

• Fats and Oils

PROBLEMS ON DEEP FRYING SHORTENINGS. Umajiro Shimamura (Nippon Oils and Fats Co., Toshima, Kita-ku, Tokyo, Japan). *Yukagaku* 19, 748-56 (1970). Topics reviewed include heat stability and the effect of shortening oxidation products on the properties of precooked fatty foods.

SOME PROBLEMS IN FRYING OILS. Etsuji Yuki (Food Ind. Exptl. Sta., Hiroshima Pref., Hijiya, Hiroshima, Japan). *Yukagaku* 19, 644-54 (1970). The deterioration of oils in frying, methods of examining its deterioration, the relation of fatty acid composition to deterioration, the relation of trace components of frying oils to deterioration, and the present state of manufacture of fried foods are discussed.

OILSEED SUPPLY AT PRESENT AND FUTURE. Yoshimichi Chiba (Sumitomo Shoji Co., Mitoshirocho, Chiyoda-ku, Tokyo). *Yukagaku* 19, 524-33 (1970). In Japan per capita edible oil consumption is increasing, although it is still low (25 g in 1969). However, domestic production of oilseeds is very small. Statistical data are given on soybeans, cottonseeds, sunflower seeds, rapeseeds, copra, peanuts, corn (maize), safflower seeds and palm oil.

HYDROGENATION OF EDIBLE OILS AND FATS. Kosaku Yasuda (Nisshin Oil Mills, Shinkawa, Chuo-ku, Tokyo, Japan). *Yukagaku* 19, 541-51 (1970). Topics reviewed include reaction mechanism, selectivity, isomerization, conjugated diene formation, cyclization during hydrogenation and the occurrence of hydrogenation off-flavor in edible oils. Hydrogenation of soybean and cottonseed oils is discussed.

MODERN OIL MILLING TECHNIQUE AND APPARATUS. Seiko Ishigai (Yoshihara Oil Mill, Imazu-Masagocho, Nishinomiya, Japan). *Yukagaku* 19, 534-40 (1970). The tonnage of oilseeds processed is increasing, while the oil milling factories are decreasing in number, from 1,333 in 1961 to 581 in 1968. Factories of large capacity are increasing. Recent developments discussed include rapeseed meal for use as fodder, solvent extraction of brown rice, manufacture of cottonseed meal with low gossypol content and special continuous extraction of soybean oil. Modern equipment is shown for raw material treatment, extraction of oil and treatment of oil cake or meal.

Erratum

The wrong figure was published over the Figure 3 caption on page 342 of the July issue of *JAOCs*. The error occurred in W.D. Stigter's paper, "On a Correlation Between the Surface Chemistry and the Felting Behavior of Wool."

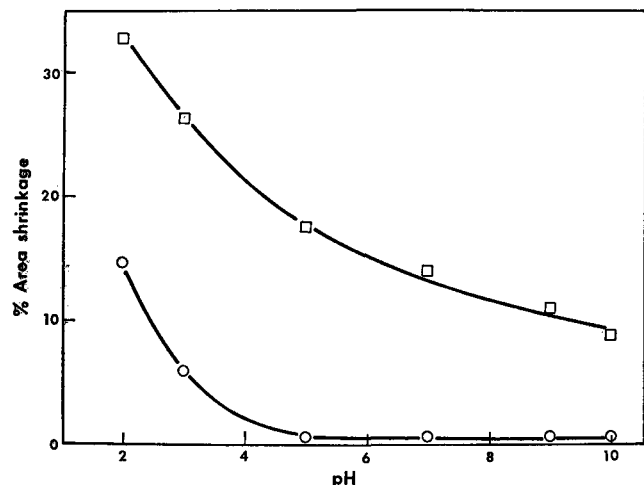


FIG. 3. Shrinkage of wool oxidized with permanganate. Squares: 3.5% permanganate-salt. Circles: 5% permanganate-salt. Data by McPhee and Feldtman (19).

PLASTICS FOR PACKAGING FATTY FOODS. Toshio Iida (Mitsubishi Petrochem. Co., Yokkaichi, Mie-ken, Japan). *Yukagaku* 19, 682-8 (1970). Such packages have suitable properties in relation to shading from light, air permeability and resistance to fats. Plastics discussed include polyethylene, polypropylene, polyvinylidene chloride, polycarbonate, polyester (from telephthalate and ethylenglycol), polystyrene and cellophane.

FOOD ADDITIVES IN THE OIL AND FAT INDUSTRY. Yoshio Yamanaka (Min. of Health and Welfare, Kasumigaseki, Chiyoda-ku, Tokyo, Japan). *Yukagaku* 19, 665-74 (1970). In Oct. 1968 toxic effect of rice oil was reported in Fukuoka Pref. It was attributed to chlorodiphenyl used as heating medium for deodorizing the oil. Additives used in fats and oils industry are classified and explained chiefly from the viewpoint of public health.

RELATION OF FATS AND OILS TO COOKERY. Fujiko Yoshimatsu (Ochanomizu Women's Univ., Otsuka, Bunkyo-ku, Tokyo, Japan). *Yukagaku* 19, 627-33 (1970). Fats and oils as additives on cooked foods, using oils for frying and the utilization of pork or beef fat in cooking are discussed.

TASTES OF FATS AND OILS. Riichiro Usuki and Takashi Kaneda (Tohoku Univ., Sendai, Japan). *Yukagaku* 19, 612-19 (1970). Topics reviewed include the taste of edible oils (liquid oils, heated oils, fats), taste of foods in relation to lipids, rheological nature of oils in relation to the taste, tastes and lipid components (aldehydes, ketones, lactones, alcohols, free fatty acids, fatty acid peroxides, hydrocarbons, sterols, tri-, di-, and monoglycerides).

X-RAY DIFFRACTION AND POLYMORPHISM OF TRIGLYCERIDES. Midori Goto (Government Chem. Ind. Res. Inst., Tokyo, Honmachi, Shibuya-ku, Tokyo, Japan). *Yukagaku* 19, 583-99 (1970). Polymorphism of triglycerides is tabulated for mp, long spacing and side spacing. In general there are 3 types α , β and β' . This phenomenon can be studied not only by X-ray diffraction, but also by other methods such as dielectric behavior, infrared and NMR spectra.

GLYCERIDE COMPOSITIONS OF NATURAL FATS. Shigeru Tsuda (Ind. Res. Inst. Osaka Pref., Enokojima, Nishi-ku, Osaka, Japan). *Yukagaku* 19, 572-6 (1970). There are to be n^3 kinds of triglycerides, when a fat contains n kinds of fatty acids. Methods of analyzing glyceride structure have progressed considerably using enzymatic, chemical and physical techniques. Several theories or hypotheses concerning fatty acid distribution in glycerides are discussed.

CATALYSTS FOR SELECTIVE HYDROGENATION OF FATTY OILS. Ichigi Nakamori (Nikki Chem. Co., Otemachi, Chiyoda-ku, Tokyo, Japan). *Yukagaku* 19, 556-61 (1970). Topics reviewed include Ni, Raney Ni, noble metals (Pt, Pd, Rh, Lu), and Cu as catalysts from practical viewpoint.

NATURAL ANTIOXIDANTS (FOR EDIBLE OILS). Shinroku Masuyama (Osaka Munic. Tech. Res. Inst., Kita-Ogimachi, Kita-ku, Osaka, Japan). *Yukagaku* 19, 675-81 (1970). Topics reviewed include tocopherols, NDGA (nordihydroguaiaretic acid), flavone derivatives, gallate derivatives, caffeate derivatives, gossypol, sesamol and natural spices.

COLOR REVERSION OF EDIBLE OILS. Ichiro Harada and Mamoru Komoda (Sugiyama Chem. Res. Inst., Inokashira, Mitaka, Tokyo, Japan). *Yukagaku* 19, 655-64 (1970). This review concerns soybean oil chiefly. Effects on color reversion of trace substances in the oil and of moisture of raw soybeans are discussed. Among trace substances tocopherol is important in relation to color reversion.

STABILITY OF EDIBLE FATS AND OILS. Shizuyuki Ota (Ajinomoto Co., Suzukicho, Kawasaki, Kanagawa-ken, Japan). *Yukagaku* 19, 634-43 (1970). Topics reviewed relate to frying oils (deterioration, relating factors, methods for its prevention) and the stability during storage at room temperature.

FLAVOR COMPOUNDS IN OILS AND FATS. Akio Kato (Govt. Chem. Ind. Res. Inst., Tokyo, Honmachi, Shibuya-ku, Tokyo, Japan). *Yukagaku* 19, 620-6 (1970). Topics reviewed include characteristic flavor of oils, secondary flavors formed by oxidative degradation of fat components and isolation and characterization of flavor compounds.

ABSTRACTS: FATS AND OILS

CRYSTAL GROWTH AND PHYSICAL PROPERTIES OF OILS AND FATS BY THE USE OF ELECTRON MICROSCOPY. Masakazu Okada (Snow Brand Milk Prod. Co., Akabane-Kita, Kita-ku, Tokyo, Japan). *Yukagaku* 19, 600-11 (1970). The main components of margarine, shortening and butter are crystalline triglycerides. Rheological properties and crystal growth are discussed.

SOLVENT FRACTIONATION (OF EDIBLE FATS). Toshimi Akiya (Food Res. Inst., Shiohama, Koto-ku, Tokyo, Japan). *Yukagaku* 19, 577-82 (1970). Topics reviewed include winterization of edible oil in hexane-acetone, fractionation of glycerides with a solvent (e.g. propyl acetate), preparation of imitation cacao butter from various fats and oil with acetone, application for dewaxing of rice bran oil, and detection of residual solvents.

RECENT TECHNOLOGY OF HYDROGENATING EDIBLE FATS AND OILS. Kazutomo Maebashi (Asahi Electro Chem. Co., Higashi-Oku, Arakawa-ku, Tokyo, Japan). *Yukagaku* 19, 552-5 (1970). Topics reviewed include processing before and after hydrogenation, selective hydrogenation of trienoic acids, isomerization and the production systems.

CURRENT SITUATION OF OILSEED PROCESSING INDUSTRY IN JAPAN AND TRADE LIBERALIZATION. Hiroshi Higashimori (Japan Oilseed Processors Assoc., Edobashi, Nihonbashi, Chuo-ku, Tokyo, Japan). *Yukagaku* 19, 518-23 (1970). Japan has very small domestic production of oilseeds. In 1969 it was only 461,277 tons (425,409 t rice bran, and 32,185 t rapeseed, etc.). Imported oilseeds amounted to 3,600,576 t: 1,974,637 t soybeans (chiefly from U.S.), 367,486 t rapeseed, 257,625 t cottonseed, 123,547 t linseed, 114,745 t copra, etc. Thus trade liberalization planned before the end of 1971 poses a great problem for Japanese oil makers.

NATURAL AND SIMULATED CACAO BUTTER. Yoshitsugu Nakanishi (Government Ind. Res. Inst. Osaka, Ikeda, Osaka, Japan). *Yukagaku* 19, 722-33 (1970). Topics reviewed include general properties of cacao butter (cb), m.p. of cb, influence of additives on the solidification of cb, simulated cb and mixing of natural and simulated cb.

MILK FAT—BUTTER OIL. Fumiyasu Tsuchiya (Meiji Milk Products Co., Higashi-Murayama, Tokyo, Japan). *Yukagaku* 19, 757-64 (1970). Production of milk fat and the preparation of butter oil and ghee are reviewed. Characteristics, components, state of existence and flavor of milk fat are discussed.

PROBLEMS IN BAKERY AND CONFECTIONER'S FATS. Nobuya Matsui (Kao Soap Co., Bunka, Sumida-ku, Tokyo, Japan). *Yukagaku* 19, 734-47 (1970). Topics reviewed include classification and characteristics of fats for bakery and confectionary, and such products as icings, butter cream, chocolate coating, biscuits, crackers, bread, cakes and prepared mixes.

A RAPID METHOD FOR EVALUATION OF AUTOXIDATION AND ANTI-OXIDANTS. Kazufumi Yagi (Kobe Coll., Nishinomiya, Hyogo-ken, Japan). *Agr. Biol. Chem.* (Tokyo) 34(1), 142-5 (1970). This paper describes a new device for measuring antioxidant activity in a given system containing autoxidizable food components. Oxidation of ascorbic acid, reductones or unsaturated lipids was recorded as the decrease in dissolved molecular oxygen in the system tested and the measurement could be completed within several min. by use of metal catalyst. Changes in oxygen content were measured with a polarographic oxygen analyzer connected to a recorder. Safflower oil oxidized to different peroxide values was used in these tests.

PRODUCTION OF FATTY ACIDS FROM OIL FOOTS. Kazuhiko Yoshitomi (Nisshin Oil Mills, Yokohama, Japan). *Yukagaku* 19, 807-18 (1970). A review with 59 references. There are two kinds of "oil foots:" that produced as by-products of degumming of crude oil and that produced on purification with alkali. Fatty acids are obtained from both "foots" by hydrolysis. Distillation and fractionation are discussed.

EDIBLE OILS DESCRIBED IN PATENTS. Tamihei Nagasaki and Yutaka Wada (Japanese Patent Office, Kasumigaseki, Chiyoda-ku, Tokyo, Japan). *Yukagaku* 19, 781-91 (1970). Japanese patents on edible oils are discussed under headings of preparation, purification, additives, and processings (212 references).

MAYONNAISE. Hirotake Watanabe (Kibun Co., Tsukiji, Chuo-ku, Tokyo, Japan). *Yukagaku* 19, 765-70 (1970). Topics reviewed include history, components (oil, egg yolk, vinegar and spice), production, and quality control of mayonnaise.

UTILIZATION OF DEODORIZER CONCENTRATES (FROM VEGETABLE OILS). Shizuyuki Ota (Ajinomoto Co., Kawasaki, Kanagawa-ken, Japan). *Yukagaku* 19, 835-40 (1970). Distillates obtained from vegetable oils by the deodorization process contain sterols, tocopherols, etc. This review describes chiefly the utilization of the sterol fractions.

UTILIZATION OF DEODORIZER SCUM FROM VEGETABLE OILS. Takahiro Takeuchi (Pref. Ind. Res. Inst., Kobe, Japan). *Yukagaku* 19, 819-25 (1970). This by-product contains high concentrations of tocopherols. These are extracted, concentrated and utilized commercially. Tocopherols are chiefly used as antioxidants.

LECITHIN. Shizuyuki Ota (Ajinomoto Co., Kawasaki, Kanagawa-ken, Japan). *Yukagaku* 19, 792-806 (1970). Topics reviewed include production and purification of lecithin from soybeans (in detail) and other sources (maize, peanuts, and rapeseeds), properties of soybean lecithin, and its uses in margarine, shortening, chocolate, caramel, ice cream, oblate, cakes, bread, macaroni, marine paste products (fish pastes), miso, shoyu, edible oils and nonfood products. Powdered and granular lecithins are also produced.

MARGARINE AND SHORTENING FOR CAKES AND BREADS. Kyoichi Ushijima (Kanegafuchi Chem. Ind. Co., Nakanoshima, Osaka, Japan). *Yukagaku* 19, 771-80 (1970). Topics reviewed include history and properties of margarine and shortening and their uses in bread, cakes, hiratio-cakes, cookies, and icing creams.

VOLATILE CARBONYL COMPOUNDS FROM HEATED BEEF FAT. Takeshi Yamato, Tadao Kurato, Hiromichi Kato and Masao Fujimaki (Univ., Tokyo). *Agr. Biol. Chem.* (Tokyo) 34(1), 88-94 (1970). Beef fat was heated at 150C in N₂. Ethanal, propanal, isobutanal, crotonal, benzaldehyde, acetone, methyl ethyl ketone, methyl isobutyl ketone, glyoxal and pyruvaldehyde were isolated as their 2,4-dinitrophenylhydrazones and identified by thin-layer chromatography, infrared, mass and proton magnetic resonance spectrometries. These carbonyls were assumed to be formed also on heating in air. When beef fat was heated at 200C in air, hexanal, heptanal, 2-heptenal, octanal, 2-octenal, nonanal, 2-nonenal, 2-decenal, 2,4-decadienal, and 2-undecenal were identified by gas-liquid chromatography.

