American Oil Chemist's Society

60th

ANNUAL MEETING

PROGRAM

APRIL 20-24, 1969

San Francisco Hilton Hotel

San Francisco, California

MONDAY MORNING—APRIL 21

10:00 A.M.—Ballroom 6

SESSION A—SOAPS AND DETERGENTS I

Chairman—**I. R. Schmolka,** Wyandotte Chemicals, Wyandotte, Michigan

10:00 INTRODUCTORY REMARKS

10:05 **1. NEW BUILDING HARD SURFACE CLEANING PROBLEMS**
Alan W. Leipnitz, Economics Laboratory Inc.

10:25 **2. DETERGENTS—HARD WATER INTERACTION IN MACHINE DISHWASHING**
Robert F. Vance, General Electric Company

10:45 **3. ANALYSIS OF DATA FROM DETERGENCY TESTS WITH A NATURAL SOIL**
J. R. Trowbridge, Colgate-Palmolive Company

11:05 **4. A TRIPLY LABELED PARTICULATE SOIL FOR DETERGENCY STUDIES**
B. E. Gordon and W. T. Shebs, Shell Development Company

11:25 **5. STAINS—FABRICS—DETERGENTS**
B. W. Terry and W. L. Groves, Continental Oil Company

MONDAY MORNING—APRIL 21

10:00 A.M.—Ballroom 7 and 8

SESSION B—BIOCHEMICAL I

Chairman—**James F. Mead,** University of California, Los Angeles, Calif.

10:00 INTRODUCTION

10:10 **6. CRITICAL MICELLE CONCENTRATION (CMC) OF DIHYDROXY AND TRIHYDROXY BILE SALTS—EFFECTS OF COUNTERION AND TEMPERATURE**
Martin C. Carey and Donald M. Small, Boston University School of Medicine

10:30 **7. RAT LIVER LIPIDS: METABOLIC RELATIONSHIPS DERIVED FROM STRUCTURAL ANALYSES OF NEUTRAL GLYCERIDES AND PHOSPHOGLYCERIDES**
Randall Wood and R. D. Harlow, Oak Ridge Associated Universities

10:50 **8. TUMOR LIPIDS: METABOLIC RELATIONSHIPS DERIVED FROM STRUCTURAL ANALYSES OF ACCL, ALKYL AND ALK-1-ENYL MOIETIES OF NEUTRAL GLYCERIDES AND PHOSPHOGLYCERIDES**
Randall Wood and Fred Snyder, Oak Ridge Associated Universities

11:10 **9. METABOLISM OF LONG CHAIN FATTY ALCOHOLS: FORMATION OF WAXES AND GLYCERYL ETHERS**
Fred Snyder, Boyd Malone and James Soodsma, Oak Ridge Associated Universities

MONDAY MORNING—APRIL 21

10:00 A.M.—Ballroom 3

SESSION C—CHEMOTAXONOMY I

Chairman—**Ivan A. Wolff,** Northern Regional Research Laboratory, USDA, Peoria, Ill.

10:00 INTRODUCTION BY CHAIRMAN

10:10 **10. SEED LIPIDS AND CHEMOTAXONOMY**
Arthur S. Barclay and Quentin Jones, Crops Research Division, USDA, Ivan A. Wolff

10:40 **11. TAXONOMIC PATTERNS IN THE LIPIDS OF PHOTOSYNTHETIC TISSUE**
B. W. Nichols and R. J. B. Wood, Unilever Research Laboratories, Bedford, England

11:10 **12. THE DISTRIBUTION OF STEROLS, ALKALOIDS AND FATTY ACIDS IN SENITA CACTUS OVER ITS RANGE IN SONORA, MEXICO**
Henry W. Kircher, University of Arizona

MONDAY MORNING—APRIL 21

10:00 A.M.—Ballroom 9

SESSION D—OILSEED MEAL AND PROTEIN

Chairman—**T. J. Potts,** Ralston Purina Co., St. Louis, Mo.

10:00 INTRODUCTION BY CHAIRMAN

10:10 **13. LIPID CONTENT AND FATTY ACID ANALYSIS OF THREE PURE VARIETIES OF OATS (AVENA SATIVA) GROWN UNDER VARIOUS AGRICULTURAL PRACTICES**
George C. Potter and Joy Cary, The Quaker Oats Co.

