent and butanone as eluting medium enables a fractionation of such adducts up to 14–16 membered homologues of the series. This separation occurs solely according to the length of the ethylene oxide chain. The eluted substances are determined in the fractions and the components present in each fraction identified by thin-layer chromatography. In two-component mixtures, the amounts of individual substances are estimated by comparing the intensity of the spots. The possibility of simplifying the apparatus is discussed.

BIOCHEMICAL DEGRADABILITY OF A SECONDARY ALKANE SULFONATE UNDER CONDITIONS EMPLOYED IN THE LABORATORY AND PRACTICE. M. Krone and G. Schneider (Farbwerke Hoechst AG, Frankfurt (M)-Hochst, Ger.). *Fette Seifen Anstrichmittel* 70, 753–757 (1968). The biochemical degradability of a secondary alkane sulfonate (SAS) developed by Farbwerke Hoechst AG was investigated. For comparison, a known, fairly degradable linear alkyl benzolsulfonate (LAS) was chosen. The products were tested under identical conditions initially in a laboratory procedure and later in a technical plant. All experiments revealed a better biological degradability of SAS than LAS.

TRANS-ALPHA-BETA-UNSATURATED FATTY ACIDS AND PRODUCTS FORMED BY THEIR REDUCTION WITH LITHIUM-ALUMINUM-HYDRIDE AND DI-ISOBUTYL-ALUMINUM-HYDRIDE. F. Falk and C. Mainas (Inst. for Fat. Chem., Ger. Acd. of Nut. of Berlin, Berlin-Adlershof, Ger.). *Fette Seifen Anstrichmittel* 70, 709–714 (1968). A method for the preparation of pure methyl esters of alpha, beta-unsaturated fatty acids is described. The reduction of these esters with lithium-aluminum-hydride led to poor yields of alpha, beta-unsaturated alcohols. Saturated alkanols and aldehydes were mainly formed. Di-isobutyl-aluminum-hydride gave near quantitative yields of alpha, beta-unsaturated alcohols. The reaction was studied with the use of gas chromatography using strongly polar stationary phases.

TURBIDITY POINT OF NONIONIC SURFACTANTS. H. Lange (Phy.-Chem. Lab. of Henkel and Cie., Dusseldorf, Ger.). *Fette Seifen Anstrichmittel* 70, 748–753 (1968). The occurrence of turbidity above a definite temperature in aqueous solutions of many nonionic surfactants, especially in those of ethylene oxide adducts, is due to separation of two liquid phases and not due to dehydration of the hydrophilic group as is often assumed. In discussing the above phenomena the thermodynamic laws governing the resolution of liquid two-component systems are taken into consideration.

THE DEVELOPMENT OF ENZYMES FOR USE IN WASHING AGENTS. J. C. Hoogerheide (Dev. Lab. Royal Neth. Ferm. Ind., Delft, Holland). *Fette Seifen Anstrichmittel* 70, 743–748 (1968). A description of the properties of enzymes which are suitable as additives to biologically active washing agents is presented. Methods for the determination of the above properties are reported. The investigations showed that the efficacy of a biologically active washing agent depends not only upon the properties of the enzyme but on the composition of the washing agent as well. The combination of enzymes and surfactants leads to a synergic effect. The further possibilities of development in the field of biologically active washing agents are given.

AUTOMATIC FINE SOAP PRODUCTION LINE AND THE EQUIPMENT AND MACHINES INVOLVED. H. Zilske (Wolfenbuttel, Ger.). *Fette Seifen Anstrichmittel* 70, 502–509 (1968). A description of the structure and working principle of a production line for the manufacture of high quality fine soap using recently developed machines is presented. An important unit of the plant is a new type of dryer which operates exclusively in the liquid phase. Soap is solidified only after the predetermined fatty acid content (up to 84%) is attained. The dust free

working condition combined with the absence of any overdried particles (spikes) substantially reduces the power requirement. Liquid drying allows soap to be dried immediately in the form of ribbons thus enabling the production of fully homogeneous, compact and non-cracked fine soaps.

FOAM STABILIZATION AND FOAM INHIBITION. E. Schmadel (Tech. Appl. Lab., Henkel and Cie, Dusseldorf, Ger.). *Fette Seifen Anstrichmittel* **70**, 491–495 (1968). A general review of the field of foam stabilization and foam inhibition in the area of washing, rinsing and cleaning agents in addition to recent developments in the field of foam inhibition is presented. It is possible to regulate foam formation by varying the temperature independent of the hardness of water. The mechanisms of foam stabilization and foam inhibition are dealt with.

DETERMINATION OF CHELATING SUBSTANCES IN WASHING AGENTS. E. Heinerth (Anal. Lab., Henkel and Cie., Dusseldorf, Ger.). *Fette Seifen Anstrichmittel* **70**, 495–498 (1968). Methods for the thin-layer chromatographic identification and titrimetric determination of chelating substances in washing agents are described.