THE CHEMICAL STRUCTURE OF GLYCOLIPIDS OF BOVINE MILK. Yasuhiko Fujino, Masao Nakano and Toru Saeki (Obihiro Zotech. Univ., Obihiro, Hokkaido). *Agr. Biol. Chem.* (Tokyo) 34(3), 442-7 (1970). Ceramide monohexoside isolated from cow milk was shown to be β -glucosyl-(1 \rightarrow 1)-N-acylsphingosine or ceramide glucoside, while ceramide dihexoside was β -galactosyl-(1 \rightarrow 4)- β -glucosyl-(1 \rightarrow 1)-N-acylsphingosine or ceramide lactoside. Infrared spectra of both glycolipids are shown.

ANTIOXIDANT EFFECT OF TOCOPHEROLS ON MILK FAT. I. ANTI-OXIDANT ACTIVITY OF TOCOPHEROLS IN FATTY ACID METHYL ESTERS OF MILK FAT. Chouemon Kanno, Mineyuki Hayashi, Kunio Yamauchi and Tomokichi Tsugo (Univ. Tokyo, Japan). *Agr. Biol. Chem.* (Tokyo) 34(6), 878-85 (1970). Antioxidant activity of α -, β -, δ -, and γ -tocopherols was studied with fatty acid methyl esters of milk fat from which unsaponifiable matter had been removed. Autoxidation was carried out at 50C and its degree was shown by peroxide value. α - or β -Tocopherol were more effective at lower concentrations (0.003 and 0.01%) than at higher concentrations (0.05, 0.1, and 0.5%). The antioxidant activity of γ - and δ -tocopherol increased with the increase of concentration within 0.001 and 0.5%. The order of antioxidant activity varied with concentration: $\gamma > \beta > \delta > \alpha$ at 0.001%, $\alpha > \gamma > \beta > \delta$ at 0.003%, $\gamma > \delta > \beta > \alpha$ at 0.01%, and $\delta > \gamma > \beta > \alpha$ at 0.05% and higher. α -Tocopherol at 0.003%, corresponding to the concentration in original milk fat, was more effective than other tocopherols at the same concentration and α -tocopherol at other concentrations.

II. ANTI-OXIDANT ACTIVITY OF TOCOPHEROLS IN THE CHURNED AND SOLVENT-EXTRACTED MILK FAT. Chouemon Kanno, Kunio

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Yamauchi, and Tomokiehi Tsugo (Univ. Tokyo, Japan). *Ibid.*, 886-90. Milk fat prepared from the same lot of cream by churning and by solvent extraction was used as the substrate for studying the antioxidant activity of added tocopherols. The churned milk fat was more quickly autoxidized than the solvent-extracted one at 60 C. α -Tocopherol added to the churned milk fat acted as pro-oxidant. The antioxidant activity of α -tocopherol added to the solvent-extracted milk fat was higher at 0.01% than at 0.05 and 0.1%. Addition of γ - and δ -tocopherols inhibited autoxidation of both types of milk fat.

LIPIDS OF SCALE INSECTS. XIV. COMPOSITION OF THE TRIGLYCERIDES OF 4 SCALE INSECTS. Akira Hashimoto, Akio Hirofumi, Katsunori Mukai and Shozaburo Kitaoka (Univ. Osaka Pref., Sakai, Osaka). *Agr. Biol. Chem.* (Tokyo) 34(12), 1839-42 (1970). Fatty acid composition of triglycerides of *Lecanium corni*, *Pulvinaria* sp., *Cerococcus muratae* and *Phenacoccus pergandei*, respectively, were: C-8:0 0.5, —, 17.1, —; C-10:0 34.9, 28.5, 22.5, 0.6; C-12:0 50.7, 33.8, 34.4, 96.8; C-14:0 11.1, 0.7, 15.2, 2.5; C-16:0 0.8, 37.0, 3.3, 0.1; C-16:1 —, —, 2.2, trace; C-18:0 trace, —, 1.1, —; C-18:1 0.4, —, 4.2, —; C-18:2 1.6, —, —, —. Glyceride structures were also deduced by determining the fatty acid composition at the 2-position.

INACTIVATION OF ENZYMES BY LINOLEIC ACID HYDROPEROXIDES AND LINOLEIC ACID. Setsuro Matsushita, Masayo Kobayashi and Yuki Nitta (Kyoto Univ., Kyoto, Japan). *Agr. Biol. Chem.* (Tokyo) 34(6), 817-24 (1970). This problem was examined in relation to the toxicity of oxidized fat. Ribonuclease was largely inhibited by linoleic acid hydroperoxides, while trypsin, chymotrypsin, and pepsin were inhibited by linoleic acid and its hydroperoxides.

EFFECT OF LINOLEIC ACID HYDROPEROXIDES ON PEPSIN ACTIVITY. Setsuro Matsushita and Masayo Kobayashi. *Ibid.*, 825-9. This effect differed considerably at different pH values. Pepsin was activated by incubation with linoleic acid hydroperoxides in acidic region except around pH 4. The activity was remarkable at pH 5-6. It is still uncertain whether the hydroperoxides were combined with the protein molecule or hydroperoxide group reacted with some special amino acid residues in the protein molecule. The reaction was very sensitive to pH change, suggesting some conformational change of the pepsin molecule.

COMPOSITION OF LIPID FOAMS FROM SWIM BLADDERS OF TWO DEEP OCEAN FISH SPECIES. S. Patton and A. J. Thomas (Lipids Lab., Pennsylvania State Univ., University Park, Penn. 16802). *J. Lipid Res.* 12, 331-35 (1971). Lipid-containing deposits within the swim bladders of *Coryphaenoides acrolepis* and *Antimora rostrata* were investigated. Lipid analysis of this material which was quite uniform from the two species, yielded the following data; neutral lipids, 36.0-41.7%; phospholipids, 53.6%-56.7%; and glycolipids, 4.3-8.9%. Cholesterol (mainly in the free form) constituted 60.4%-77.8% of the neutral lipids. Sphingomyelin and phosphatidylcholine were the principal phospholipids, with sphingomyelin highest in the material from *C. acrolepis* and phosphatidylcholine predominant in that from *A. rostrata*. The overall pattern of lipids shows a resemblance to that of plasma membrane, particularly in the relatively high levels of free cholesterol, sphingomyelin and phosphatidylserine. The lipid-to-protein ratio of the material is approximately 1.5-2 to 1. The lipids of the fine inner lining (tunica interna) of the swim bladder from a shallow water fish, the kelp bass (*Paralabrax clathratus*), had essentially the same composition as the much more abundant swim bladder material from the deep ocean fishes.

NEW SYNTHETIC PHOSPHINATE ANALOGUES OF LECITHIN. A. F. Rosenthal and S. V. Chodsky (Dept. of Labs., Long Island Jewish Med. Cent., New Hyde Park, N.Y. 11040). *J. Lipid Res.* 12, 277-85 (1971). The chemical syntheses of two new, completely nonhydrolyzable phosphinate analogues of lecithin are described. These have the structures $\text{ROCH}_2\text{CH}(\text{OR})\text{-CH}_2\text{CH}_2\text{-P}(\text{O})(\text{O}^-)\text{CH}_2\text{CH}_2\text{N}^+(\text{CH}_3)_3$ and $\text{ROCH}_2\text{CH}(\text{OR}')\text{CH}_2\text{P}(\text{O})(\text{O}^-)\text{CH}_2\text{CH}_2\text{N}^+(\text{CH}_3)_3$, where $\text{R} = \text{C}_{15}\text{H}_{37}$ and $\text{R}' = \text{C}_6\text{H}_{13}$. Each is thus isosteric with lecithin on either side of the phosphorus function. The infrared spectra of these compounds undergo unexpected changes under mild acid, base or adsorptive treatment. These are discussed and compared with related lecithin analogues, including the simple phosphinate

$\text{C}_{15}\text{H}_{37}\text{P}(\text{O})(\text{O}^-)\text{CH}_2\text{CH}_2\text{N}^+(\text{CH}_3)_3$, whose synthesis is also reported.

FREEZING METHODS INFLUENCE ON FAT AND MOISTURE COMPOSITION OF PRECOOKED THIGHS. L. D. Yingst and T. L. Goodwin (Dept. of Animal Sciences, Univ. of Arkansas, Fayetteville, Ark. 72701). *Poultry Sci.* 50, 957-59 (1971). Commercially breaded, precooked, frozen thighs were obtained from a local processor and evaluated for chemical composition and tenderness. The chicken parts were frozen by each of the following methods: liquid dichlorodifluoromethane, liquid dichlorodifluoromethane plus 5% chicken fat and conventional blast freezing. The muscle tissue of the precooked thighs contained more extractable fat prior to deep-fat frying than did comparable parts after being deep-fried. The blast frozen muscle tissue was lower in fat content than the muscle tissue frozen by the dichlorodifluoromethane methods. The addition of 5% chicken fat to the dichlorodifluoromethane gave a skin-breading complex which was considerably higher in fat content. Deep fat frying of the frozen products resulted in desiccation of both muscle tissue and the skin-breading complex. Treatments had no effect on the tenderness of the thighs.

MASS SPECTRAL ANALYSIS OF GLYCEROPHOSPHOLIPIDS. J. H. Duncan, W. J. Lennarz and Catherine C. Fenselau. *Biochemistry* 10, 927-31 (1971). Mass spectrometry and combined gas chromatography-mass spectrometry have been applied to the determination of the "backbone" components of glycerophospholipids. By chemical modification of the parent phospholipids, the characteristic glycerophosphate esters have been isolated and analyzed as their trimethylsilyl derivatives.

DETERMINATION OF EPOXIDE POSITION AND CONFIGURATION AT THE MICROGRAM LEVEL AND RECOGNITION OF EPOXIDES BY REACTION THIN-LAYER CHROMATOGRAPHY. B.A. Bierl, M. Beroza, Mary H. Aldridge (Chem. Dept., the Amer. Univ., Washington, D.C. 20016 and U.S. Dept. of Agri, Agri. Res. Service, Beltsville, Md. 20705). *Anal. Chem.* 43, 636-645 (1971). The position of epoxide groups in microgram amounts of compound is located by reacting the compound in a halogenated solvent with dry, powdered HIO₄ for 5 minutes; the aldehyde and ketone fragments produced by cleavage between the carbon atoms of the epoxide group are then determined by gas chromatography of the reaction solution. Epoxides are detected by applying them over a phosphoric acid spot on a silica gel thin-layer chromatographic plate and allowing the reaction to proceed for 1 hour before plate development; the products of the epoxide, being much more polar than the original compound, remain near the origin, and the original spot is no longer visible. The geometrical configuration of disubstituted epoxides is determined by the same procedure, except that a 5-minute reaction interval is used before plate development. Cis epoxides react almost completely within 5 minutes; steric hindrance (α -alkyl substituent) delays the reaction some, and trans configuration delays the reaction even more. The procedures, which were applied to a variety of epoxides and compounds with other oxygen-containing functional groups, are rapidly and easily carried out.

SULFOLIPID I OF MYCOBACTERIUM TUBERCULOSIS, STRAIN H37RV. NATURE OF THE ACYL SUBSTITUENTS. M. B. Goren, Olga Brokl, B. C. Das and E. Lederer (Div. of Res., Nat. Jewish Hos. and Res. Center, Denver, Colo., and Inst. de Chimie des Sub. Naturelles, Gif-sur-Yvette, France). *Biochemistry* 10, 72-81 (1971). Sulfolipid I of *Mycobacterium tuberculosis*, strain H37Rv, was previously characterized as a 2,3,6,6'-tetraacyl-trehalose 2'-sulfate. The structures of the acyl functions have been elucidated largely by mass spectrometry and are reported herein. Three principal (and related) series of carboxylic acids were found: palmitic-stearic acids with minor

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amounts of other homologs; a multibranched series, the "average" member of which is 2,4,6,8,10,12,14-heptamethyltriacontanoic acid; and a second, related, oxygenated multibranched acid consisting principally of 17-hydroxy-2,4,6,8,10,12,14,16-octamethyltriacontanoic acid. Homology by 42 mass units is prominent in both series and suggests a biogenesis involving successive incorporations of propionate onto a palmitate residue. All representatives of the two methyl-branched series are dextrorotatory; they are therefore very likely of the L configuration and related to the phtienic (mucolipenic) acids.

COULOMETRIC DETERMINATION OF OIL ACIDITY. M. Kucera and K. Novak. *Chem. Průmysl* 21, No. 1, 27-9 (1971). A coulometric titration method was elaborated, suitable for the determination of oil acidity. The method allows the determination of very low acidities with minimum consumption of the sample. The titration is carried out in 2-propanol. A Pt cathode is used for generation of the base (tetrabutylammonium hydroxide) and a Ag electrode serves as a reference anode. (World Surface Coatings Abs. No. 348)

EFFECT OF TEMPERATURE AND HYDROCARBON CHAIN STRUCTURES ON THE TITRATION PROPERTIES OF LECITHIN AND SOME RELATED PHOSPHOLIPIDS. M. B. Abramson. *J. Colloid Interface Sci.* 34, No. 4, 571-9 (1970). The titration characteristics of lecithin and phosphatidyl ethanolamine are changed when the lipid in water undergoes a mesomorphic transition. The temp. at which this transition occurs depends on the chain length and unsaturation of the hydrocarbon groups. (World Surface Coatings Abs. No. 348)

VARIATION IN THE FATTY ACID DISTRIBUTION OF FILLED MILK BEVERAGES. R.A. Horvath, W.H. Brown and J.W. Stull (Dept. of Dairy and Food Sci., The Univ. of Arizona, Tucson, Ari. 85721). *Am. J. Clin. Nutr.* 24, 397-400 (1971). Samples of eight filled milk beverages and four liquid coffee whiteners were collected on or about the first day of 6 consecutive months. Fatty acid distributions were determined by gas-liquid chromatography, and percentages of total solids and fat were measured by standard methods. There were two basic types of fatty acid distributions. One was characterized by predominantly lauric, myristic, palmitic and stearic acids with a total saturated fatty acid content of more than 90%. The second contained predominantly oleic, linoleic, palmitic and stearic acids with a total unsaturated fatty acid content of more than 75%. There was considerable variation in total solids and fat content.

CHEMICAL COMPOSITION OF AN OESTROGEN-INDUCED CALCIUM-BINDING GLYCOLIPOPHOSPHOPROTEIN IN *XENOPUS LAEVIS*. A.Q. Ansari, P.J. Dolphin, C.B. Lazier, K.A. Munday and M. Akhtar (Dept. of Physiol. and Biochem., Univ. of Southampton, Southampton S09 5NH, U.K.). *Biochem. J.*, 122, 107-13 (1971). Oestrogen treatment has previously been shown to induce the formation of large amounts of a serum protein, vitellogenin (xenoprotein), in *Xenopus laevis*. Vitellogenin was purified from serum by dimethylformamide precipitation and was shown to be homogeneous by a variety of electrophoretic techniques. The molecular weight of vitellogenin was estimated by gel filtration to be about 6×10^6 . The chemical constituents of vitellogenin were determined and lead to the characterization of this protein as a serum calcium-binding glycolipophosphoprotein. The extractable lipid accounted for 12% of vitellogenin. Gas-liquid-chromatographic analysis of the saponified lipid moiety showed the presence of palmitic acid, palmitoleic acid, stearic acid, oleic acid and linoleic acid in the molecular proportions 6.8 : 1.5 : 1.0 : 3.6 : 1.4.

AN ANAEROBIC REACTION BETWEEN LIPOXYGENASE, LINOLEIC ACID AND ITS HYDROPEROXIDES. G.J. Garssen, J.F.G. Vliegert-hart and J. Boldingh (Lab. of Org. Chem., State Univ. of Utrecht, Croesestraat 79, Utrecht, The Netherlands). *Biochem. J.* 122, 327-32 (1971). In an anaerobic system soya bean lipoxygenase together with linoleic acid induces a structural rearrangement of 13-hydroperoxyoctadeca-*cis*-9-*trans*-11-dienoic acid leading to the formation of 13-oxotrideca-*cis*(*trans*)-9-*trans*-11-dienoic acid and n-pentane as well as 13-oxo-octadeca-9,11-dienoic acid. It is proposed that the 13-peroxyoctadeca-*cis*-9-*trans*-11-dienoic acid radical formed through hydrogen radical abstraction by the linoleic acid radical is the key intermediate for three reactions.

FATTY ACID ETHYL ESTERS OF RHIZOPUS ARRHIZUS. J.L. Laseter and J.D. Weete (Dept. of Biol. Sci., Louisiana St. Univ., New

Orleans, La. 70122). *Science* 172, 864-65 (1971). Gas chromatographic and mass spectrometric analyses on selected lipid fractions revealed for the first time the presence of ethyl esters of long-chain fatty acids as biological products. Ethyl esters of oleic, palmitic and stearic acids were detected in relative concentrations of 21.2, 2.4, and 1.5%, respectively, of the total methyl and ethyl ester fraction.