10:30 **14. CONTINUOUS EXTRUSION COOKING OF COTTONSEED KERNELS AND PARTIALLY DEFATTED MEAL**
S. P. Clark, Texas A & M University

10:50 **15. REACTION OF GOSSYPOL WITH AMINO ACIDS, PEPTIDES AND OTHER AMINO-COMPOUNDS—PREPARATION, PURIFICATION AND CHARACTERIZATION**
Carl M. Cater and Carl M. Lyman, Texas A & M University

11:10 **16. CHEMICAL INACTIVATION OF CYCLOPROPENOID FATTY ACIDS IN COTTONSEED MEALS**
H. J. O'Neill and H. G. Reilich, IIT Research Institute, J. Proctor and W. A. Pons, Jr.

MONDAY MORNING—APRIL 21

11:00 A.M.—Ballroom 1 and 2

SESSION E—OZONOLYSIS

Chairman—**O. S. Privett,** Hormel Institute, University of Minnessota, Austin, Minn.

10:30 **17. STRUCTURAL ANALYSIS OF FATTY ACIDS VIA OZONOLYSIS (COLOR MOVIE)**
O. S. Privett, Hormel Institute

11:15 **18. MICRO-OZONIZER: AN APPARATUS FOR OZONOLYSIS**
Nicholas Pelick, Supelco Inc.

MONDAY AFTERNOON—APRIL 21

2:00 P.M.—Ballroom 6

SESSION F—SOAPS AND DETERGENTS II

Chairman—**Charles P. McClain,** Purex Corp. Ltd., Willmington, Calif.

2:00 **19. AUTOMATED METHOD FOR THE DETERMINATION OF TOTAL ENZYME IN PRESOAK DETERGENTS**
L. M. Paixao, E. J. Maitheny and C. Benz, Colgate-Palmolive Research Center

2:20 **20. FLUORESCENT WHITENING AGENTS FOR ENZYME-CONTAINING LAUNDRY PRODUCTS**
Per S. Stensby, William R. Findley and Charles W. Liebert, Geigy Chemical Corp.

2:40 **21. AN AUTOMATED DETERMINATION OF LINEAR ALKYL SULFONATES IN SPRAY DRIED DETERGENTS**
E. H. Brandli and R. M. Kelley, Colgate-Palmolive Co.

3:00 INTERMISSION

3:10 **22. THE COURSE OF BIODEGRADATION OF ANIONIC DETERGENTS**
T. C. Cordon, E. W. Maurer and A. J. Stirton, Eastern Utilization Research and Development Division, USDA

3:30 **23. N→O ACYL MIGRATION STUDIES OF N-(2-HYDROXYETHYL)LAURAMIDES**
J. A. Loboda and T. M. Muzyczko, The Richardson Co.

MONDAY AFTERNOON—APRIL 21

2:00 P.M.—Ballroom 7 and 8

SESSION G—BIOCHEMICAL II

Chairman—**F. Lee Avera,** Corn Products Co., Alameda, Calif.

2:00 **24. LIPIDS FROM THE MICROSOMAL, MITOCHONDRIAL AND MYELIN FRACTIONS OF MOUSE BRAIN**
Grace Y. Sun, Cleveland Psychiatric Institute, and Lloyd A. Horrocks

2:20 **25. LIPID CONTENT AND FATTY ACID PATTERNS IN DEVELOPING STEELHEAD SAC FRY**
Lyle Hayes, Ian J. Tinsley, Robert R. Lowry and Gary A. Chapman, Oregon State University

2:40 **26. HYDROCARBON BIOSYNTHESIS BY COCKROACH INTEGUMENT**

Charles W. Conrad and Larry L. Jackson, Montana State University

3:00 INTERMISSION

3:10 27. **STUDIES ON THE METABOLIC FATE OF GOSSYPOL IN THE RAT USING ^{14}C LABELED GOSSYPOL**
Mohamed Bahi Abou-Donia and Carl M. Lyman, Texas A & M University

3:30 28. **CHANGES IN FATTY ACID AND LIPID CLASS COMPOSITION OF SOYBEANS DURING MATURATION**
O. S. Privett, R. A. Gross and K. Beutel, Hormel Institute

3:50 29. **EFFECTS OF PARATHION ON LIPOLYSIS IN ISOLATED ADIPOSE CELLS**
Richard M. Caley and Robert G. Jensen, University of Connecticut

MONDAY AFTERNOON—APRIL 21

2:00 P.M.—Ballroom 3

SESSION H—CHEMOTAXONOMY II

Chairman—**Carter Litchfield,** Texas Agricultural Experiment Station, College Station, Texas

2:00 **INTRODUCTION**
Ivan A. Wolff, Northern Regional Research Laboratory, USDA

2:10 30. **PHYLOGENERIC PATTERNS IN THE FATTY ACIDS OF AQUATIC ORGANISMS**
R. G. Ackman, Fisheries Research Board of Canada, Halifax, N.S., Canada