DETERGENTS AND SURFACTANTS. L. Raphael. *Mfg. Chemist* **39**, 41–45 (1968). A progress report covering advances in such topics as economics, nonionics, biodegradable rinse additives, synthetic fatty acids, autoxidation, odor, stability of emulsions and the question of the use of detergents to clean up pollution of the sea by marine disasters.

DETERMINATION OF ETHANOLAMIDES IN MIXTURES BY DIFFERENTIAL SAPONIFICATION RATES. F. Lohman and T. Mulligan (Procter and Gamble Co., Cincinnati, Ohio) *Anal. Chem.* **41**, 243–47 (1969). The fatty alkanolamides can be saponified in alcoholic KOH to give two moles of weak base for every mole of KOH reacting with one mole of the amides. This serves to distinguish them from amines, amine soaps, amine esters, and the ester function of amide esters and the other by-products present in the commercial alkanolamides which yield only one mole of weak base per mole of KOH consumed during saponification. This formation of extra base has been used to follow the saponification of mixtures of commercial mono- and diethanolamide and makes possible the application of the differential reaction rate technique to these systems. The pseudo-first-order rate constant for lauric diethanloamide was found to be 70 times that of lauric monoethamolamide, which condition makes the determination of these two materials by this technique inherently precise.

SYNTHESIS AND SOME SURFACE ACTIVE PROPERTIES OF FATTY DERIVATIVES OF PROPANE SULTONE. V. CELLULOSE DERIVATIVES. Hisao Hirai, Yozo Ishikawa, Kyoichi Suga and Shoji Watanabe. *Yukagaku* **17**, 623–8 (1968). Sodium sulfopropyl celluloses (Na-SPC) have been prepared from cellulose and propane sultone. The effects of concentration, temperature, pH and electrolytes on the viscosity of aqueous solutions of Na-SPC were determined as well as their compatibility with heavy duty liquid detergents. Anti-redeposition and soil removing characteristics were examined. In comparison with sodium carboxymethyl cellulose (Na-CMC), aqueous solution of Na-SPC showed lower viscosity which was rather insensitive to the concentration, temperature and pH. The synergistic effect of Na-SPC in built detergent was slightly inferior to that of Na-CMC; however, their marked solubility suggested utilization in heavy duty liquid detergents.

ALKYLBENZENE SULFONATE IN THE WATER OF TAMA RIVER. IV. DATA FROM DEC. 1966 TO DEC. 1967. Kazuaki Miura, Yoshio Suzuki, Katsuhiko Tsuchikura, Akihisa Baba, Yuzo Hayakawa, Tomoyuki Hayashi, Sadaharu Moriyama, Tetsuro Ishiguro and Yukio Yoshida. *Yukagaku* **17**, 635–7 (1968). The results showed that methylene blue active substance (MBAS) concentration has been increasing continuously. Correlations were found between MBAS and chloride ion concentration, and between MBAS and ammonia nitrogen concentration.

BIOCHEMICAL STUDIES OF N-α-OLEFIN SULFONATES. II. ACUTE TOXICITY, SKIN AND EYE IRRITATION, AND SOME OTHER PHYSIOLOGICAL PROPERTIES. Kenkichi Oba, Akira Mori and Shinichi Tomiyama (Lion Fat & Oil Co., Ltd., Tokyo). *Yukagaku* **17**, 628–34 (1968). The toxicity of n-α-C_{15-18} olefin sulfonates (AOS) was determined, including acute toxicity by various routes, rabbit eye irritation, primary skin irritation by patch test and skin mildness by repeated immersion test. A linear alkylate sulfonate and a dodecyl sulfate were also tested. AOS showed the mildest response in all tests.

SYNTHESIS AND PROPERTIES OF N-ACYL-N-ALKYL-β-ALANINES. Mutsuo Ishii, Hidemitsu Takizawa, Yasunori Usuba, Kazuo Ishikawa, Keisuke Morimoto and Hajime Akiba (Kawaken Fine Chem. Co., Kawagoe, Japan). *Yukagaku* **17**, 616–22 (1968). N-acyl-N-alkyl-β-alanines and their salts were prepared by acylating N-alkyl-β-alanines and N-alkyl-β-alanine methyl esters in water or in organic solvent in the presence of organic alkali or sodium hydroxide catalyst. The products showed excellent foaming and wetting properties even in hard water. N-methyl, N-lauroyl and myristoyl derivatives showed excellent properties in the pH 6–7 range.

BIOCHEMICAL STUDIES OF N-α-OLEFIN SULFONATES. I. BIODEGRADABILITY UNDER AEROBIC CONDITION. Kenkichi Oba, Akira Mori and Shinichi Tomiyama (Lion Fat & Oil Co., Tokyo). *Yukagaku* **17**, 517–20 (1968). The aerobic biodegradability of C_{15} alkenyl sulfonate, C_{14} hydroxy alkane sulfonate, and n-α-olefin sulfonates was measured by the Japanese Industrial Standard Method. Good reproducibility of a methylene blue colorimetric method was confirmed. The alkenylsulfonate, hydroxyalkane sulfonate and n-α-olefin sulfonate were completely biodegradable under these conditions.