ELECTRIC CHARGE SPEEDS WASTE DISPOSAL. O.A. Clemens (Gen. Mgr., Eng. Res. Dept., Swift and Co., Oakbrook, Ill.) and J.V. Ziemba. *Food Eng.* 43(8), 47-9 (1971). Chemical flocculation combined with an impressed electric current aids in the liquid-waste treatment from an edible oil refinery. The flocculants used in this case are alum and a specially formulated polymer. BOD, fats and suspended solids are reduced to acceptable levels for discharge into a municipal sewerage system.

PREDICTS OIL STABILITY. V. Spiehler. (Associate Application Chem., Application Res., Beckman Instruments, Inc., Fullerton, Calif.). *Food Eng.* 43(8), 77-8 (1971). A recently introduced oxygen analyzer permits detection of dissolved oxygen in oil. A polarographic sensor measures partial pressure of oxygen in physical solution and is not influenced by chemically bound peroxide compounds. Dissolved oxygen in oil gives a simple indication of oil stability and a guide to antioxidant selection. The instrument also has a number of other applications.

ALASKA KING CRAB: FATTY ACID COMPOSITION, CAROTENOID INDEX AND PROXIMATE ANALYSIS. R.A. Krzeczowski, R.D. Tenney and C. Kelley (USDC National Oceanic and Atmospheric Adm., National Marine Fisheries Service, Marine Fisheries Center, Box 1638, Kodiak, Alaska 99615). *J. Food Sci.* 36, 604-6 (1971). The proximate analysis, carotenoid index and fatty acid composition of the lipids were determined on five separate types of cooked and frozen king crab meat. Similar fatty acid content was found in all types of meat. Eight fatty acids accounted for about 77% of the total fatty acid content; 26 others were found in low quantities. Fatty acid 20:5 was predominant followed by 18:1 and 22:6. Polyunsaturated acids were predominant (50.2-56.4%) in all types of meat followed by 29.8-33.5% mono-unsaturated acids and 14.6-17.0% saturated acids. The proximate analysis varied slightly in some types of meat with 16.3-20.7% protein, 1.3-1.8% ash, 0.9-3.3% lipid and 76.2-79.6% moisture. The carotenoid content was highest in the propodus-carpus meat and lowest in the shoulder meat. The skin contained most of the lipid and carotenoids.

FOOD PRODUCTS FROM CORN GERM: ENZYME ACTIVITY AND OIL STABILITY. H.W. Gardner and G.E. Inglett (USDA NRRL, ARS, Peoria, Ill. 61604). *J. Food Sci.* 36, 645-48 (1971). Various temperatures were used during roll-cooking to inactivate the lipase, lipoxygenase, linoleic acid-hydroperoxide-isomerase and peroxidase in full-fat corn germ. Due to the heat inactivation of lipase, the oil of roll-cooked germ was stable to lipolysis during storage, except at moistures sufficient to support mold growth. Peroxide values of the extracted oil increased during storage only in samples in which linoleic acid-hydroperoxide-isomerase was inactivated.

MYCOTOXINS OTHER THAN AFLATOXIN IN OILSEEDS. M.T. Juillet (ITERG, Paris.) *Rev. Franc. Corps Gras* 18, 301-7

SOS/70 Proceedings Available

Proceedings of the Third International Congress on Food Science and Technology, "Science of Survival" (SOS/70), will be available for distribution by November 1 from the Institute of Food Technologists, host of the August, 1970, gathering of world wide food scientists in Washington, D.C.

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(1971). Many fungi can grow and produce potentially dangerous toxins on oilseeds. Structural formulas where available and LD₅₀ data are listed for a number of toxins produced by the following genera of molds: *Aspergillus*, *Fusarium*, *Penicillium*, *Rhizopus* and *Claviceps*. During storage and handling prior to use, proper precautions should be taken to prevent contamination and growth of molds on the seeds.

DETERMINATION OF INSECTICIDE RESIDUES IN FATS AND OILS. A. Karleskind and J.P. Wolff (Wolff Labs., Paris). *Rev. Franc. Corps Gras* 18, 285-95 (1971). Of the various methods used for the fractionation and analysis of chlorinated pesticides, Storherr's method (*JAOC* 50, 605-15 (1967)) was found to be the best for continuous control purposes. It was possible to determine 0.01 ppm of isomers of HCH, aldrin, heptachlor, heptachlor oxide, and 0.02 ppm of dieldrin and DDT. Using this method, the amounts of pesticides in various animal fats and vegetable oils were determined. Also studied was the effect of refining on the pesticide level. The pesticides were virtually completely removed in well-refined oils.

LIQUID CAKE SHORTENING CONTAINING ALKYL GALLATES. D.H. Hughes (Procter and Gamble). *U.S. 3,597,225*. Alkyl gallates are used as high temperature batter stabilizers in combination with liquid glyceride oils and alpha-phase crystal tending emulsifiers as ingredients in cake mixes to provide cakes of improved volume, texture and eating quality.

LIQUID CAKE SHORTENING CONTAINING KOJYL ACYLATE. D.H. Hughes (Procter and Gamble). *U.S. 3,597,226*. Kojyl acylate used in liquid cake shortenings provides benefits similar to those described in U.S. 3,597,225.

BUTTERLIKE MARGARINE. A. Mijnders and H.W. Lincklaen (Lever Bros.). *U.S. 3,579,229*. A margarine having butter-like properties is prepared from a fat blend comprising an olein fraction, melting at 30-36C, obtained from a rapidly crystallizing hydrogenated fat.

STABLE BETA PRIME PLASTIC SHORTENING. E.E. Colby and C.H. Japikse (Procter and Gamble). *U.S. 3,597,230*. A hardstock composition comprising two components combined in certain proportions provides improved beta prime phase stability. The hardstock components are a beta prime-tending C₁₄-C₂₀ hardstock containing at least a minimum amount of C₂₀-C₂₂ acids and a beta prime-tending C₁₄-C₁₈ hardstock containing at least a minimum amount of C₁₀-C₁₈ fatty acids.

MARGARINE OIL. H.W.L. Westenberg (Lever Bros.). *U.S. 3,600,195*. The oil contains specified proportions of an interesterified mixture of a liquid fat, containing at least 40% polyunsaturated fatty acids, and a hard fat melting at 42-48C and containing at least 10% C₂₀-C₂₂ acids. The remainder of the fat may consist of fats hardened to 32-38C with at least 10% of other fats hardened to 42-48C. This mixture may also be interesterified.

NOVEL CREAM PRODUCTS. C. Heine and C. von Schilcher (Henkel & Cie). *U.S. 3,600,196*. Cream products comprising 50-70% of shortening with 10-15% solids at 20C and 5-10% solids at 30C, 20-30% sugar, and up to 30% of other powdered ingredients is described. From 20-150 cc of inert gas are dispersed through 100 g of product.

FLAVOR ENHANCING COMPOSITIONS FOR FOODS AND BEVERAGES. H.D. Spangler, P.A. Hammes and C.W. Everson (Merck & Co.). *U.S. 3,600,197*. The composition prevents stratification and segregation of ingredients without affecting flowability. It consists of a premix of a salt with a blend of (a) a glycerol or propylene glycol monoester or acetylated monoester of C₁₂-C₁₈ fatty acids, and (b) a mixture of sodium glutamate with 5'-nucleotides.

• Fatty Acid Derivatives

CONVERSION OF SOME SATURATED FATTY ACIDS, ALDEHYDES AND ALCOHOLS INTO γ - AND δ -LACTONES. Kenji Watanabe and Yasushi Sato (Nagoya Univ., Nagoya, Japan). *Agr. Biol. Chem.* (Tokyo) 34(3), 464-72 (1970). A series of lactones were found in the thermally oxidized products of normal saturated acids, aldehydes and alcohols of C₆, C₁₀ and C₁₂, when heated at 180C in the presence of 0.1% KMnO₄. They were identified by gas chromatography, infrared spectroscopy, and mass spectroscopy. They could be detected also in the volatile components produced by heating C:10 acid, aldehyde and alcohol mixed with pork fat. Thus lactones in meat fat

flavor might be secondary products converted from saturated acids, aldehydes and alcohols formed by oxidative degradation of meat fats. The lactones might be derived through mono- or dihydroperoxides of acids, aldehydes, and alcohols.

ESTERIFICATION AND INTERESTERIFICATION (OF EDIBLE OILS). Kazuo Sakurai and Kiyota Murakawa (Riken Vitamin Oil Co., Nishi-Kanda, Chiyoda-ku, Tokyo, Japan). *Yukagaku* 19, 562-71 (1970). Recent reports are systematically reviewed. Gas chromatograms are shown for Span 40, 60, and 80, and commercial polyglycerol esters. Special derivatives of mono-glyceride lactate and citrate, etc. are mentioned. Acetolysis, alcoholysis, ester interchange are discussed under interesterification.

LOW CALORIE FAT-CONTAINING COMPOSITIONS. F.H. Mattson and R.A. Volpenhein (Procter and Gamble). *U.S. 3,600,186*. Part of the fat content of a conventional food is replaced with a sugar fatty acid ester or sugar alcohol ester having at least 4 ester groups, with each fatty acid having from 8 to 22 carbon atoms.

• Biochemistry and Nutrition

IS FAT-CORRECTED MILK SUFFICIENT? R.A. Brog (Dairy Consultant, Dairy Monitoring of America, North Logan, Utah 84321). *J. Dairy Sci.* 54, 1137-41 (1971). Since milk plant administrators are investigating protein as a milk accounting and pricing variable, a study determined whether the fat content of milk and dairy products with the weight, were sound criteria for estimating their respective values in the market place. Clearly the fat content and weight of milks and milk products failed to describe their respective wholesale prices. An alternative approach for evaluating milk from lactating dairy cows is suggested. This approach reduces milk equivalents, based on protein and fat contents, to dollars for a hypothetical milk arbitrarily valued at \$5 per hundred-weight. Several advantages inherent in the suggested milk evaluation system are discussed including benefits which could accrue to milk producers and milk consumers.

LIPIDS AND SERUM CHOLESTEROL. Teruo Ono and Yoh Imai (Hokkaido Univ., Sapporo, Japan). *Yukagaku* 19, 705-12 (1970). First the state of cholesterol is discussed in relation to absorption, metabolism in the liver, and content in serum. Second, the effect of diet on serum cholesterol is discussed chiefly in relation to atherosclerosis.

FAT METABOLISM IN OBESITY. Shigeaki Baba (Kobe Univ., School Med., Ikuta-ku, Kobe, Japan). *Yukagaku* 19, 689-94 (1970). Topics reviewed include regulation of lipid metabolism, metabolic features of obesity (such as rise in blood insulin, decrease in oxidation of glucose, palmitate, and β -hydroxybutyrate, and rise in blood fatty acids) and mechanism of production of obesity.

TOXICITY OF OXIDIZED AND HEATED FATS. Kagenori Matano (Natl. Inst. Health, Shinagawa-ku, Tokyo, Japan). *Yukagaku* 19, 713-21 (1970). Topics reviewed include the toxicity of heated oils and autoxidized oils (especially long-term experiments with monkeys) and deterioration and toxicity of fatty foods (instant foods).

LIPIDS FROM THE VIEWPOINT OF NUTRITION. Teruo Ono and Yoh Imai (Hokkaido Univ., Sapporo, Japan). *Yukagaku* 19, 695-705 (1970). Topics reviewed include digestion and absorption of fats, translocation of fats and cholesterol, oxidation of fatty acids and essential fatty acids (deficiency, metabolism, fatty acid antagonism, and prostaglandin).

PRODUCTION AND UTILIZATION OF SOYBEAN PROTEIN PRODUCTS. Shiro Yamamoto and Kazuhiko Yoshitomi (Nisshin Oil Mills, Yokohama, Japan). *Yukagaku* 19, 826-34 (1970). Topics reviewed include components of soybeans, protein solubility of defatted soybean meal, components of various soybean products, solubility of protein and food uses, soybean varieties and protein yields, and uses of newer soybean protein foods.

EFFECT OF PHENOLIC ANTIOXIDANTS ON LIPOXYGENASE REACTION. Kyoden Yasumoto, Aijiro Yamamoto and Hisateru Mitsuda (Kyoto Univ., Kyoto, Japan). *Agr. Biol. Chem.* (Tokyo) 34(8), 1162-8 (1970). The effect of conventional antioxidants on soybean lipoxygenase reaction was examined. *o*-Diphenols such as pyrocatechol, homocatechol, propyl gallate and NDGA had higher inhibitory activity than *m*- and *p*-diphenols. NDGA showed a mode of inhibition conforming to the competitive type and not to the induction period type. Under certain conditions NDGA could be an irreversible inactivator for the enzyme. The inactivation by NDGA was effectively prevented

by incubation under anaerobic condition or at low pH values, or by adding borate. Thus the inactivation of lipoxygenase took place in parallel with autooxidation of NDGA.

LIPASE FROM CANDIDA PARALIPOLYTICA. IV. PURIFICATION, SOME PROPERTIES, AND MODIFICATION OF THE PURIFIED ENZYME WITH THE CONCENTRATED SOLUTION OF SODIUM CHLORIDE. Yasuhide Ota, Teruaki Nakamiya and Koichi Yamada (Univ. Tokyo, Japan). *Agr. Biol. Chem.* (Tokyo) 34(9), 1368-74 (1970). Lipase from *C. paraliipolytica* was purified 132-fold, as judged by disc electrophoresis. After purification, modification of the enzyme was performed by dialysing its solution against *M* NaCl in acetate buffer at room temperature and by separating the modified enzyme with Sephadex G-75. Purified lipase had optimum activity at pH 8.0, while the modified enzyme had its optimum at 7.0.

INCORPORATION OF SERINE-3-¹⁴C INTO SPHINGOLIPID BY RAT LIVER PARTICULATES. Yasuhiko Fujino and Masuo Nakano (Obihiro Zotech. Univ., Obihiro, Hokkaido, Japan). *Agr. Biol. Chem.* (Tokyo) 34(6), 974-6 (1970). Palmitoyl-Co A, labeled serine, and the tissue particulates were mixed, emulsified and incubated with cofactors. After incubation at 37C for 2 hours, small amounts of ceramide, cerebroside and sphingomyelin were added as carrier sphingolipids. More than 70% of the initial radioactivity was recovered from the long-chain base (sphingosine) fraction of the acid hydrolyzate not only in the presence but also in the absence of NADPH.

ISOLATION OF LIPOXYGENASE ISOZYMES AND COMPARISON OF THEIR PROPERTIES. Aijiro Yamamoto, Kyoden Yasumoto and Hisateru Mitsuda (Kyoto Univ., Kyoto, Japan). *Agr. Biol. Chem.* (Tokyo) 34(8), 1169-77 (1970). Lipoxygenase *b*, dependent on Ca ions, was isolated from soybean meal. Its protein and catalytic natures were compared with those of lipoxygenase *a*. They were different in electrophoretic mobility on polyacrylamide gel, pH-activity profile and sensitivity to added Ca⁺⁺.

PERIODICAL CHANGE IN THE CARCASS COMPOSITION OF CHICKS AFTER CHANGING TO LIPOGENIC OR LIPOLYTIC DIETS. Minoru Yoshida and Hiroshi Morimoto (Natl. Inst. Animal Ind., Chiba, Japan). *Agr. Biol. Chem.* (Tokyo) 34(3), 423-31 (1970). The change in carcass fat level was rapid and reversible. When the lipolytic diet was switched to the lipogenic diet, carcass fat increased gradually and reached apparent equilibrium after 12 (9-17) days. In the reverse switching, carcass fat decreased to reach apparent equilibrium after 7 days. The lipolytic or lipogenic effect of diet was not simply due to the change in supply of dietary energy to the chick.

AGAROSE-STARCH GEL ELECTROPHORESIS OF RAT SERUM LIPOPROTEINS. A. Chalvardjian (Res. Inst., Hospital for Sick Children, Toronto 101, Ontario, Canada). *J. Lipid Res.* 12, 265-69 (1971). Rat serum lipoproteins were separated into at least four fractions by agarose-starch gel electrophoresis. The system used was discontinuous in that glycine and sodium barbitone buffer was used in the reservoirs and Tris buffer was used for the gels. The four major bands could be related to the pattern obtained by ultracentrifugation. The high density lipoproteins consisted of at least two poorly resolved bands and were not separated from albumin. The vertical gel apparatus was further modified to accept 0.4 ml of rat plasma, which was prestained with Sudan black. After electrophoresis the different lipoprotein bands could conveniently be cut out and the lipid phosphorus determined. The addition of Sudan Black B decreased the recovery of the low and high density lipoproteins by 5-9%. However, the recovery of phospholipids was reproducible (80 ± 2%) and the high density lipoproteins contained over two-thirds of the plasma lipid phosphorus.