2:40 31. **TAXONOMIC PATTERNS IN THE TRIGLYCERIDE COMPOSITION OF NATURAL FATS**
Carter Litchfield, Texas Agricultural Experiment Station

3:10 32. **PHYLOGENETIC RELATIONSHIPS IN LIPID METABOLISM**
James F. Mead, University of California

3:40 INTERMISSION

3:50 33. **A COMPARATIVE STUDY OF SPHINGOSINE BASES IN CENTRAL NERVE TISSUE**
Kathleen M. Gilliland and Ezio A. Moscatelli, University of Texas, Southwestern Medical School

4:20 34. **PHOSPHOLIPIDS AND GLYCOLIPIDS OF ANIMAL CELL MEMBRANES, ORGANELLES AND ORGANS: SPECIES AND AGE VARIATIONS**
George Rouser, City of Hope Medical Center

MONDAY AFTERNOON—APRIL 21

2:00 P.M.—Ballroom 9

SESSION I—SEMINAR: NON-FOOD USES OF COCONUT OIL—WHERE ARE WE HEADING?

Chairman—**N. O. V. Sonntag,** Glyco Chemicals, Inc., Williamsport, Penna.

Panel Members: Henry A. Mohteni, Drew Chemical Co., Karl T. Zilch, Emery Industries, Inc., Herman W. Zobel, Roger Williams T & E Services, Inc., Henry Fineberg, Ashland Chemical Co.

A panel discussion and seminar on growth and future of inedible uses of coconut oil and its fatty acids including review of trends, possible influence of synthetic lauric acid, price fluctuation, supply and economic factors. The recent increase in relative proportion of oil used edibly will be pinpointed. Future applications and research trends will also be discussed.

MONDAY AFTERNOON—APRIL 21

2:00 P.M.—Ballroom 1 and 2

SESSION J—SYMPOSIUM: NATURAL WAXES

Chairman—**Nicholas Nicolaides,** U.S.C. Medical School, Los Angeles, Calif.

2:00 **INTRODUCTORY REMARKS**

2:10 35. **BACTERIAL HYDROCARBONS: STRUCTURE AND BIOSYNTHESIS**
Phillip W. Albro and John C. Dittmer, St. Louis University Medical School

2:40 36. **PLANT WAXES**
P. E. Kolattukudy, Connecticut Agricultural Experiment Station

3:10 37. **THE CUTICULAR LIPIDS OF INSECTS**
Larry L. Jackson and Graeme L. Baker, Montana State University

3:40 INTERMISSION

3:50 38. **DETERMINATION OF THE COMPOSITION OF UNHYDROLYZED BEESWAX**
A. P. Tulloch, National Research Council of Canada

4:20 39. **THE OCCURRENCE, FUNCTION AND BIOSYNTHESIS OF WAX ESTERS IN MARINE ORGANISMS**
Judd C. Nevenzel, University of California

4:50 40. **WAXES OF ANIMAL SKIN SURFACES**
N. Nicolaides, Hwei C. Fu and M. N. A. Ansari, University of Southern California School of Medicine

TUESDAY MORNING—APRIL 22

9:00 A.M.—Ballroom 6

SESSION K—THE USE OF SURFACTANTS IN MINERAL RECOVERY

Chairman—**K. R. McKennon,** Dow Chemical Co., Walnut Creek, Calif.

9:00 41. **SULFONATE FLOTATION OF PHOSPHORITE AND COMMONLY ASSOCIATED MINERALS**
M. C. Fuerstenau and J. D. Miller, University of Utah, R. O. Juareghi

9:20 42. **THE ROLE OF SURFACTANTS IN THE FLOTATION OF MOLYBDENITE AT CLIMAX**
Richard A. Ronzio, Climax Molybdenum Co.

9:40 43. **THE EFFECTS OF SURFACTANTS AND ALCOHOLS ON THE FLOTATION COLLECTION PERFORMANCE OF AN ALKYL THIONOCARBAMATE**
D. J. Collins and T. F. Izzo, Dow Chemical Co.

10:10 44. **SOME OBSERVATIONS ON THE EFFECTS OF SURFACTANTS IN THE MICROBIOLOGICAL LEACHING OF LOW-GRADE SULFIDE ORES**
J. P. Kass, Technical Counsel for the Food and Chemical Industries

10:30 INTERMISSION

10:40 45. **EFFECT OF SURFACTANTS ON THE RHEOLOGY OF HEMATITE SLURRIES**
Harley Y. Jennings, Jr., Chevron Oil Field Research Co.

11:00 46. **SURFACTANTS FOR OIL RECOVERY**
G. P. Ahearn, Esso Production Research Co.