PVP AS A SOIL ANTIREDEPOSITION AGENT. R. A. Grifo and R. P. Berni (GAF Corp.). *Soap Chem. Specialties* **44**(9), 48–9 (1968). Polyvinylpyrrolidone (PVP) has been tested as a soil anti-redeposition agent in launderometer tests using artificially soiled swatches (cotton and various synthetic fibers). Over the whole range of conditions tested (100–160F, 0.025 to

0.2% ingredient concentration in product, and on all fabrics) PVP gave significantly better results than the commonly used CMC. PVP is compatible with anionic and nonionic detergent formulations, has no adverse effect on their foaming characteristics and retains its soil suspending ability even in the presence of relatively high calcium ion concentration.

SOME PHYSICAL ASPECTS OF SOIL RELEASE. A MICROSCOPIC LOOK. J. H. Hoffman (Dan River Mills). *Am. Dyestuff Rept.* **57**, 992–7 (1968). Oil retention on resin-treated fabrics is primarily a surface or interface phenomenon involving capillarity and other oil to fiber attractive forces. Yarn and fabric structure which determine the number and size of the capillary spaces in a fabric, can influence soil release properties. The principle of fiber coating with hydrophilic or oleophobic substances to obtain oil release has been shown to be valid. Finishing based on this principle should be successful provided a durable coating having good adhesion to fiber surface can be achieved without sacrificing desirable fabric properties.

CMC, A VERSATILE WATER-SOLUBLE POLYMER. Y. L. Meltzer (H. Kohnstamm & Co.). *Soap Chem. Specialties* **44**(11), 72–6; **44**(12), 122–4, 135–6 (1968). Current knowledge is reviewed concerning carboxymethylcellulose as a soil suspending agent in detergent products and for other applications (food, textiles, etc.).

ADVANCES IN SAPONIFICATION, DRYING AND SOAP-FINISHING TECHNOLOGY. L. Spitz (Mazzoni S.p.A.). *Soap Chem. Specialties* **44**(10), 60–6 (1968). Commercially available processes in fatty acid neutralization, drying and finishing are reviewed and discussed, with special emphasis on proprietary equipment engineered by the Mazzoni firm.

SOILING AND SOIL REMOVAL STUDIES ON COTTON AND POLYESTER FABRICS. W. A. Reeves, J. V. Beninate, R. M. Perkins and G. L. Drake, Jr. (Southern Reg. Res. Lab., New Orleans, La.). *Am. Dyestuff Rept.* **57**, 1053–6 (1968). Results of a preliminary study are reported on the soiling and soil removal from polyester, polyester/cotton blends and cotton, portions of which fabrics were resin treated and resin-CMC treated. Reflectance measurements were taken before and after soiling and after one to five launderings to determine the degree of soiling and soil removal. Factors affecting soil and soil removal are discussed, as well as means to improve soil release and the application of some tailor-made fluorocarbons to produce finishes that resist oily stains but can be cleaned by laundering.

BUILDING FREE-FLOW PROPERTIES INTO LAS DETERGENTS. M. Mausner and E. Rainer (Witco Chemical Co.). *Soap Chem. Specialties* **44**(8), 34–7, 64–9; **44**(9), 56–8, 100–1 (1968). Detergents containing linear alkylbenzene sulfonates (LAS) have a generally higher degree of tackiness than those made with their tetrapropylene, non-biodegradable counterparts. Experiments show that bone-dry LAS has perfect free-flow characteristics, but that it develops tackiness very quickly upon picking up moisture. The effectiveness of sodium sulfosuccinate and sodium xylene sulfonate in reducing the tackiness of LAS formulations has been studied and a synergistic effect has been demonstrated for a mixture of these two additives. The mechanism by which tackiness is reduced is not yet understood. The presence of tripolyphosphate also improves the free flow properties of the detergent powder. Molecular weight of the alkylate is not a factor in tackiness, but alkylates having a high 2-phenyl content are more likely to develop tackiness. A new humidity-tackiness test is proposed as a useful tool in studies of detergent powder behavior.

ART OF CLEANING CARPETS. O. D. Hoxie (Bissell Inc.). *U.S. 3,418,243.* A dry cleaning composition suitable for cleaning carpets is composed of a finely divided inert carrier, a volatile organic solvent, a water-soluble surfactant and water. The formulation is in the form of a free-flowing powder which is only slightly moist to the touch. When applied to the carpet, the inert carrier functions as a fiber scouring medium, while the solvent attacks greases and oils and the aqueous phase serves to dissolve water soluble soil. Upon evaporation of the solvent and water, the resulting solid residue can be readily removed from the carpet by sweeping or vacuuming.

FATTY POLYAMINES AND THEIR ALKYL AND ALKOXY DERIVATIVES. E. J. Miller, Jr., D. J. Berenschot and R. L. Berger (Armour and Co.). *U.S. 3,418,374.* Fatty polyamines and derivatives are prepared by hydrogenating the cyanonitrile precursor with H_2 and Raney Ni, and then, if desired, alkylating or alkoxylating the fatty polyamine that is formed.