DIGLUCOSYLDIGLYCERIDE FROM *B. CEREUS*. K. Saito and K. Mukoyama (Dept. of Biochem., Kansai Med. Schl., Moriguchi, Osaka). *J. Biochemistry* 69, 83-90 (1971). Diglucosyldiglyceride from *B. cereus* was purified on columns of silicic acid and florasil. The structure proposed was glucosyl-(1 → 6)-glucosyl-(1 → 1)-diglyceride. The crystalline lipase (EC 3.1.1.3.) of *Eh. delemar*, which was known to attack the terminal ester linkage of the "synthetic" triglycerides, liberated from the diglucosyldiglyceride mainly the higher members of the constituent fatty acids, i.e., *br*-C₁₅, *br*- and *n*-C₁₆ and *br*-C₁₇ acids with a formation of diglucosylmonoglyceride. The constituent fatty acids of the diglucosylmonoglyceride, esterified at C-2 position, were mainly *br*-C₁₅, *br*-C₁₆ and *br*-C₁₇.

STIMULATION BY PHENOLS OF THE REOXIDATION MICROSOMAL BOUND CYTOCHROME *b*₅ AND ITS IMPLICATION TO FATTY ACID DESATURATION. N. Oshino and R. Sato (Inst. for Protein Res., Osaka Univ., Osaka). *J. Biochemistry* 69, 169-80 (1971). Various phenols, notably *p*-cresol, and two non-phenolic compounds have been found to stimulate the aerobic reoxidation of cytochrome *b*₅, reduced by NADH, in rat liver microsomes having a high activity of stearyl CoA desaturation. This stimulation is accompanied by simultaneous increases in the oxidation of NADH and consumption of molecular oxygen. The phenols added also seem to be oxidized. Evidence has been obtained that cytochrome *b*₅ located in a microsomal vesicle undergoes oxidation independently of that present in the other vesicles during the phenol-stimulated process. As in the similar stimulation of cytochrome *b*₅ reoxidation by stearyl CoA, the phenol effect is inhibited by cyanide. The magnitude of phenol effect can be correlated with the stearyl CoA desaturation activity of the microsomes; it is negligible in liver microsomes from fasted rats, but can be induced profoundly by refeeding the animals on a high carbohydrate diet. The phenol effect is also detectable in adipose tissue microsomes, which show a high desaturation activity. It is concluded that the phenols interact with the cyanide-sensitive factor, the termination enzyme of the microsomal desaturation system, resulting in an increased utilization by oxygen of electrons of reduced cytochrome *b*₅. Since the phenol effect is depressed by low concentrations of stearyl CoA, the cyanide-sensitive factor seems to react with stearyl CoA in preference to the phenols.

LIPID SYNTHESIS IN RATS DURING COLD ACCLIMATIZATION. M. Bhattathiry (Dept. of Biochem., Faculty of Med., Univ. of Malaya, Kuala Lumpur, Malaysia). *J. Biochemistry*, 69, 415-20 (1971). The effect of cold environment on the biosynthesis of lipids in warm-blooded animals was studied in terms of incorporation of acetate-¹⁴C into cholesterol and fatty acids of serum, liver and carcass of rats exposed to cold at 0-2C, for up to 28 days. The results were compared with those of normothermic rats. In vivo studies, the serum cholesterol concentration of cold-exposed rats was found to be higher, but the liver cholesterol concentration showed little changes. An initial decrease followed by a slight increase in total lipids of both serum and liver of cold-exposed rats was observed. The specific activity of total lipids from serum and liver and of serum cholesterol was very low during the first few days of cold-exposure but gradually reached normal values. At the end of 28 days the specific activity of serum cholesterol was higher in the case of cold-exposed rats, whereas the specific activity of liver cholesterol was higher for the first few days of exposure and slowly reached normal values in 28 days. In vitro studies using liver slices from cold-exposed and normothermic rats, the specific activity of both total cholesterol and fatty acids from livers of cold-exposed rats was low during the first few days, but gradually increased and showed 1.5 times and 3 times activity, respectively, in cholesterol and fatty acids at the end of 28 days of exposure to cold.

THE ELUTION BEHAVIORS OF ACIDIC PHOSPHOLIPIDS ON COLUMN CHROMATOGRAPHY. T. Shimojo, H. Kanoh and K. Ohno (Dept. of Biochem., Sapporo Med. College, Sapporo). *J. Biochemistry* 69, 255-63 (1971). Elution behavior of acidic phospholipids on silicic acid, cellulose, and Sephadex LH-20 column chromatography was investigated with special attention to the cations bound to phospholipids and to their purity, and the following results were obtained. The binding cations are not a primary factor determining their elution behavior but rather cause a secondary effect, affecting micelle formation of phospholipids. The difference in the chromatographic elution behavior between different cationic forms of acidic phospholipids seemed to be due to a difference in the stability and polarity of micelles. Na- and K-forms of isolated acidic phospholipids tend to form micelles which are smaller, more polar and probably less stable than those of their Ca- or Mg-forms in organic solvents. Acidic phospholipids with mixed cations in tissue extract are apt to form less stable or more polar micelles than those with a single species of cation. The mixed cation form is readily converted to the Ca-form by treatment of tissue extracts with CaCl₂.

EXTRACTION OF BILE ACIDS FROM RAT FECES CONTAINING CHOLESTYRAMINE. J.D. Manes and D.L. Schneider (Dept. of Nutritional Res., Mead Johnson Res. Center, Evansville, Ind. 47721). *J. Lipid Res.* 12, 376-77 (1971). The fecal extraction procedure described by Evrard and Janssen was inadequate for the complete extraction of conjugated bile acids from

feces containing the bile acid sequestrant, cholestyramine. As judged by gas-liquid chromatographic analysis, substitution of 0.5 N HCl in absolute ethanol for glacial acetic acid allowed for complete recovery (98-104%) of three different conjugated bile salts in the presence of the resin.

BIOSYNTHESIS OF SQUALENE AND STEROLS BY RAT AOETA. Marie M. Daly (Depts. of Biochem. and Med. and the Unit for Res. in Aging, Albert Einstein College of Med., Yeshiva Univ., Bronx, N.Y. 10461). *J. Lipid Res.* 12, 367-75 (1971). The synthesis of nonsaponifiable compounds from radioactive mevalonate by segments of adult rat aorta was studied *in vitro*. The labeled products consisted largely of substances with the chromatographic and chemical behavior of squalene, lanosterol, lathosterol and cholesterol. Even after 3 or 4 hr of incubation, the incorporation of mevalonate into squalene was higher than its incorporation into C₂₇ sterols; cholesterol contained less than 20% of the radioactivity in the total sterols. Lanosterol was the most highly labeled sterol. The level of radioactivity in lathosterol was comparable to the level in cholesterol. Small amounts of radioactivity were found in other sterols. Material with the same mobility on TLC as 7-dehydrocholesterol had less radioactivity than cholesterol, but more than sterols with the mobility of desmosterol. The results of measurements made after short periods of incubation showed that squalene and lanosterol became labeled before the other nonsaponifiable compounds.

NEUTRAL GLYCOLIPIDS IN LEUKEMIC AND NONLEUKEMIC LEUKOCYTES. J. Hildebrand, P. Stryckmans and P. Stoffyn (Dept. of Internal Med. and Clinical Invest., Inst. Jules Bordet, Brussels Univ., Brussels, Belgium). *J. Lipid Res.* 12, 361-66 (1971). Neutral lipids, free and total cholesterol, glycolipids and phospholipids were determined in 20 preparations of leukocytes distributed in four groups. Group I consisted of leukocytes from nonleukemic patients; group II, from patients with chronic myelogenous leukemia; group III, from patients with chronic lymphocytic leukemia; and group IV, from patients with acute leukemia. Two neutral glycolipids were found in nonleukemic mixed leukocyte populations. They were identified as glucosylceramide and lactosylceramide. The same glycolipids were also present in leukemic cells, but striking differences in glycolipid composition were found in various types of leukocytes. Glycolipids accounted for 8.9-12.6% of the total lipids in leukocytes from group I, 11.4-20.4% in group II, 1.2-1.6% in group III, and 0.5-4.9% in group IV. Glucosylceramide was the only glycolipid found in seven out of eight analyzed samples of lymphocytes, both normal and leukemic. Lactosylceramide was the major glycolipid in preparations consisting mainly of polymorphonuclear, myeloid and blastic cells. Only lactosylceramide was found in platelets where its concentration was about 100 times lower than in mixed leukocyte populations.

STRUCTURE OF SULFATIDES BIOSYNTHESIZED *IN VITRO*. P. Stoffyn, Anne Stoffyn and G. Hauser (Res. Lab McLean Hosp., Belmont, Mass. 02178). *J. Lipid Res.* 12, 318-23 (1971). Starting from galactose-¹⁴C-labeled phrenosine and 3'-phosphoadenosine-5'-phosphosulfate, radioactive sulfatides have been obtained *in vitro* with a biosynthetic system. Exogenous cerebrosides can act as acceptors of sulfate. The specific radioactivity of the synthetic phrenosine used as precursor was sufficiently high to permit the proof of the structure of the resulting sulfatides to be done by methylation on an amount estimated at 0.1 μg. The sulfate group was found only at C-3 of galactose, the position at which it is located in sulfatides isolated from tissues. This observation indicates the specificity of the sulfotransferase involved in the *in vivo* synthesis of sulfatides.

CHOLESTEROL BIOSYNTHESIS IN TRANSPLANTABLE HEPATOMAS: EVIDENCE FOR IMPAIRMENT OF UPTAKE AND STORAGE OF DIETARY CHOLESTEROL. D.S. Harry, H.P. Morris and N. McIntyre (Dept. of Med., Royal Free Hosp., London, N.W. 3 England). *J. Lipid Res.* 12, 313-17 (1971). Cholesterol feeding inhibits cholesterol biosynthesis in normal but not in malignant liver tissue. It has been postulated that hepatomas have suffered a specific intracellular deletion of the cholesterol feedback control mechanism, but there is little direct evidence to support this hypothesis. Rats bearing Morris transplantable hepatomas were fed high cholesterol diets for periods of up to 21 days. Cholesterol biosynthesis, as expected, was suppressed in the normal liver but not in hepatomas. The livers accumulated large amounts of cholesteryl ester but the hepatomas showed little or no increase in ester content. Cholesterol-¹⁴H was administered intragastrically to other tumor-bearing rats. Uptake of radioactivity by the tumors was much slower than

by normal liver. Comparison of the specific activities of liver and tumor cholesterol with that of the plasma suggested that the liver took up dietary cholesterol selectively from the blood, while the appearance of radioactivity in the tumors could be explained by slow equilibration with plasma cholesterol. Results suggest that the insensitivity of cholesterol biosynthesis to dietary cholesterol in hepatomas could be explained by an impairment in the uptake and storage of dietary cholesterol and that the concept of an intracellular deletion of the feedback mechanism requires further evidence.

FURTHER STUDIES OF THE EFFECTS OF AN ANOVULATORY DRUG ON LIPID METABOLISM IN THE RAT. L. Aftergood and R.R. Alfin-Slater (Schl. of Public Health, Univ. of Cal., Los Angeles, Cal. 90024). *J. Lipid Res.* 12, 306-11 (1971). The effect of various levels of the oral contraceptive drug, Enovid E, on serum and liver lipid levels of adult female rats has been investigated. Doses ranging from 0.052 to 1.04 mg/day have been employed in rats fed control or cholesterol-containing diets. It has been confirmed that after administration of even low, physiological doses of the drug, esterified cholesterol in serum and adrenals decreases rapidly while at the same time it accumulates in the liver; cholesteryl oleate is increased while the relative amount of cholesteryl arachidonate is reduced. Serum phospholipids also are decreased; the α/β lipoprotein ratio is significantly reduced due to the decrease of α-lipoproteins. Most of these changes also occur in cholesterol-fed rats. The observed effects are not related to a decreased food intake.

EFFECT OF FREE FATTY ACID MOBILIZATION ON THE ELECTROPHORETIC MOBILITY OF α-LIPOPROTEINS IN THE DOG. M.J. Lipson and S. Naimi (New England Med. Cent. Hospitals and the Dept. of Med., Tufts Univ. Schl. of Med., Boston, Mass. 02111). *J. Lipid Res.* 12, 294-305 (1971). Dogs were given infusions of norepinephrine and subsequent additional infusions of propranolol and nicotinic acid over a 4-hr period. Under different physiological conditions, α-lipoproteins of three different electrophoretic mobilities were identified by means of paper electrophoresis; they were designated α-lipoproteins X, Y and Z. During norepinephrine infusion, α-lipoprotein Y fell from 45% (of all lipoproteins) to 14%. There was a reciprocal rise in α-lipoprotein Z. On the other hand, α-lipoprotein X was not significantly changed. There was evidence that α-lipoprotein Y was progressively transformed into α-lipoprotein Z by increasing plasma FFA concentrations. The percentages of both α-lipoproteins Y and Z returned to original values after the dogs were given either nicotinic acid or propranolol. The alterations in the α-lipoprotein peaks Y and Z were rapid, being noted within 5 min of change in plasma FFA concentration. However, there appeared to be a threshold of plasma FFA concentration of 1200 μEq/liter, below which no changes in α-lipoproteins were noted. It was concluded that α-lipoproteins were noted. It was concluded that α-lipoprotein Y is rapidly, progressively, but reversibly transformed into α-lipoprotein Z by binding to plasma FFA above a threshold level of 1200 μEq/liter. However, α-lipoprotein X does not appear to be involved in the binding of plasma FFA.

GROWTH AND LIPOLYSIS OF RAT ADIPOSE TISSUE: EFFECT OF AGE, BODY WEIGHT AND FOOD INTAKE. R.W. Hubbard and W.T. Matthew (Biochem., Pharmacol. Lab., U.S. Army Res. Inst. of Environmental Med., Natick, Mass. 01760). *J. Lipid Res.* 12, 286-93 (1971). The purpose of the present work was to study age- and weight-controlled rats to determine which is the primary factor in reducing the lipolytic response of free fat cells to non-fat cells in adipose tissue. The method for estimating fat cell and nonfat cell numbers is based on the analysis of adipose tissue and fat cell DNA and lipid. In adequately fed rats, epididymal adipocyte hyperplasia is complete between 9 and 14 wk of age. Chronic underfeeding delays, but does not eliminate, normal fat cell hyperplasia and is accompanied by a net loss in the nonfat cell population. During 9-14 wk of age, epididymal adipose tissue enlarges mainly through adipocyte hypertrophy. Total fat cells from the epididymal adipose tissue of control rats represent only 20-23% of the total cell population. Chronic underfeeding increases the percentage of fat cells in the fat pad from 23 to 28%. Noradrenaline-stimulated lipolysis is proportional to fat cell numbers but is inhibited when fat cell lipid increases to over 80% of fat pad wet weight. Rat age is apparently not primarily responsible for the decreased noradrenaline-stimulated lipolysis in fat cells of 350-g rats *in vitro*.

ENZYMIC FORMATION OF ESTERS OF METHYL STEROL PRECURSORS

OF CHOLESTEROL. D.R. Brady and J.L. Gaylor (Grad. School of Nutr., Cornell Univ., Ithaca, N.Y. 14850). *J. Lipid Res.* 12, 270-76 (1971). For investigation of the reactions of cholesterol biosynthesis, a number of workers use the 10,000 g supernatant fraction (or similar preparations) obtained from cell-free homogenates of rat liver. Esters of methyl sterol biosynthetic intermediates are formed by this crude source of enzymes. Esters of C₂₀, C₂₆, C₂₈, and C₂₇-sterol intermediates have been isolated by silicic acid chromatography of an acetone extract of incubation mixtures. Competition between ester formation and demethylation of the C₂₈-sterol intermediate has been demonstrated. With 4 α -methyl-5 α -cholest-7-en-3 β -ol as substrate, maximal velocities of ester formation (0.36 nmole/30 min per mg of protein) were almost equivalent to maximal velocities of demethylation (0.45 nmole/30 min per mg of protein). Ester formation may be eliminated by carrying out incubations with microsomal preparations; ester formation may be restored completely upon addition (to the microsomes) of either coenzyme A and ATP or the supernatant fraction resulting from centrifugation at 105,000 g. Ester formation has been examined similarly with broken-cell preparations of rat skin. With 4 α -methyl-5 α -cholest-7-en-3 β -ol as substrate, the rate of ester formation was more than six times the rate of methyl sterol demethylation. The very significant competition between esterification and demethylation of methyl sterol intermediates of skin suggests that sterol intermediates accumulate in rat skin because of the rapid formation of esters that may not be further metabolized.