TUESDAY MORNING—APRIL 22

9:00 A.M.—Teakwood Room

SESSION L—CHEMICAL SYNTHESIS SESSION I

Chairman—**Kenneth E. Holt,** Experience, Inc., Minneapolis, Minn.

9:00 47. **N-PHENYLAMINOMETHYLATION: A ONE-STEP ROUTE TO N-SUBSTITUTED ANILINES FROM UNSATURATED FATTY DERIVATIVES**
Robert A. Grimm, Ashland Chemical Co.

9:20 48. **OLEYL ALCOHOL FROM ANIMAL FATS BY CATATYLIC HYDROGENOLYSIS**
R. S. Klonowski, T. W. Findley, C. M. Josefson and A. J. Stirton, Swift & Co.

9:40 49. **HF CATALYSIS I—A NOVEL SYNTHESIS OF GLYCEROL MONOESTERS**
Eugene J. Miller and Harlan E. Tiefenthal, Armour Industrial Chemical Co., and Ago Mais

10:00 50. **COMPETITIVE HYDROGENATION RATES OF ISOMERIC METHYL OCTADECENOATES**
C. R. Scholfield, T. L. Mounts, R. O. Butterfield and H. J. Dutton, Northern Regional Research Laboratory, USDA

10:20 51. **HOMOGENEOUS CATALYTIC CONJUGATION OF POLYUNSATURATED FATS BY METHYL BENZOATE-Cr $(CO)_3$**
E. N. Frankel, Northern Regional Research Laboratory, USDA

10:40 52. **REDUCTIVE AMINATION OF 12-KETOSTEARIC ACID**
Bernard Freedman and Glenn Fuller, Western Regional Research Laboratory, USDA

11:00 53. **THERMAL POLYMERIZATION OF SAFFLOWER SEED OIL**
Yeshwant K. Purandare, State University, A & T College

11:20 54. **CATALYTIC ISOMERIZATION OF SAFFLOWER SEED OIL**
Yeshwant K. Purandare, State University, A & T College

11:40 **55. URETHANE FOAMS FROM ANIMAL FATS: V. FLAME RESISTANT FOAMS FROM HYPOHALOGENATED GLYCERIDES**
F. Scholnick, E. J. Saggese, A. N. Wrigley and G. R. Riser, Eastern Regional Research Laboratory, USDA

TUESDAY MORNING—APRIL 22

9:00 A.M.—Ballroom 7 and 8

SESSION M—NUTRITION I

Chairman—**Ruth Okey,** University of California, Berkeley, Calif.

9:00 **56. EFFECTS OF DIET ON CHOLESTEROL METABOLISM IN RABBITS**
K. K. Carroll, University of Western Ontario, Canada

9:20 **57. DIETARY INDUCED CHANGES IN CANINE ERYTHROCYTE FATTY ACID COMPOSITION**
Antanas Butkus, L. Allen Ehrhart, Lena A. Lewis and F. Merlin Bumpus, Cleveland Clinic Foundation

9:40 **58. THE EFFECT OF DIETARY CHOLESTEROL ON THE COMPOSITION, MORPHOLOGY AND FUNCTIONS OF GUINEA PIG RED CELLS**
R. Ostwald, W. Yamanaka, J. Kroes and M. Light, University of California

10:00 **59. EFFECT .OF .DIETARY .FAT .LEVEL .AND CALORIC INTAKE ON BODY WEIGHT, DISTRIBUTION OF FAT DEPOTS AND FATTY ACID COMPOSITION OF CARCASS, LIVER AND ADIPOSE TISSUE OF RATS**
Mildred J. Bennett and Shirley Barber, Children's Hospital Medical Center

10:20 **60. NUTRITIONAL EVALUATION OF FILLED MILKS IN MONKEYS AND RATS**
Hans Kaunitz and Jaime Sanyer, College of Physicians and Surgeons

10:40 **61. SEX DIFFERENCES IN INTESTINAL PROTEIN SYNTHESIS AND ABSORPTION OF LIPIDS**
George V. Vahouny, M. Ito and C. R. Treadwell, George Washington University, Washington, D.C.

11:00 **62. EFFECT OF DIETARY STERCULIC ACID ON LIPID COMPOSITION OF RAT TISSUES**
Barry J. Burns, R. B. Alfin-Slater and James F. Mead, University of California

TUESDAY MORNING—APRIL 22

9:00 A.M.—Ballroom 9

SESSION N1—SYMPOSIUM: MEMBRANE MODEL SYSTEMS

Chairman—**G. Colacicco,** City University of New York, N.Y.
Co-chairman—**J. M. Steim,** Brown University, Providence, R.I.