THE FORMATION AND REDUCTION OF THE 14,15-DOUBLE BOND IN CHOLESTEROL BIOSYNTHESIS. I.A. Watkinson, D.C. Wilton, K.A. Munday and M. Akhtar (Dept. of Physiol. and Biochem., Univ. Southampton, Southampton SO9 5NH, U.K.). *Biochem. J.* 131-37 (1971). It was shown that 100 μ g quantities of 4,4-dimethyl(2-³H₂)cholesta-8,14-dien-3 β -ol (IIIa), tritiated cholesta-8,14-dien-3 β -ol, 4,4-dimethyl(2-³H₂)cholesta-7,14-dien-3 β -ol, dihydro(2-³H₂)lanosterol and (24-³H)lanosterol were converted by a 10,000 g supernatant of rat liver homogenate into cholesterol in 17%, 54%, 6%, 9.5% and 24% yields, respectively. From an incubation of dihydro(3 α -³H)lanosterol with a rat liver homogenate in the presence of a trap, up to 38% of the radioactivity was found to be associated with a fraction that was unambiguously shown to be 4,4'-dimethylcholesta-8,14-dien-3 β -ol. Another related compound, 4,4-dimethylcholesta-7,14-dien-3 β -ol was also shown to be equally effective in its ability to trap compound (IIIa) from an incubation of dihydro(3 α -³H)lanosterol. The mechanism of the further conversion of the compound (IIIa) into cholesterol occurred by the reduction of the 14,15-double bond and involved the addition of a hydrogen atom from the medium to C-15 and another from the 4-position of NADPH to C-14. Two possible mechanisms for the removal of the 14 α -methyl group in sterol biosynthesis are discussed.

ISOLATION OF THE CADMIUM DERIVATIVE OF LIPOYL DEHYDROGENASE. A.M. Stein and Jeanne H. Stein (Dept. of Biochem., Univ. of Florida College of Med., Gainesville, Fla. 32601). *J. Biol. Chem.* 246, 670-76 (1971). Reduced lipoamide dehydrogenase reacts with cadmium ion to form a moderately stable compound characterized by 1 g atom of cadmium bound per mole of flavin adenine dinucleotide, a modified spectrum in the visible region, and altered reactivity with electron acceptor substrates. The 450-m μ band is shifted to the blue and the 370-m μ shoulder is lost. On storage, the spectrum of the cadmium-lipoil dehydrogenase complex tends to return to that of the unmodified enzyme. Alkylated lipoil dehydrogenase, inactive in the DPNH-lipoamide dehydrogenase reaction, forms the corresponding cadmium compound. The latter appears more stable than the cadmium compound of the unalkylated enzyme. The activity profile of the cadmium compound is characterized by increased rates of 2,6-dichloroindophenol and quinone reductase, little change in ferriyanide reductase, decreased pyridine nucleotide transhydrogenase, and highly inhibited DPNH-lipoamide dehydrogenase activity.

REDUCING THE EGG CHOLESTEROL LEVEL BY INCLUDING EMULSIFIED SITOSTEROL IN STANDARD CHICKEN DIET. R. Clarenburg, I.A. Kim Chung and Lucille M. Wakefield (Dept. of Physiol. Sci. and Dept. of Foods and Nutr., Kansas State Univ., Manhattan, Ks. 66502). *J. Nutr.* 101, 289-98 (1971). During a 2-month control period, laying hens were fed a standard ration; cholesterol levels in eggs laid were remarkable constant for each hen, but varied significantly from hen to hen. During the next 2 months, the hens were fed the standard ration supplemented with 0.5% carboxymethylcellulose and 0, 1, 2, or 4% ³H-sitosterol emulsion. Intestinal absorption

of the plant sterol from the 2% diet amounted to 60% for laying hens and 85% for non-layers. Lowered egg cholesterol levels and sitosterol incorporation in eggs were established. Similar maximal effects—reducing egg cholesterol levels by as much as 35% and incorporating 42 mg of sitosterol per egg—were observed when either the 2% or the 4% sitosterol diet was fed, but at the higher sitosterol level responses were more prompt. When the hens again were fed the standard layer ration, egg cholesterol levels reverted toward normal.

EFFECT OF PREVIOUS HIGH FAT DIET ON BODY PROTEIN METABOLISM IN RATS. K. Nakano, S. Kurimoto and K. Ashida (Lab. of Nutr. Biochem., Dept. of Agr. Chem., Nagoya Univ., Chikusa, Nagoya, Japan). *J. Nutr.* 101, 895-900 (1971). When rats were fed a high fat diet followed by a high carbohydrate test diet, they excreted less urinary nitrogen than rats fed only a high carbohydrate diet. Concomitantly, activities of two hepatic amino acid-catabolizing enzymes, e.g., threonine dehydratase and arginase, were significantly decreased in rats fed a high fat diet followed by a high carbohydrate diet. The ratio of radioactivity recovered from injected ¹⁴C-amino acids, ¹⁴C-body protein/respiratory ¹⁴CO₂, indicates that the body protein metabolism in rats fed a high fat diet followed by a high carbohydrate test diet is shifted to the anticatabolic fashion as compared with rats fed constantly a high carbohydrate diet. The analytical results of blood glucose in rats fed a high carbohydrate test diet indicate that the utilization of glucose is much reduced by the previous feeding of a high fat diet compared with the previous feeding of a high carbohydrate diet. It is inferred that a marked rise of blood glucose in fat-fed rats responding to the feeding of a high carbohydrate test diet may result in an increased secretion of insulin, which is known to exhibit an anticatabolic effect on the body protein metabolism in the animal.

VITAMIN D STIMULATED CALCIUM BINDING PROTEIN FROM RAT INTESTINAL MUCOSA. PURIFICATION AND SOME PROPERTIES. D. Drescher and H.F. DeLuca (Dept. of Biochem., Univ. of Wis., Madison, Wis. 53706). *Biochemistry* 10, 2302-7 (1971). Rat intestinal calcium binding protein made in response to vitamin D has been purified to homogeneity. Two main purification procedures have been employed, each of which is suitable for preparative purposes. The calcium binding activity from the supernatant fraction of intestinal mucosa has been purified several 100-fold by means of column chromatography with Sephadex G-100 followed CM-Sephadex. A second procedure involving gel filtration on Bio-Gel P series resins also yields a homogeneous calcium binding protein. Using the latter procedure another protein having similar charge to the calcium binding protein has been isolated, and is thought to be a precursor of the calcium binding protein. Homogeneity of the calcium binding protein obtained by these methods has been established by column chromatography, urea disk gel electrophoresis, and ultracentrifugation. A molecular weight of between 8000 and 9000 has been estimated by sedimentation equilibrium ultracentrifugation measurements while a molecular weight of 13,000 has been estimated by means of gel filtration and disc gel electrophoresis.

POSSIBLE PRECURSOR OF VITAMIN D STIMULATED CALCIUM BINDING PROTEIN IN RATS. *Ibid.*, 2308-12. The supernatant fraction of intestinal mucosa has been fractionated by means of urea disk gel electrophoresis. Seventy-two hours after administration of vitamin D₃ to vitamin D deficient rats, a new protein (band C) appears in the supernatant fraction. Concomitant with the appearance of the new protein, there is a depression of a preexisting protein (band A) of apparently higher molecular weight than the newly appearing one (band C). These changes are confirmed by experiments involving incorporation of radioactive amino acids into these proteins. The molecular weight range, time course of appearance, and effect of dietary strontium all suggested that the newly appearing protein (band C) might represent the calcium binding protein formed in response to vitamin D. Using chromatographically purified calcium binding protein from rat intestine, it was shown that this protein (band C) is most probably the calcium binding protein. The major contaminating protein in the final chromatographic purification of rat calcium binding protein has also been identified on gels as the preexisting protein (band A), which is probably the precursor of the calcium binding protein.

ISOLATION AND IDENTIFICATION OF 1,25-DIHYDROXYCHOLECALCIFEROL, A METABOLITE OF VITAMIN D ACTIVE IN INTESTINE.

M.F. Holick, H.K. Schoes, H.F. DeLuca, T. Suda and T.J. Cousins (Dept. of Biochem., College of Agr. and Life Sciences, Univ. of Wis., Madison, Wis. 53706). *Biochemistry* 10, 2799-2804 (1971). A metabolite of vitamin D₃, thought to be the "tissue-active" form of the vitamin in the intestine, has been isolated from chicken intestines in pure form as the mono-trimethylsilyl ether derivative. The structure of this metabolite has been identified as 1,25-dihydroxycholecalciferol by means of mass spectrometry, ultraviolet absorption spectrophotometry and specific chemical reactions.

VITAMIN D₂ AND CHOLESTEROLEMIA IN THE GROWING RAT. M.H. Jurgens, C.T. Blunn and E.R. Peo, Jr. (Nebraska Agr. Expt. Stat., Lincoln, Neb. 68503). *J. Nutr.* 101, 153-60 (1971). The influence of dietary supplements of vitamin D₂ or cholesterol, or both, upon blood serum cholesterol and the cholesterol and fatty acid content of certain tissue was studied in rats. In all treatments, female rats had higher serum but lower liver cholesterol concentrations than intact males. Rats fed cholesterol had elevated levels of serum and liver cholesterol, increased percentage of body fat, but reduced body cholesterol concentration as compared to noncholesterol-fed rats. The inclusion of excessive vitamin D₂ when fed in the presence of dietary cholesterol resulted in lowered serum cholesterol levels ($P < 0.01$) and dramatically increased liver cholesterol content. Total saturated fatty acids tended to be reduced while oleic acid was increased in the liver and body of rats fed a high level of vitamin D₂ and cholesterol in combination, compared to levels in rats fed the basal diet. The data suggest that the serum cholesterol-lowering effect of vitamin D₂ may be related in part to increased retention of cholesterol by the liver.

UTILIZATION OF FAT AND GLYCEROL FOR GLYCOGENESIS BY THE NEONATAL RAT. J.D. Johnson, Ruth Hurwitz and N. Kretschmer (Dept. of Pediatrics, Stanford Univ. Schl. of Med., Stanford, Cal. 94305). *J. Nutr.* 101, 299-306 (1971). Hepatic glycogenesis from fat and glycerol has been studied in the fasted rat during development. Administration of either olive oil or glycerol resulted in marked hepatic glycogenesis in 5- to 10-day-old animals; this response was diminished in weaned rats and was almost absent in animals less than 48 hours old. The incorporation of ¹⁴C-glycerol into hepatic glycogen was greater in 9-day-old neonatal rats than in weaned animals. Although glycerol kinase activity was low in liver of animals less than 48 hours of age, these young animals readily converted ¹⁴C-glycerol into ¹⁴CO₂. Our results indicate that glycerol is utilized via different pathways depending on the age of the animal. Hepatic glycogenesis, following fat administration, in the 5- to 10-day-old rat can be explained on the basis of increased utilization of triglyceride-glycerol for glycogenesis during this period of neonatal life.

THE ROLE OF BETA-LIPOPROTEIN, CHOLESTEROL AND VARIOUS SERA IN TISSUE CULTURE INTRACELLULAR LIPIDOSIS. R.D. Maca and K.D. Rose (Univ. Health Cent., Univ. of Nebraska, Lincoln, Neb. 68508). *Proc. Soc. Exp. Biol. Med.* 136, 457-60 (1971). Serum from human, calf, fetal calf, horse and roosters fed either a regular or high cholesterol diet produced sudanophilic cytoplasmic inclusions within cloned mouse fibroblasts ("Low line" NCTC No. 2445) in tissue culture. Serum from roosters either fed a 1.0% cholesterol diet or subjected to heat stress exhibited an increased ability to produce cytoplasmic inclusions. The serum protein factor responsible for these inclusions proved to be the low density β -lipoprotein. α -Lipoprotein produced inclusions only in high concentrations. Besides varying with different sera, the amount of lipodosis was found to be directly proportional to total cholesterol and to the percentage of β -lipoprotein-bound unesterified cholesterol, but inversely proportional to the percentage of bound esterified cholesterol. Furthermore, bovine albumin-bound cholesterol in the unesterified form produced similar appearing sudanophilic, cytoplasmic inclusions. These observations suggest that the protein-bound, and more specifically, the β -lipoprotein-bound, unesterified cholesterol plays a significant role in inclusion formation, although other factors are probably involved in this process.

THE COMPOSITION OF CHOLESTEROL ESTERS IN FATTY STREAKS AND ATHEROSCLEROTIC PLAQUES OF THE HUMAN AORTA. HISTOCHEMOTOPHATIC INVESTIGATIONS. Brigitte Kunnert and H. Krug (Dept. of Pathol., Karl Marx Univ., Leipzig (G.D.R.)). *J. Atheroscler. Res.* 13, 93-101 (1971). The histochemotopograms of fatty streaks and atherosclerotic lesions showed the following six cholesterol ester fractions: saturated fatty acids, acids with one, two, three, four and more than four double bonds. The two largest fractions were of the oleic acid type

and of the linoleic acid type. Most of the fatty streaks belonged to the oleic acid type, while most of the atherosclerotic plaques belonged to the linoleic acid type. But we also found lipidoses of the linoleic type and atherosclerotic plaques of the oleic acid type as well as chromatograms with equal fractions of oleic and linoleic acid ester fractions. Our parallel histological and histochemical investigations suggest that the cellular activity plays an important role in the genesis of the cholesterol ester composition. We believe that the plasma cholesterol esters (linoleic acid type) that entered the arterial wall were converted intracellularly to esters with more double bonds in their fatty acid moiety. By this mechanism (overloading by lipids or cell injury caused by a noxa) we may see an accumulation of oleic acid cholesterol esters, a further transition to more unsaturated acids being impossible physiologically. The differences in the cholesterol ester composition of arteriosclerotic lesions are the results of extracellular infiltration of plasma cholesterol esters, intracellular transformation and secondary extracellular deposition by necrosis of lipid-laden (foam) cells.

STUDIES ON THE RELATIONSHIP BETWEEN LIPOGENESIS AND THE LEVEL OF COENZYME A DERIVATIVES, LACTATE AND PYRUVATE IN CHICK LIVER. Y. Yeh and G.A. Leveille (Lab. of Nutr. Biochem., Dept. of Animal Sci., Univ. of Illinois, Urbana, Ill. 61801). *J. Nutr.* 101, 911-18 (1971). An attempt was made to elucidate the mechanisms by which hepatic lipogenesis is regulated by dietary protein and fat and by food restriction and refeeding in the growing chick. The reduction in hepatic fatty acid synthesis in fat-fed and short-term fasted chicks was accompanied by a decrease in the hepatic concentration of free CoA and in the lactate/pyruvate ratio and by an increase in the level of plasma free fatty acids and liver long-chain acyl-CoA derivatives. The data indicate that the depression in hepatic fatty acid synthesis caused by either fat feeding or a short fast can be attributed to: 1) a reduced availability of free CoA for citrate cleavage activity which would limit cytoplasmic acetyl-CoA generation for fatty acid synthesis; and 2) a decreased availability of cytoplasmic reducing equivalents to support reductive fatty acid biosynthesis. Increased dietary protein levels depressed hepatic lipogenesis and decreased plasma free fatty acid levels and the liver lactate/pyruvate ratio. Increased dietary protein did not, however, influence the hepatic levels of free CoA or acetyl-CoA in spite of the fact that long-chain acyl-CoA derivative levels were increased. The decreased ratio of lactate/pyruvate suggests that a limitation in the availability of cytoplasmic reducing equivalents may initiate the reduction in hepatic fatty acid synthesis in chicks fed high protein diets.

REMOVAL OF LIPIDS FROM HUMAN PLASMA LOW-DENSITY LIPOPROTEIN BY DETERGENTS. A. Helenius and K. Simons (Dept. of Serology and Bact., Univ. of Helsinki, Haartmaninkatu 3, Helsinki 29, Finland). *Biochemistry* 10, 2542-47 (1971). Four different detergents have been found to remove all major lipids from human plasma low-density lipoprotein (LDL). The detergents used were a bile salt-sodium deoxycholate, a neutral detergent (Nonidet P40), an anionic detergent (sodium dodecyl sulfate) and a cationic detergent (cetyltrimethylammonium bromide). High concentrations of these detergents were added to aqueous solution of LDL. The protein moiety could then be separated from the mixed lipid-detergent micelles by gel filtration in the presence of micellar concentrations of the detergent. The lipid-free protein obtained with Nonidet P40 or with sodium deoxycholate retained the immunological properties of the native LDL as shown by double diffusion in agarose gels against anti-LDL-sera. The lipid-free protein obtained with sodium dodecyl sulfate or cetyltrimethylammonium bromide had altered immunological properties.

PLASMA CHOLESTEROL LEVELS AND LIVER CHOLESTEROL BIOSYNTHESIS IN RABBITS FED COMMERCIAL OR SEMISYNTHETIC DIETS WITH AND WITHOUT ADDED FATS OR OILS. K.K. Carroll (Dept. of Biochem., Univ. of Western Ontario, London 72, Ont. (Canada)). *J. Atheroscler. Res.* 13, 67-76 (1971). Rabbits were fed semisynthetic or commercial diets, low in fat or containing 15% by weight of various added fats or oils, and after 2 weeks on diet, plasma cholesterol levels were measured and the ability of liver slices to incorporate acetate into cholesterol and fatty acids was determined. Plasma cholesterol levels were high in rabbits fed semisynthetic diets containing no fat or saturated fats. They were normal or only slightly elevated on the corresponding commercial diets and on either semisynthetic or commercial diets containing polyunsaturated oils. Addition of casein to commercial diets elevated plasma

cholesterol except when polyunsaturated oils were present but added dextrose had little effect. Growth was better on commercial than on semisynthetic diets and was stimulated by adding casein but was inhibited by adding dextrose to the commercial diet. Acetate incorporation into cholesterol was generally lower on semisynthetic than on commercial diets, and added fats had no consistent effects. Incorporation into fatty acids tended to be higher on semisynthetic than on commercial diets but was decreased by adding fats to either diet. Liver levels of cholesterol and fatty acids were generally higher on high fat diets. Addition of casein to commercial diet depressed acetate incorporation into cholesterol. Added dextrose had no effect and neither casein nor dextrose had any consistent effect on incorporation into fatty acids.

PARTIAL PURIFICATION AND CHARACTERIZATION OF THE PHOSPHOLIPASE A₂ FROM RAT LIVER MITOCHONDRIA. M. Waite and Patricia Sisson (Dept. of Biochem., Bowman Gray School of Med. of Wake Forest Univ., Winston-Salem, N.C. 27103). *Biochemistry* 10, 2377-83 (1971). Phospholipase A (phosphatide acyl-hydrolase, EC 3.1.1.4) was purified 160-fold from rat liver mitochondria by precipitating the proteins with ammoniacal acetone, extracting the precipitated proteins with potassium chloride solution, fractionating the extract by ammonium sulfate precipitation, and separating the enzyme by gel filtration through Sephadex G-200. The purified enzyme catalyzed production of equal amounts of unsaturated fatty acid and 1-acylglycerophosphorylethanolamine from phospholipids which indicates that the enzyme is specific for the 2 position and is not contaminated with a lysophospholipase. Phosphatidylethanolamine was most rapidly hydrolyzed at pH 9.5, less at pH 7.4. In contrast phosphatidylserine was hydrolyzed more extensively at pH 7.4 than pH 9.5. Phosphatidylcholine, phosphatidic acid, phosphatidylinositol and cardiolipin were less extensively hydrolyzed. Mixtures of phosphatidylethanolamine and cardiolipin were hydrolyzed more completely than was either compound alone at pH 7.4 but not at 9.5. Both hexadecyltrimethylammonium chloride and di-n-octadecyl phosphate stimulated hydrolysis of phosphatidylethanolamine at pH 7.4. Hexadecyltrimethylammonium chloride in low concentrations stimulated and di-n-octadecyl phosphate inhibited hydrolysis at pH 9.5. Monoacylglycerophosphorylethanolamine inhibited the reaction 50-75%. The rate at which fatty acids were removed from phosphatidylethanolamine (in decreasing order) was oleic, linoleic, linolenic and arachidonic acids. This was uninfluenced by the nature of the acid in the 1 position. Hydrolysis of phosphatidylethanolamine was first order only during the first 3-5 min.

ORIGIN OF PLASMA FATTY ACIDS IN LACTATING COWS FED HIGH GRAIN OR HIGH FAT DIETS. D.L. Palmquist and H.R. Conrad (Dept. of Dairy Sci., Ohio Agr. Res. Center, Wooster, Ohio 44691). *J. Dairy Sci.* 54, 1025-33 (1971). Palmitate-1-¹⁴C was given to lactating cows in 20 trials involving four dietary treatments and two different routes of tracer administration. The specific activity-time curve of ¹⁴C-activity in milk fat was resolved by curve analysis into two components, a rapid-turnover component attributed to exogenous (dietary) fatty acid and a slower turnover component attributed to adipose tissue. Administration of palmitate-1-¹⁴C abomasally or orally gave greater ($P < 0.001$) estimates of exogenous fatty acid than did intravenous dosing. A high grain-restricted roughage diet reduced ($P < 0.01$) the exogenous estimate, presumably due to increased uptake of dietary fatty acid by adipose tissue. A low-fat diet also lowered the estimate of exogenous fatty acid. A high-fat diet reduced the turnover time of the adipose tissue pool by 30% whereas the high grain, restricted roughage diet increased the turnover time by 26%. Estimates of the effects of dietary treatments on rate constants of fatty acid transfer in a 2-pool model are presented. Low fat and high grain restricted roughage diets reduced plasma free fatty acids ($P < 0.01$ and 0.001). The high grain restricted roughage diet increased plasma glucose and serum heparin-precipitable lipoprotein esters ($P < 0.001$ and 0.001).

ON THE MODE OF ACTION OF LIPID-LOWERING AGENTS—KINETICS OF THE INHIBITION IN VITRO OF RAT ACETYL COENZYME A CARBOXYLASE. M.E. Maragoudakis and Hilda Hankin (Ciba Pharma. Co., Res. Dept., Summit, N.J. 07901). *J. Biol. Chem.* 246, 348-358 (1971). Rat liver acetyl coenzyme A carboxylase was purified about 200-fold and the inhibition of this enzyme by certain hypolipidemic drugs was studied. The inhibition was more pronounced if the drugs were added before rather than after the citrate activation of the enzyme. Kinetic analysis revealed noncompetitive inhibition of the drugs with

respect to the substrates acetyl-CoA, ATP and HCO₃⁻, and competitive inhibition with respect to the activator, citrate. Sucrose density gradient centrifugations showed that the drugs reverse the aggregating effect of citrate to form the active polymeric forms of the enzyme from the inactive monomers. Arrhenius plots and heat-inactivation studies suggest gross conformational changes of the enzyme protein in the presence of the drugs. Relative affinities of acetyl coenzyme A carboxylase for citrate and the drugs are expressed by the calculated dissociation constant for citrate and the inhibition constants of the drugs. Derangement of fatty acid synthesis *in vivo* is conceivable by competition of the drugs with the activation of acetyl-CoA carboxylase at low and physiologically possible concentrations of the drugs and citrate.

OBSERVATIONS ON THE LIPOLYTIC AND MELANOTROPIC ACTIVITIES OF THE PINEAL GLAND. D. Rudman, A.E. Del Rio, Bettye Hollins and Diane H. Houser (Depts. of Med. and Biochem., Emory Univ. Schl. of Med., Atlanta, Ga. 30322). *J. Biol. Chem.* 246, 324-330 (1971). An extract of ovine and bovine pineal glands (labeled Precipitate II) has previously been shown to possess lipolytic activity on rabbit adipose tissue. This extract also causes darkening of the frog's skin *in vivo* and *in vitro*. Both lipolytic and melanotropic activities are abolished by trypsin and chymotrypsin. Fractionation of ovine pineal Precipitate II by the sequence, chromatography on diethylaminoethyl cellulose → chromatography on carboxymethyl cellulose → gel filtration (repeated once) on Sephadex G-25, concentrated both activities 100- to 1000-fold. Only one component was visible in acrylamide gel electrophoretograms of the final concentrate; its volume of elution from Sephadex G-25 indicated a molecular weight of 1000 to 3500; its lipolytic and melanotropic potencies were about 10 to 80% those of adrenocorticotropin. The bovine peptide, prepared in a similar way, was closely similar to or identical with the ovine counterpart in electrophoretic mobility, molecular weight, lipolytic and melanotropic potencies, and amino acid composition.

MECHANISM OF PROSTAGLANDIN BIOSYNTHESIS. I. CHARACTERIZATION AND ASSAY OF BOVINE PROSTAGLANDIN SYNTHETASE. C. Takeguchi, E. Kohno and C.J. Sih (School of Phar., Univ. of Wis., Madison, Wis. 53706). *Biochemistry* 10, 2372-76 (1971). The Zimmerman reaction was found to be adaptable for the assay of prostaglandin synthetase, which is capable of catalyzing the conversion of arachidonic acid into 11 α -15 α -dihydroxy-9-oxo-5-cis,13-trans-prostadienoic acid. The bovine seminal vesicle microsomes possess active prostaglandin synthetase activity when a heat-labile, nondialyzable inhibitor, present in the supernatant fraction was removed. Various parameters affecting the rate of this biosynthetic reaction have been quantitatively defined. To achieve maximal rate, both GSH and a cofactor must be present. The following compounds were found to be suitable cofactors: p-aminophenol, hydroquinone, L-norepinephrine, L-pinephrine, serotonin and 5-hydroxyindolacetic acid. The pH optimum of this reaction depends on the specific coenzyme used.

LOCAL STIMULATORY EFFECT OF VITAMIN A ON SPERMATOGENESIS IN THE RAT. B. Ahluwalia and J.C. Bieri (Lab. Nutr. and Endocrinology, Nat'l Inst. of Arthritis and Metabolic Diseases, Nat'l Inst. of Health, Bethesda, Md. 20014). *J. Nutr.* 101, 141-51 (1971). The objectives of this study were: (1) to see if a localized effect of retinol could be demonstrated on the germinal epithelium of rat testes and (2) to determine if retinoic acid administered directly into the testis would support spermatogenesis. When 10 μ g of retinol in oil was injected intratesticularly into vitamin A-deficient rats, marked stimulation of the germinal epithelium around the injection site occurred in 14 days. A larger amount of retinol, 400 μ g, injected under the testicular capsule or the implantation of a 3 mg pellet restored spermatogenesis throughout the testis, and also in the corresponding untreated testis, in 50 days. Substitution of retinoic acid or its methyl ester in the above procedures was completely ineffective. Retinoic acid when combined with retinol in an implanted pellet did not interfere with the stimulatory activity of the retinol. It is concluded that retinol has a local direct biochemical action on the germinal epithelium and that retinoic acid is totally inactive.

INFLUENCE OF FEEDING COTTONSEED OIL TO LAYING HENS ON THE LIPOVITELLINS OF THEIR EGGS. R.J. Evans, Doris H. Bauer and C.J. Flegal (Depts. of Biochem. and Poultry Sci., Michigan State Univ., East Lansing, Mich. 48823). *J. Nutr.* 101, 355-62 (1971). Lipovitellins were isolated from normal eggs

and from eggs laid by hens fed crude cottonseed oil (cottonseed lipovitellin). Cottonseed lipovitellin contained 23% lipid compared to 17% in normal lipovitellin, and the lipids of cottonseed lipovitellin contained more stearic acid and less oleic acid than those of normal lipovitellin. Amino acid compositions of the vitellins (proteins) were similar. Ether extracted 49% of the lipid from cottonseed lipovitellin compared to 16% of the lipid from normal lipovitellin. Progressive digestion with proteolytic enzymes released similar amounts of lipids and peptides from ether-extracted normal and cottonseed lipovitellins. Chromatography on a TEAE-cellulose column separated lipovitellins into α - and β -lipovitellins. Normal lipovitellins were better separated than cottonseed lipovitellins, and the ratio of α -lipovitellin to β -lipovitellin was smaller in normal than in cottonseed lipovitellins. The principal difference between normal and cottonseed lipovitellins appeared to be in the higher content of saturated fatty acids in the lipid, the larger amount of lipid bound on the outer surface of the molecule, and the higher proportion of α -lipovitellin in cottonseed lipovitellins than in normal lipovitellins.

INTERACTION OF DIETARY FAT AND THYROID FUNCTION WITH HEPATIC AND RENAL GLUCONEOGENESIS OF RATS. H. Suzuki and H. Fuwa (Lab. Nutr. Biochem., Dept. Agr. Chem., Nagoya Univ., Nagoya, Japan). *J. Nutr.* 101, 919-26 (1971). The influence of thyroid function on the capacity for glucose formation in rats fed a high carbohydrate or a high fat diet was investigated with the use of propylthiouracil and L-thyroxine. The food intake, growth rate and blood glucose level of rats fed the high fat diet were depressed in hypothyroidism produced by feeding 0.01% propylthiouracil. However, rats fed the high carbohydrate diet were unaffected by feeding 0.01% or 0.05% propylthiouracil. The hepatic activity of glucose-6-phosphatase was decreased significantly by feeding propylthiouracil, while the renal activity of glucose-6-phosphatase did not show any significant change in either of these dietary groups. Moreover, percentage decline of activity due to propylthiouracil was greater in the high fat group than in the high carbohydrate group. L-Thyroxine injected intraperitoneally caused significant increases in the hepatic and renal activities of glucose-6-phosphatase of rats fed the high carbohydrate diet. The effects of thyroxine and the high fat diet were additive in the liver, whereas no change in the renal activity was observed. These findings support the hypothesis that increases in the capacity for glucose formation and glucose-6-phosphatase activity with dietary fat, which are observed in the liver only, might be related to increased thyroid activity as a consequence of feeding a high fat diet.

INSIDE-OUTSIDE TRANSITIONS OF PHOSPHOLIPIDS IN VESICLE MEMBRANES. R.D. Kornberg and H.M. McConnell (Stauffer Lab. for Phys. Chem., Stanford Univ., Stanford, Cal.). *Biochemistry* 10, 1111-20 (1971). Spin-labeled phosphatidylcholine is a paramagnetic analog of phosphatidylcholine. The vesicle which results from prolonged sonication of egg phosphatidylcholine and spin-labeled phosphatidylcholine in salt solution has an aqueous compartment and a bilayer membrane. Sodium ascorbate at 0°C abolishes the paramagnetism of spin-labeled molecules in the external monolayer of the vesicle membrane (65% of the total paramagnetism of a vesicle) without affecting the paramagnetism of internal molecules. The consequent asymmetry in the distribution of paramagnetic molecules between the two monolayers of the vesicle membrane decays with a half-time of 6.5 hr at 30°C. It follows that the probability of a spin-labeled phosphatidylcholine molecule passing from the internal monolayer of the vesicle membrane to the external monolayer is 0.07/hr at 30°C, and the probability of a spin-labeled phosphatidylcholine molecule passing from the external monolayer of the vesicle membrane to the internal monolayer is 0.04/hr at 30°C.

IDENTIFICATION OF PHOSPHOLIPASE A₁ AND A₂ IN THE SOLUBLE FRACTION OF RAT LIVER LYSOSOMES. R. Franson, M. Waite and Mariano LaVia (Dept. of Biochem. and Pathol., Bowman Gray School of Med. of Wake Forest Univ., Winston-Salem, N.C. 27103). *Biochemistry* 10, 1942-46 (1971). Rat liver lysosomes were isolated by sucrose density gradient centrifugation from rats previously injected with Triton WR-1339. As measured by acid phosphatase activity, the lysosomes were purified 31-fold over the homogenate with an average yield of 6.0%. Mitochondrial, microsomal and peroxisomal contaminations were each less than 0.05% of the total activity of the homogenate. When the lysosomes were incubated at

pH 4.0 with 1.0 mM EDTA, (¹⁴C)linoleic acid and (¹⁴C)-monoacylglycerophosphorylethanolamine were produced from 1-acyl-2-(¹⁴C)linoleyl-3-glycerophosphorylethanolamine. After osmotic rupture of purified lysosomes the phospholipases (A₁ and A₂) were in the soluble fraction entirely. The two phospholipases were not inhibited to the same extent by increasing concentrations of Ca²⁺ or EDTA. Phospholipases A₁ and A₂ were separated by gel filtration on Sephadex G-200.