9:00 **OPENING OF SYMPOSIUM**
G. Colacicco, City University of New York

9:05 **GENERAL INTRODUCTION**
F. S. Sjostrand, University of California

SESSION I—LIPIDS

Chairman—**H. T. Tien,** Michigan State University

9:25 **INTRODUCTORY REMARKS**
H. T. Tien, Michigan State University

9:40 **63. LIPID MONOLAYERS: INFLUENCE OF CHEMICAL STRUCTURE ON SURFACE PROPERTIES**
G. Colacicco, City University of New York

10:00 **DISCUSSION**

10:20 **64. BLM AS BIOLOGICAL MEMBRANE MODELS: AN EVALUTION AND A SUGGESTION FOR ITS SPONTANEOUS FORMATION IN NATURE**
H. T. Tien, Michigan State University

10:40 **DISCUSSION**

11:00 **INTERMISSION**

11:10 **65. EFFECT OF CHOLESTEROL ON PERMEABILITY AND ELECTRICAL PROPERTIES OF PHOSPHOLIPID MODEL MEMBRANES**
D. Papahadjopoulos and S. Ohki, State University of New York at Buffalo

11:30 **DISCUSSION**

11:50 **66. FRACTURE SURFACES IN BULK-PHASE LIPID SYSTEMS**
D. W. Deamer, University of California

12:10 **DISCUSSION**

TUESDAY AFTERNOON—APRIL 22

2:00 P.M.—Ballroom 9

SESSION N2—SYMPOSIUM: MEMBRANE MODEL SYSTEMS

Chairman—**G. Colacicco,** City University of New York, N.Y.
Co-chairman—**J. M. Steim,** Brown University, Providence, R.I.

INTERNATIONAL MEMBRANE WORKSHOP: LIPID-PROTEIN ASSOCIATION IN MEMBRANES—HYDROPHOBIC OR ELECTROSTATIC?

2:00 **OPENING OF WORKSHOP**
G. Colacicco, City University of New York, N.Y.

WORKSHOP SESSION I: MEMBRANE LIPIDS

Chairman—**D. Branton,** University of California, Berkeley, Calif.

2:05 **INTRODUCTORY REMARKS**
D. Branton, University of California

2:25 **DISCUSSION**

TUESDAY MORNING—APRIL 22

9:00 A.M.—Ballroom 1 and 2

SESSION O—PROCESSING OF FATS AND OILS

Chairman—**Frank E. Sullivan,** Frank E. Sullivan Co., Tiburon, Calif.

9:00 **67. FILTRATION IN THE FATS AND OILS INDUSTRY**
Robert J. Zilli, Johns-Manville Corp.

9:30 **68. DEODORIZATION OF FATS AND OIL IN COMMERCIAL PRACTICE**
George Kreutzer, Swift & Co., Chicago

10:00 **69. HYDROGENATION OF SOYBEAN OIL WITH COPPER-CHROMITE CATALYST: WINTERIZATION OF LOW-LINOLENATE OILS**
K. J. Moulton, R. E. Beal and E. L. Griffin, Jr., Northern Utilization Research and Development, USDA

10:30 **70. THE EFFECTS OF HYDROGENATION PROCESS VARIABLES ON THE SELECTIVITY AND ISOMERIZATION CHARACTERISTICS OF NICKEL CATALYSTS**
Robert R. Allen, M. C. Moore and J. E. Covey, Jr., Anderson, Clayton & Co.

11:00 **71. POSITIONAL AND GEOMETRICAL ISOMERIZATION DURING PARTIAL HYDROGENATION OF TRILINOLEIN—A COMPARISON OF COPPER AND NICKEL CATALYSTS**
E. R. Lowrey, Procter & Gamble, and Ehud Kirschner

WEDNESDAY MORNING—APRIL 23

9:00 A.M.—Teakwood Room

SESSION P—ANTIOXIDANTS

Chairman—**H. S. Olcott,** University of California, Berkeley, Calif.

9:00 **INTRODUCTION BY CHAIRMAN**

9:10 **72. PRESERVATION OF LIPIDS WITH MALIC ACID**
S. Edmund Berger, Casimir V. Krolewski and Leslie C. Wizemann, Allied Chemical Corp.