FATTY ACID ESTERIFICATION BY HOMOGENATES OF BOVINE LIVER AND ADIPOSE TISSUE. J.D. Benson and R.S. Emery (Dairy Dept., Mich. State Univ., East Lansing, Mich. 48823). *J. Dairy Sci.* 54, 1034-40 (1971). Glycerol-3-phosphate, adenosine triphosphate, coenzyme A, MgCl₂ and NaF were necessary cofactors for maximal esterification of palmitate-1-¹⁴C by homogenates of bovine liver and adipose tissue. Dithiothreitol enhanced esterification in liver, but not in adipose tissue homogenates, whereas esterification was stimulated by bovine serum albumin in adipose but not hepatic tissue. Both systems incorporated palmitate, stearate, oleate and linoleate into glycerides. Linoleate incorporation was scant by either system. Liver homogenates esterified a greater percentage of the fatty acids into phospholipids and mono-glycerides than did adipose tissue. Triglycerides accounted for 55% of the product in adipose tissue and 18 to 44% in liver.

BIOSYNTHESIS OF WAX ESTERS IN FISH. METABOLISM OF DIETARY ALCOHOLS. D.M. Sand, Jean L. Hehl and H. Schlenk (Hormel Inst., Univ. of Minn., Austin, Minn. 55912). *Biochemistry* 10, 2536-41 (1971). Dietary fatty alcohols are incorporated by the gourami (*Trichogaster cosby*) as alcohols and acids into the roe wax esters. The course of this incorporation was studied by feeding 1-³H- and 1-³H, U-¹⁴C-labeled palmityl and oleyl alcohols and by analyzing the lipids 24 hr after ingestion. Levels of incorporation into roe wax esters were 15-60% of ¹⁴C but only 0.5-4% of the ³H that had been offered. The ratios ³H/¹⁴C in the alcohols of those wax esters showed that the dietary alcohols had undergone extensive oxidation to the corresponding fatty acids and were then reduced again for esterification. Some direct esterification of dietary alcohols is indicated by a small amount of tritium in position 1 of the alcohols recovered but it did not exceed 16% of the ¹⁴C-labeled chains that were found as alcohols in the wax esters. Direct esterification may occur to a relatively greater extent in body wax esters which are present only in trace amounts. There an enrichment of ³H compared to ¹⁴C was observed which remains unexplained. Some of the tritium derived from labeled dietary alcohols is used for reduction of fatty acids since it is found in position 1 of alcohols other than the ones fed. Amounts of tritium similar to those in roe wax esters have entered the glycerol moiety of body triglycerides and phosphatidylcholines, likely by reduction of a triose phosphate. Very little tritium had been used for synthesis of lipid chains.

BIOSYNTHESIS OF ALKYLDIACYLGLYCEROLS AND TRIACYLGLYCEROLS IN A CELL-FREE SYSTEM FROM THE LIVER OF DOGFISH. D.C. Malins and J.R. Sargent (Pioneer Res. Lab., Nat'l. Marine Fisheries Serv., Nat'l. Oceanic and Atmos. Admin., Seattle, Wash. 98102). *Biochemistry* 10, 1107-10 (1971). A cell-free preparation of dogfish (*Squalus acanthias*) liver was incubated with oleic acid-1-¹⁴C and oleyl alcohol-9,10-³H to study the relative rates of biosynthesis of triacylglycerols and alkyldiacylglycerols. The biosynthesis of triacylglycerols greatly exceeded that of alkyldiacylglycerols. Furthermore, radioactivity from both precursors was significantly higher in acyl chains of alkyldiacylglycerols than in alkyl chains. Fatty alcohol is extensively oxidized to fatty acid in this system; whereas fatty acid is reduced to fatty alcohol to only a slight extent. Nevertheless, fatty alcohol is strongly favored over fatty acid as a precursor of the 0-alkyl bond of alkyldiacylglycerols. The data may reflect regulation of alkyldiacylglycerol metabolism by enzymes controlling the equilibrium fatty acid \rightleftharpoons fatty alcohol.

BILE ACIDS. ALLOCHOLIC ACID, A METABOLITE OF ALLOCHENOXYCHOLIC ACID IN BILE FISTULA RATS. M.M. Mui and W.H. Elliott (Dept. Biochem., St. Louis Univ. Schl. of Med., St. Louis, Mo. 63104). *J. Biol. Chem.* 246, 302-4 (1971). 3 β -³H-Allochenodeoxycholic acid was administered by stomach tube to rats with bile fistulas. Bile collected during the first 21 hours contained 53 to 59% of the administered ³H. After hydrolysis and separation of the free bile acids by acetic acid partition chromatography, 95% of the chromato-

graphed ^3H was recovered as the unchanged acid, but 0.3% and 4.5% appeared in two trihydroxy acid regions. At least 85% of the latter material was identified as allocholic acid. One of the metabolites in the former fraction appeared to be allohydrocholic acid. These results show a distinct difference in the metabolism of the 5 α - and 5 β -cholic acids and demonstrate for the first time the 12 α -hydroxylation of a cholic acid nucleus in the rat.

ADRENERGIC NEUROHUMORAL INFLUENCES ON CIRCULATION AND LIPOLYSIS IN CANINE OMENTAL ADIPOSE TISSUE. Kathryn Ballard and S. Rosell (Dept. Phar., Karolinska Inst, 104 01 Stockholm 60 Sweden). *J. Nutr.* 28, 389-95 (1971). Vascular resistance, capillary filtration coefficient (CFC) and changes in blood volume were determined in canine omental adipose tissue. Release of glycerol and free fatty acids (FFA) were measured. Stimulation of the sympathetic nerves (1 to 9 Hz) caused initial vasoconstriction, maintained for 1 to 3 minutes. At lower frequencies (1 to 3 Hz), the blood flow increased gradually after the initial decrease. At higher frequencies, the constriction often reverted to vasodilatation. The CFC was not changed or increased initially. The clearance rate of locally injected ^{125}I decreased in spite of a constant blood flow and blood volume was reduced initially. The release of glycerol and FFA was increased by nerve stimulation and by norepinephrine. It is concluded that sympathetic nerves are physiologically important for the regulation of vascular reactions and control of lipid metabolism in omental adipose tissue. Whether this is also the case for circulating catecholamines remains to be established.

OCCURRENCE OF EICOSADIENOIC ACIDS IN LIVER LIPIDS OF RATS FED PARTIALLY HYDROGENATED SOYBEAN FAT. P.O. Egwin and D.S. Sgoutas (Burnsides Res. Lab., Univ. of Illinois, Urbana, Ill. 61801). *J. Nutr.* 101, 307-14 (1971). Male rats were fed for 5 months diets containing 10% or 20% partially hydrogenated soybean fat. The liver lipids were extracted, fractionated and the fatty acid composition of the fractions determined. Among the fatty acids in the phospholipid fraction from rats fed the diet containing 20% partially hydrogenated fat were substantial amounts of eicosadienoic acids. The structures: 5,11-20:2; 6,11-20:2; 5,13-20:2; 8,13-20:2 and 8,11-20:2 have been suggested. Their appearance was accompanied by a decrease in the concentration of eicosatrienoic and eicosatetraenoic acids, indicating a hindrance in the interconversion of unsaturated fatty acids, in particular at the acyl desaturation step.

EFFECT OF DIETARY LINOLEIC ACID ON FATTY ACID COMPOSITION OF EGG YOLK, LIVER AND ADIPOSE TISSUE. W. Guenter, D.B. Bragg and P.A. Knodra (Dept. of Animal Sci. Univ. of Manitoba, Winnipeg, Man. 19, Canada). *Poultry Sci.* 50, 845-50 (1971). Laying hens were fed five levels of dietary corn oil (0, 0.94, 1.89, 3.77 and 7.55%) in order to test the effect of dietary linoleic acid on egg weight, yolk weight and fatty acid metabolism during an 84 day test period. Addition of corn oil, at the expense of coconut oil in the semi-purified diet was used to maintain an equal energy level in test diets. Different levels of dietary linoleic acid had no effect on egg yolk weight, however, egg weight increased with higher levels of dietary linoleic acid. Fatty acid patterns of egg yolk, liver and adipose tissue resembled the fatty acid pattern of test diets. Higher levels of dietary linoleic acid increased egg yolk and tissue linoleic acid. Oleic acid in yolk and tissue showed an inverse relationship to linoleic acid content. The same relationship was observed between linoleic acid and saturated fatty acid in yolk and tissue. Low dietary linoleic acid appeared to increase body synthesis and deposition of myristoleic and palmitoleic acid. Linoleic acid depletion in yolk and adipose tissue was observed during the first 14 days of feeding the 0.20% linoleic acid diet. Linoleic acid equilibrium was maintained between adipose tissue and egg yolk with 0.84% linoleic acid and a highly significant increase in linoleic acid of the yolk and adipose tissue resulted from feeding 5.33% dietary linoleic acid.

TIMING OF MUCOPEPTIDE AND PHOSPHOLIPID SYNTHESIS IN SPORULATING *BACILLUS MEGATERIUM*. D.W. Pitel and C. Gilvarg (Dept. of Biochem. Sci., Frick Chem. Lab., Princeton Univ., Princeton, N.J. 08540). *J. Biol. Chem.* 246, 3720-24 (1971). During sporulation, the processes of septation and envelopment create a double membraned structure (the forespore) which is destined to become the spore. In this paper we present biochemical data to support the hypothesis that septation and envelopment result from the continued synthesis of plasma membrane in the absence of cell wall

synthesis. Using a double auxotroph of *Bacillus megaterium* requiring both diaminopimelic acid and lysine, it has been possible to follow mucopeptide formation unambiguously by measuring incorporation of radioactive diaminopimelic acid. Membrane synthesis has been determined by measuring the incorporation of ^{14}C -ethanolamine or ^{14}C -acetate into phospholipid. Upon glucose exhaustion and the initiation of sporulation, vegetative cell mucopeptide synthesis ceased for a period of 3 hours. Approximately 1 hour after the cessation of mucopeptide synthesis, an accelerated rate of phospholipid synthesis was observed that continued for about 3 hours. This biochemical data was correlated with morphological changes observed during the initial stages of sporulation.

THE LOCATION OF THE 4-PRO-R PROTONS OF MEVALONIC ACID IN CHOLESTEROL. L.J. Mulheirn and E. Caspi (Worcester Found. for Exper. Biol., Shrewsbury, Mass. 01545). *J. Biol. Chem.* 246, 3948-52 (1971). Evidence is presented that a proton derived from the 4-pro-R position of mevalonic acid is located at C-20 of cholesterol. It is now clear that the 3 protons derived from 4-pro-R position of mevalonic acid of cholesterol are located at the 17 α , 20, and 24-pro-R positions as predicted.

BIOHYDROGENATION OF UNSATURATED FATTY ACIDS. Carol R. Kepler, W.P. Tucker and S.B. Tove (Depts. of Biochem. and Chem., N. Carolina Univ., Raleigh, N.C. 27607). *J. Biol. Chem.* 246, 2765-71 (1971). The specificity of linoleic acid isomerase from *Butyrivibrio fibrisolvens* for cis-9,cis-12-dienoic fatty acids with an ω chain length varying from 4 to 8 carbons has been examined. The enzyme was found to be highly specific for a straight chain fatty acid bearing an ω chain length of 6 carbon atoms. Stereospecific addition of hydrogen to carbon atom 13 of linoleic acid in the D configuration was demonstrated. It was deduced that the substrate was bound to the enzyme in the form of a loop and that mechanism of isomerization involves either the protonation of an enzyme-bound carbanion or a concerted reaction. A tentative model with the carboxyl oxygens of the substrate participating in the isomerization reaction is proposed.

ON THE MODE OF ACTION OF LIPID-LOWERING AGENTS. M.E. Maragoudakis (Ciba Pharma. Co., Summit, N.J. 07901). *J. Biol. Chem.* 246, 4046-52 (1971). Cultured mammary cells, prepared from lactating rat mammary glands by treatment with collagenase, synthesize extensively cellular and secretory lipids, mostly triglycerides, from exogenous acetate or glucose via malonyl coenzyme A as an obligatory intermediate. Certain hypolipidemic drugs of clinical interest, which act as inhibitors of acetyl-CoA carboxylase in vitro drastically reduce lipogenesis in mammary cells from 1- ^{14}C -acetate or U- ^{14}C -glucose in a dose-dependent fashion. The effect is most pronounced in the triglyceride fraction of the cell lipids, but other lipid classes including cholesterol are also reduced. Production of $^{14}\text{CO}_2$ and incorporation of radioactivity from 1- ^{14}C -acetate into nonlipid material, cellular or secretory, are unaffected by the presence of the drugs in the culture media. Lipogenic capacity of the cells utilizing 1,3- ^{14}C -malonyl-CoA is not altered by the drugs; thus the derangement of fatty acid synthesis occurs before malonyl-CoA formation. Exogenous citrate in the culture medium can, under certain conditions, prevent or reverse the drastic inhibition of lipogenesis caused by the hypolipidemic drugs. Competition of these structurally unrelated compounds at the cellular level is in agreement with the in vitro competition between the hypolipidemic drugs and citrate, the activator of acetyl-CoA carboxylase.

THE METABOLIC EFFICIENCY OF ENERGY UTILIZATION OF GLUCOSE, SOYBEAN OIL AND DIFFERENT ANIMAL FATS BY GROWING CHICKS. G.D. Groote, N. Reyntens and J.A. Gali (Gov. Res. Station for Small Stock Husbandry, Merelbeke, Belgium). *Poultry Sci.* 50, 808-19 (1971). An experiment was conducted to study the metabolic energy efficiency in growing chicks of glucose, soybean oil and different animal fats, including a brown grease with a high quantity (25%) of free fatty acids. The metabolic efficiency of M. E. utilization for maintenance and growth together was higher for the different fats compared to glucose. The relative net availabilities were respectively 103.1% for lard, 102.9% for degummed soybean oil, 107.3% for fancy tallow, 108.1% for prime tallow and 102.9% for brown grease. The efficiencies were significantly higher for the two tallows relative to glucose. Conversion rates could not be found to be significantly different among the different fats. Recognizing the influence of observed variability in carcass

fat deposition as percentage of digested test fat intake on the availability of the M. E. of the different fats, it could be concluded that no important differences exist between the commonly used vegetable and animal fats soybean oil, lard, fancy and prime tallow and brown grease, concerning the utilization of their M. E. for maintenance plus growth by chicks. Tissue energy retention per unit M. E. intake was also significantly higher for the high fat diets (37.1-40%), compared to the low fat diets (33.2-34.7%). A significantly higher tissue energy gain on the prime tallow diet (40%) relative to the lard and soybean oil diets (37.1 and 37.7% respectively) was obtained, suggesting that tallow might be more efficiently utilized for growth than lard and soybean oil. Additional experiments however appear necessary to be able to draw a definite conclusion.

EFFECT OF ALPHA-TOCOPHEROL ADMINISTRATION ON RED CELL SURVIVAL IN VITAMIN E-DEFICIENT HUMAN SUBJECTS. P.J. Leonard and M.S. Losowsky (Dept. of Med., St. James's Hosp., Leeds, LS9 7TF, England). *Am. J. Clin. Nutr.* 24, 388-93 (1971). The effect of α -tocopherol therapy on red cell survival using ^{51}Cr -tagged cells was studied in eight subjects with evidence of vitamin E deficiency. In five of the eight subjects the survival curve was fitted significantly better by two straight lines than by one. Of the remaining three subjects, the $T_{0.5}$ was initially within the normal range in one; the value was only just below normal in the second; and in the third, a normal plasma vitamin E level was not achieved by therapy. Before the start of therapy the $T_{0.5}$ for ^{51}Cr in all eight subjects ranged from 10 to 28 days with a mean of 19.2 days. Following therapy, the values ranged from 19 to 30 days with a mean of 24.9 days, which is significantly different from the pretreatment value ($P < 0.025$).