9:30 **73. STUDIES ON ANTIOXIDANT TREATMENTS OF CRUDE VEGETABLE OILS**
E. R. Sherwin and B. M. Luckadoo, Eastman Chemical Products

9:50 **74. EFFECTS OF HUMIDIFICATION ON ACTIVITY OF CATALYSTS AND ANTIOXIDANTS IN MODEL SYSTEMS**
K. H. Tjhio, T. P. Labuza and M. Karel, Massachusetts Institute of Technology

10:10 **75. EFFECTS OF NITROGEN BASES OF PHOSPHATIDYL CHOLINE AND PHOSPHATIDYL ETHANOLAMINE ON AUTOXIDATION OF METHYL LINOLEATE EMULSION**

Lloyd M. Smith and Lee-Shin Tsai, University of California

10:30 **76. THE INHIBITION OF OXIDATION BY AROMATIC AMINES**
K. U. Ingold, K. Adamic and D. F. Bowman, National Research Council of Canada

10:50 **77. ANTIOXIDANT PROPERTIES OF α-TOCOPHEROL DERIVATIVES AND RELATIONSHIP OF ANTIOXIDANT ACTIVITY TO BIOLOGICAL ACTIVITY**
W. A. Skinner and R. M. Parkhurst, Stanford Research Institute

11:10 **78. FERRIC IRON-CATALYZED REACTIONS OF BIOLOGICAL ANTIOXIDANTS WITH PREFORMED LIPID HYDROPEROXIDES**
E. H. Gruger, Jr., Bureau of Commercial Fisheries and A. L. Tappel

11:30 **79. ANTIOXIDANT PROPERTIES OF TOCOPHERAMINES**
H. S. Olcott and J. Van der Veen, University of California

WEDNESDAY MORNING—APRIL 23

9:00 A.M.—Ballroom 1 and 2

SESSION Q—CHEMICAL SYNTHESIS SESSION II

Chairman—**William E. Link,** Ashland Chemical Co., Minneapolis, Minn.

9:00 **80. ALIPHATIC NITROGEN DERIVATIVES: I. ADDITION OF N,N-DIBROMOSULFONAMIDES TO INTERNAL OLEFINS**
T. A. Foglia, E. T. Haeberer and G. Maerker, Eastern Regional Research Laboratory, USDA

9:20 **81. ALIPHATIC NITROGEN DERIVATIVES: II. REACTION OF CIS-9, 10-EPIMINOOCTA-DECANE WITH CARBOXYLIC ACIDS**
G. Maerker, E. T. Haeberer, E. T. Donahue and T. A. Foglia, Eastern Regional Research Laboratory, USDA

9:40 **82. FRIEDEL CRAFTS REACTION OF N-ALKENOIC ACIDS**
M. F. Ansell and G. F. Whitfield, Queen Mary College, University of London, London, E. 1., England

10:00 **83. DIOL DIRICINOLEATES FROM DIHALOALKANES**
C. K. Lyon and V. H. Garrett, Western Regional Research Laboratory, USDA

10:20 **84. ACRYLATE ESTERS OF LONG CHAIN HYDROXY ACYL CHLORIDES**
M. J. Diamond, Western Regional Research Laboratory, USDA

10:40 **85. PREPARATION OF SULFATE ESTERS BY CARBODIIMIDE-MEDIATED SULFATION**
Ralph O. Mumma, F. J. Vastola and C. P. Hoiberg, Pennsylvania State University

11:00 **86. STEREOSPECIFIC HYDRATION OF UNSATURATED FATTY ACIDS BY BACTERIA**
L. L. Wallen and E. N. Davis, Northern Regional Research Laboratory, USDA

11:20 **87. ALUMINUM CHLORIDE-CATALYZED ACYLATION REACTIONS USING ISOPROPENYL ESTERS AS ACYLATING AGENTS**
E. S. Rothman and G. G. Moore, Eastern Regional Research Laboratory, USDA

WEDNESDAY MORNING—APRIL 23

9:00 A.M.—Ballroom 7 and 8

SESSION R—SYMPOSIUM: THE LAURANCE W. KINSELL MEMORIAL SYMPOSIUM

Chairman—**R. B. Alfin-Slater,** University of California, Los Angeles, Calif.

9:00 **INTRODUCTION BY CHAIRMAN**

9:05 **88. THE CONTRIBUTIONS OF DR. LAURANCE W. KINSELL AND CO-WORKERS TO ATHEROSCLEROSIS RESEARCH**
Hugh Sinclair, Magdalen College, Oxford University, England

9:30 **89. CHOLESTYRAMINE AND POLYUNSATURATED FAT: SIMILARITY OF EFFECTS IN MAN**
Peter Wood, Highland General Hospital

10:00 **90. EFFECT OF HYDROGENATED FATS ON BLOOD CHOLESTEROL IN MAN**
Fred Mattson, The Procter & Gamble Co.

10:30 **91. DIETARY MANAGEMENT OF HYPERLIPEMIC STATES IN MAN**
Edwin L. Bierman, VA Hospital and University of Washington

11:00 **92. GRAPHIC PRESENTATION AND ANALYSIS OF COMPUTER-DERIVED SCHLIEREN LIPOPROTEIN DATA**
Lin C. Jensen, Thomas H. Rich and Frank T. Lindgren, University of California

11:30 **93. THE METABOLISM OF UNSATURATED FATTY ACIDS IN RATS FED DL-METHIONINE**
Richard L. Lyman, C. Giotas, M. A. Fosmire and P. Miljanich, University of California

12:00 **94. SOME RELATIONSHIPS OF POLYUNSATURATED FATTY ACID METABOLISM TO ATHEROSCLEROSIS**
James F. Mead and D. F. Haggerty, Jr., University of California

WEDNESDAY MORNING—APRIL 23

9:00 A.M.—Ballroom 9

SESSION N3—SYMPOSIUM: MEMBRANE MODEL SYSTEMS

SESSION II—PROTEINS AND LIPID-PROTEIN INTERACTIONS

Chairman—**S. J. Singer,** University of California, San Diego, Calif.

9:00 **INTRODUCTORY REMARKS**
D. W. Urry, American Medical Association

9:20 **95. USE OF OPTICAL ROTATION IN DETERMINING CONFORMATION OF PROTEIN WITHIN MEMBRANES**
D. W. Urry, American Medical Association

9:40 **DISCUSSION**

10:00 **96. PROTEIN CONFORMATION AND MEMBRANE STRUCTURE**
S. J. Singer, University of California

10:20 **DISCUSSION**

10:40 **INTERMISSION**

10:50 **97. TEMPERATURE DEPENDENCE OF THE CONFORMATION OF HUMAN SERUM LOW AND HIGH DENSITY LIPOPROTEINS**
A. M. Scanu, University of Chicago

11:10 **DISCUSSION**

11:30 **98. PHYSICAL STUDIES OF LIPID-POLYPEPTIDE AND LIPID-PROTEIN INTERACTIONS**
D. Chapman, Unilever, England

11:50 **DISCUSSION**

WEDNESDAY MORNING—APRIL 23

9:00 A.M.—Ballroom 6

SESSION S—ANALYTICAL I

Chairman—**James A. Thompson,** Basic Vegetable Products, Vacaville, Calif.

9:00 **99. PREPARATIVE FRACTIONATION BY FRONTAL COUNTERCURRENT DISTRIBUTION**
R. A. Barford, R. J. Bertsch and H. L. Rothbart, Eastern Utilization Research and Development Division, USDA

9:20 **100. AN UNUSUAL NITROGEN-CONTAINING LIPID FROM CORDIA VERBENACEAE SEED OIL**
K. L. Mikolajczak, D. S. Seigler, C. R. Smith, Jr. and I. A. Wolff, Northern Regional Research Laboratory, USDA

9:40 **101. NEW SOURCES OF 9-D-HYDROXY-CIS-12-OCTADECENOIC ACID**
R. G. Powell, R. Kleiman and C. R. Smith, Jr. Northern Regional Research Laboratory, USDA

10:00 **102. OXYGENATED TRANS-3-OLEFINIC ACIDS OF STENACHAENIUM SEED OIL**
R. Kleiman, G. F. Spencer, L. W. Tjarks and F. R. Earle, Northern Regional Research Laboratory, USDA

10:20 **103. CORRELATION OF SOLUBILITY DATA: III. THE ISOPLETH REFERENCE METHOD FOR PREDICTING SOLUBILITY DATA FOR LONG CHAIN HOMOLOGOUS AND ANALOGOUS COMPOUNDS**
August V. Bailey, James A. Harris and Evald L. Skau, Southern Utilization Research and Development Division, USDA

10:40 104. **KINETIC RATE CONSTANTS DETERMINED BY A DIGITAL COMPUTER**
R. O. Butterfield, Northern Regional Research Laboratory, USDA

11:00 105. **SOME PARAMETERS OF CUPRIC SALTS OF FATTY ACIDS WITH REFERENCE TO ANALYTICAL USAGE**
Robert R. Lowry and Ian J. Tinsley, Oregon State University

11:20 106. **ACOUSTIC CHARACTERISTICS OF SOME FATS AND OILS**
G. O. Hustad, T. Richardson, W. C. Winder and M. P. Dean, University of Wisconsin

WEDNESDAY AFTERNOON—APRIL 23

2:00 P.M.—Teakwood Room

SESSION T—CHROMATOGRAPHY IN LIPID ANALYSIS

Chairman—**Randall Wood,** Oakridge Associated University, Oakridge, Tenn.