THE METABOLISM OF GLYCERIDE GLYCOLIPIDS. R.T. Ambron and R.A. Pieringer (Dept. of Biochem., Temple Univ. School of Med., Philadelphia, Penn. 19140). *J. Biol. Chem.* 246, 4216-25 (1971). Particulate enzyme preparations of *Streptococcus faecalis* ATCC 9790 catalyze the conversion of diglycosyl diglyceride to a new type of phosphoglycolipid which has been identified as an acylated derivative of 3-O-(2-O- α -D-glucopyranosyl-6-O-phosphoryl-3'(1')-O-sn-glycerol)- α -D-glucopyranosyl-sn-glycerol. The characterization of the lipid was greatly facilitated after the development of methods which specifically labeled with radioisotopes either the glucose, glycerol or phosphate portions of the molecule. Periodate oxidation and differential hydrolysis catalyzed by base, acid and enzyme (α -glucosidase) of the specifically labeled compound were the major techniques used in the identification. The deacylated water-soluble derivative of the lipid yielded a hexose-glycerol-phosphate ratio of 2.0:2.08:1.05. Analysis of the intact lipid yielded a fatty acid ester-phosphate-hexose ratio of 3.87:1.0:1.82. However, the fatty acid ester content of four is considered tentative because of certain difficulties inherent to the procedures used to isolate and purify the intact lipid. Periodate oxidation of the lipid released no formaldehyde, indicating that at least one of the fatty acids is esterified to the glycerophosphate moiety of the lipid.

STIMULATION OF PHOSPHOLIPID EXCHANGE BETWEEN MITOCHONDRIA AND ARTIFICIALLY PREPARED PHOSPHOLIPID AGGREGATES BY A SOLUBLE FRACTION FROM LIVER. D.B. Zilversmit (Grad. Schl. of Nutr., Section of Biochem. and Molecular Biol., Div. of Biological Sci., Cornell Univ., Ithaca, N.Y. 14850). *J. Biol. Chem.* 246, 2645-49 (1971). Exchange of phospholipids between chylomicrons, artificial fat emulsions, phosphatidylcholine liquid crystals (liposomes) and mitochondria was observed. Exchange of phosphatidylcholine also occurred, in the absence of a biological membrane fraction, between liposomes and a fat emulsion stabilized with phospholipid. A protein fraction in the soluble portion of a rat liver homogenate accelerated phospholipid exchange in all systems from 3- to 30-fold. Exchange of phosphatidylcholine between labeled liposomes and unlabeled mitochondria showed a roughly proportional increase with the amount of cytosol protein added. This system appears suitable and convenient for the assay of the phospholipid exchange-stimulating fraction.

EFFECTS OF SIMPLE DIETS ON CHOLESTEROL SYNTHESIS IN RAT LIVER. E.B. Terrell and D.M. Regen (Dept. of Physiol., Vanderbilt Univ. Med. School, Nashville, Tenn. 37203). *J. Nutr.* 101, 437-44 (1971). Judging from the incorporation of $2\text{-}^{14}\text{C}$ -acetate into cholesterol by perfused rat livers, the relative rates of hepatic cholesterol synthesis under several

conditions were as follows: 1) fed laboratory ration throughout, 100; 2) fasting 1 day, 19; 3) fasting with bile fistula 1 day, 81; 4) fasting 2 days, 11; 5) fed a calorie-free diet (cellulose) 2 days, 9; 6) fed the calorie-free diet with 3% cholestyramine 2 days, 174; 7) fed corn oil at 1 kcal/g (in cellulose) 2 days, 17; 8) fed sucrose and starch at 0.25 and 0.75 kcal/g (in cellulose) 2 days, 43; 9) fed casein, bovine serum albumin, and sodium glutamate at about 0.48, 0.47 and 0.06 kcal/g, respectively, 2 days, 55. It is seen that enhanced removal of secreted bile steroids from the enterohepatic circulation (3 vs. 2, 6 vs. 5) is a potent stimulus to cholesterol synthesis. However, 1 day of bile drainage was no greater stimulus than continued laboratory ration feeding (3 vs. 1), suggesting that factors other than escape of bile steroids from the enterohepatic circulation can influence steroid turnover. The flow of bile through the intestines did not stimulate synthesis (5 vs. 4) showing that mechanical events associated with feeding are not entirely responsible for the effects of the laboratory diet; i.e., chemical properties of the diet are essential to the feeding effect (1 vs. 2 or 4). Dietary carbohydrate and protein both seem to contribute to the stimulatory effect of a laboratory diet (8 and 9 vs. 5). The role of fat in this respect appears to be very different from the other two nutrients (7 vs. 8 and 9).

ISOLATION AND CHARACTERIZATION OF MULTIPLE FORMS OF HYDROXYINDOLE-0-METHYLTRANSFERASE. R.L. Jackson and W. Lovenberg (Experi. Therapeutics Branch, National Heart and Lung Inst., National Inst. of Health, Bethesda, Md. 20014). *J. Biol. Chem.* 246, 4280-85 (1971). Hydroxyindole-0-methyltransferase has been isolated in pure form from bovine pineal glands. The enzyme has a molecular weight of about 76,000 to 78,000 but also occurs as higher molecular weight aggregates. The enzyme also exists as two differently charged molecular species which can be separated on DEAE-Sephadex. These two forms are identical in all other chemical and physical properties. Amino acid analysis reveals a relatively high leucine content (15% of the residues). Gel electrophoresis of the reduced enzyme in sodium dodecyl sulfate and dithiothreitol indicates the enzyme has two subunits with a monomer molecular weight of 39,000 which appear to be identical by fingerprint analysis.

DIETARY AND HORMONAL EFFECTS ON EXTENDED LACTATION AND LIPID METABOLISM IN RATS. R.E. Emery J.D. Benson and H.A. Tucker (Dairy Dept., Mich. State Univ., East Lansing, Mich. 48823). *J. Nutr.* 101, 831-38 (1971). To study the influence of extra dietary protein and fat on prolonged lactation and the mechanism whereby glucocorticoid maintains lactation, litters were replaced at days 16 and 24 of lactation with 8-day old foster litters. Control (22% protein), 41% protein and 50% fat diets, were fed throughout lactation. Litter weight gains were greater with the normal diet between days 8 and 16 of lactation but the high fat diet supported 59% greater gains than the control diet between days 16 and 32 of lactation. Fat deposition accounted for about 75% of this extra litter weight gain and the caloric efficiency of the 50% fat diet during prolonged lactation was 1.5 times greater than with the control diet. Daily subcutaneous injections of 50 μg 9-fluoroprednisolone acetate into the mothers between days 16 and 32 lactation increased litter weight gains by 21 and 52% with the three rations. This increased litter weight could be attributed largely to increased loss of body substance from the mothers. Both the high fat diet and exogenous glucocorticoid helped maintain mammary size, protein content, protein synthesizing activity and glyceride synthesizing activity although these effects were not uniformly significant. It is concluded that a high fat diet and exogenous glucocorticoid augment prolonged lactation in a largely independent and additive manner.

AFLATOXIN AS A POSSIBLE CAUSE OF FATTY LIVER SYNDROME IN LAYING HENS. P.B. Hamilton and J.D. Garlich (Dept. of Poultry Sci. and Dept. of Microbiol., N. Carolina State Univ., Raleigh, N.C. 27607). *Poultry Sci.* 50, 800-4 (1971). A cause has not been established previously for the fatty liver syndrome of laying hens. This disease is characterized by pale, friable livers with a lipid content about double that of the liver of normal hens and is accompanied by a marked reduction in egg production. An experiment was designed to establish whether dietary aflatoxin can induce the fatty liver syndrome. Groups of eight hens selected for similar age, weight and egg production were fed graded doses of aflatoxin for three weeks. Aflatoxin decreased

egg production in a dose related manner. It decreased the mean egg weight but it had no significant effect of egg shell thickness or percent of egg as shell. Necropsy revealed that the liver weight relative to the body weight was increased, but the relative weights of the spleen and pancreas were unaffected. Analysis of the liver for total lipid revealed that the mean lipid content as percent of dry liver weight was 36, 33, 43, 59, 55 and 55 for the hens consuming diets containing 0.0, 1.25, 2.5, 5.0, 10, and 20 μg of aflatoxin per gram, respectively. These results suggest that dietary aflatoxin can cause a fatty liver syndrome in laying hens.

MEDIUM-CHAIN TRIGLYCERIDES AS STRESS AGENTS IN VITAMIN K DEFICIENCY. P. Griminger and H. Fisher (Dept. of Nutr., Rutgers Univ., N. Brunswick, N.J. 08903). *Poultry Sci.* 50, 707-10 (1971). Growing, as well as adult, chickens showed more severe symptoms of a vitamin K deficiency when their diet contained medium chain triglycerides (MCT) than when it contained corn oil. Their vitamin K status in the presence of MCT was similar to that obtained with an essentially fat-free (glycerol-containing) diet. Since diets containing MCT are more palatable to chickens than those not containing any dietary fat, MCT may be a useful aid where residual vitamin K activity in the feed or a degree of intestinal synthesis make it difficult to deplete chickens of this vitamin.

METABOLIC STUDIES ON RETINOIC ACID IN THE RAT. P.R. Sundaresan and H.N. Bhagavan (Lipids Lab., Res. Inst., St. Joseph Hosp., Lancaster, Pa. 17604). *Biochem. J.* 122, 1-4 (1971). The nature of metabolites in the urine arising from differentially labelled retinoic acid was investigated after injection of physiological doses into retinol-deficient rats. Distribution of radioactivity after partition of urine into ether-soluble, acidic and water-soluble fractions revealed that there were at least six metabolites in urine. Of these, the major metabolite(s) was one lacking both C-14 and C-15 of retinoic acid. Enzymic or alkaline hydrolysis of acidic and water-soluble fractions did not release any retinoic acid, thus indicating that retinoyl β -glucuronide was not present in urine in significant amounts.

VITAMIN D₃: INDUCTION OF CALCIUM-BINDING PROTEIN IN EMBRYONIC CHICK INTESTINE IN VITRO. R.A. Corradino and R.H. Wasserman (Dept. of Phys. Bio., New York State Vet. College, Cornell Univ., Ithaca, N.Y. 14850). *Science* 172, 731-33 (1971). Induction of the synthesis of calcium-binding protein in chick embryonic intestine maintained *in vitro* was accomplished by simply adding vitamin D₃ to the culture medium. Accompanying the induction of this protein, there was enhanced radiocalcium uptake by the intestine. These observations represent the first demonstration of an *in vitro* physiological effect of vitamin D₃ on the calcium absorptive mechanism of the intestine.

POST-HEPARIN LIPOLYTIC AND MONOGLYCERIDASE ACTIVITIES IN FASTED MAN. D.L. Arons, P.H. Schreiber and R.A. Arky (Thorndike Memorial Lab. and Harvard Medical Unit, Boston City Hosp. and Dept. of Med., Harvard Med. School, Boston, Mass. 02118). *Pro. Soc. Exp. Biol. Med.* 137, 780-82 (1971). Prolonged fasting in man causes a significant decrease in post-heparin lipolytic and monoglyceridase activities. Activity is restored after refeeding. Plasma triglycerides declined in the face of diminished clearing factor lipase.

GANGLIOSIDES, GLYCOSIDASES AND SIALIDASE IN THE BRAIN AND EYE OF DEVELOPING CHICKENS. Cara-Lynne Schengrund and A. Rosenberg (Dept. of Biol. Chem., Pennsylvania State Univ., Hershey, Penn., 17033). *Biochemistry* 10, 2424-28 (1971). Ganglioside patterns and the activities of glycosidases and sialidase were followed in eye and brain during embryonic development of the chicken beginning with the 5-day-old embryo. Acetone powders were used for enzyme assays. Gangliosides were isolated by Folch extraction and partition dialysis. In early embryonic brain and eye, monosialoganglioside was predominant and disialoganglioside was a relatively minor ganglioside component. Trisialoganglioside was present in brain but not in eye. With embryonic development, the relative amounts of monosialoganglioside decreased and disialoganglioside increased until disialoganglioside became the predominant fraction in adult eye as well as brain.

EVALUATION OF CAROTENOID CONCENTRATION IN CHICKEN TISSUES. H.A. Stone, W.M. Collins and W.E. Urban Jr. (Dept. of Animal Sci., Univ. of New Hampshire, Durham, N.H. 03824). *Poultry Sci.* 50, 675-80 (1971). The purpose of this study was two-fold. The first was to evaluate the

relationship between subjective measurements of shank pigmentation intensity and objective measurements of total carotenoid concentration of chicken tissues. The second was to choose one tissue which, from objective measurement of carotenoid concentration and other available data, appeared to merit genetic study. To accomplish these objectives, one hundred and eighty meat-type chickens were categorized into age, hatch, type of housing and sex sub-groups. They were weighed, scored for shank pigmentation intensity, and sampled for total carotenoid concentration of the blood, skin, fat and liver, and vitamin A concentration of the liver. The data were analyzed statistically to determine the repeatability of duplicate determinations of carotenoid concentration, the product-moment correlations between shank score and carotenoid concentration. An analysis of variance was used to evaluate the importance of age, hatch, housing and sex upon the variation in the carotenoid concentration of the various tissues studied. The repeatability of duplicate determinations of carotenoids in blood were above 0.90, but for the other tissue traits were lower and ranged from approximately 0.45 to 0.85. The analysis of variance indicated that one or more of the following factors (age, hatch, housing, sex and some of the first order interactions) significantly influenced the variation in carotenoid concentration of each tissue. Blood at 8 weeks of age was selected as the most appropriate tissue for further genetic studies of total carotenoid concentration.

THE METABOLISM OF PHOSPHATIDYLINOSITOL IN THE THYROID GLAND OF THE PIG. F.B. Jungalwala, N. Freinkel and R.M.C. Dawson (Dept. of Biochem., Agr. Res. Council Inst. of Animal Physio., Babraham, Cambridge, U.K.). *Biochem. J.* 123, 19-33 (1971). The metabolism of phosphatidylinositol in pig thyroid has been investigated as a basis for understanding the specific stimulation of the synthesis of this phospholipid in the gland by thyrotropin. The gland contained an active Ca²⁺-dependent phosphatidylinositol-splitting enzyme with an optimum pH of 5.3-5.5. The major water-soluble product (65%) formed by this catabolic enzyme was not phosphorylinositol but a related compound, which may be a cyclic phosphorylinositol. Both this and phosphorylinositol (35%) were released simultaneously from the phosphatidylinositol substrate. The phosphatidylinositol-splitting enzyme was found almost exclusively in the supernatant fraction obtained by homogenization of the gland. It is postulated that the phosphatidylinositol is split into inositol and a phosphorus-containing lipid precursor of the phospholipid that remains on the microsomal membrane and is recycled. Isolated thyroidal mitochondria synthesized phosphatidylinositol from (2-³H)inositol only because of their contaminating microsomal component.

EFFECT OF DIETARY SUPPLEMENTS OF VITAMINS A, D AND E ON BODY BURDENS OF DDT IN THE RAT. W.E.J. Phillips, G.V. Hatima, D.C. Villemeuve and D.L. Grant (Res. Lab., Food and Drug Directorate, Dept. of Nat. Health and Welfare, Ottawa, Canada). *J. Agr. Food Chem.* 19, 780-84 (1971). In a study of nutrient-pesticide interactions, the rate of depletion of DDT in adipose tissue of the rat was measured when the diets contained various levels of supplemental vitamin A, D or E. The vitamin A status of the rat did not influence the rate of depletion or metabolism of DDT. Similarly, vitamin D and the supplementation of a nutritionally adequate diet (laboratory cubes) with vitamin E did not hasten the depletion of body burdens of DDT. The use of dietary supplements of fat-soluble vitamins does not appear to have practical significance for decreasing body burdens of DDT.

DISRUPTION OF LOW- AND HIGH-DENSITY HUMAN PLASMA LIPOPROTEINS AND PHOSPHOLIPID DISPERSIONS BY 1-ANILINONAPHTHALENE-8-SULFONATE. R.A. Muesing and T. Nishida (Univ. of Ill., Urbana, Ill. 61801). *Biochemistry* 10, 2952-62 (1971). A hydrophobic probe, 1-anilinonaphthalene-8-sulfonate (ANS), at high concentrations was found to alter profoundly the structure of both low-density lipoproteins of the S₁ 0-10 class and high-density lipoproteins of d 1.063-1.21. The disruption of these lipoproteins by 0.1 M NH₄ANS yielded essentially lipid-free proteins and phospholipid-rich and neutral lipid-rich fractions. Although all of the fractions obtained by the disruption of the lipoproteins were initially stabilized by association with ANS, the neutral lipid-rich fraction aggregated gradually upon loss of the ANS-stabilized phospholipid components. Lecithin dispersions were also disrupted extensively in 0.1 M ANS yielding an ANS-

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