2:00 107. **A SIMPLIFIED PREPARATIVE THIN LAYER CHROMATOGRAPHY OF PHOSPHOLIPIDS**
James M. Iacono and Terry T. Ishikawa, University of Cincinnati Medical College

2:20 108. **THE PHOSPHATIDES OF SAFFLOWER SEEDS AND THEIR CONTRIBUTION TO PIGMENT FORMATION OCCASIONALLY OCCURRING IN EXTRACTED OILS**
H. J. Burkhardt, Western Regional Research Laboratory, USDA

2:40 109. **GAS CHROMATOGRAPHIC EQUIVALENT CHAIN LENGTHS OF ISOMERIC METHYL OCTADECENOATES AND OCTADECYNOATS**
C. R. Scholfield and H. J. Dutton, Northern Regional Research Laboratory, USDA

3:00 110. **DETECTION OF UNUSUAL COMPONENTS BY DIRECT GAS LIQUID CHROMATOGRAPHY OF SEED OILS**
R. Kleiman, J. W. Hagemann, F. R. Earle and I. A. Wolff, Northern Regional Research Laboratory, USDA

3:20 **INTERMISSION**

3:30 111. **QUANTITATIVE DETERMINATION OF MONO- AND DIGLYCERIDES BY GAS LIQUID CHROMATOGRAPHY**
J. Blum and W. R. Koehler, Lever Brothers Co.

3:50 112. **COMBINED ULTRAMICRO DRY COLUMN CHROMATOGRAPHY AND MASS SPECTROMETRY OF LIPID CLASSES**
A. J. Bauman and Heinz G. Boettger, California Institute of Technology

4:10 113. **TECHNIQUES AND QUANTITATION OF PROGRAMMED TEMPERATURE GLC FOR DETECTING FATTY ACIDS PRESENT IN SUB-MICROGRAM AMOUNTS**
John L. Iverson, Food and Drug Administration

4:30 114. **ISOLATION AND DETERMINATION OF TRACE AMOUNTS OF FATTY AMINES AND RELATED COMPOUNDS**
L. D. Metcalfe, R. J. Martin and W. A. Wagner, Armour Industrial Chemical Co.

WEDNESDAY AFTERNOON—APRIL 23

2:00 P.M.—Ballroom 7 and 8

SESSION U—AFLATOXIN I

Chairman—**Walter A. Pons, Jr.,** Southern Regional Research Laboratory, New Orleans, La.

2:00 115. **DETERMINATION OF AFLATOXINS IN PEANUT SOAPSTOCKS**
A. F. Cucullu, L. S. Lee, W. A. Pons, Jr. and L. A. Goldblatt, Southern Utilization Research and Development Division, USDA

2:20 116. **RECOVERY OF AFLATOXINS FROM ARTIFICIALLY INOCULATED TOBACCO**
C. Y. Yang and F. F. Fannin, University of Kentucky

2:40 117. **DECOMPOSITION OF AFLATOXINS IN AQUEOUS SOLUTION: EFFECTS OF PH AND HEAT**
Harry W. Schroeder and Hugo Hein, Jr., Market Quality Research Division, USDA

3:00 118. **OXIDATION OF 6-METHOXYDIFUROCOUMARONE**
Mabry Wiley and Anthony C. Waiss, Jr., Western Utilization Research and Development Division, USDA

3:20 119. **STABILITY OF AFLATOXIN STANDARDS IN SOLUTION AND DRY FILM**
J. A. Robertson, W. A. Pons, Jr. and L. A. Goldblatt, Southern Utilization Research and Development Division, USDA

3:40 120. **A STUDY OF THE VARIABILITY ASSOCIATED WITH SAMPLING PEANUTS FOR AFLATOXIN**
Peter J. Tiemstra, Derby Foods, Inc.

4:00 121. **AFLATOXINS IN COTTONSEED HULLS**
M. E. Whitten, Market Quality Research Division, USDA

4:20 122. **SURVEY OF CORN FOR THE PRESENCE OF AFLATOXIN, ZEARALENONE AND OCHRATOXIN**
Odette L. Shotwell, C. W. Hesseltine and Marion L. Goulden, Northern Utilization Research and Development Division, USDA

4:40 123. **CHEMICAL INACTIVATION OF AFLATOXINS IN OILSEED MEALS**
G. E. Mann, L. P. Codifer, Jr., H. K. Gardner, Jr., F. G. Dollear and S. P. Koltun, Southern Utilization Research and Development Division, USDA

5:00 124. **ALTERNATIVES IN AFLATOXIN METHODOLOGY**
M. S. Masri and Jon R. Page, Western Utilization Research and Development Division, USDA

WEDNESDAY AFTERNOON—APRIL 23

2:00 P.M.—Ballroom 9

SESSION N4—SYMPOSIUM: MEMBRANE MODEL SYSTEMS

SESSION III—BIOLOGICAL MEMBRANES

Chairman—**A. A. Benson,** University of California, San Diego, Calif.

2:00 **INTRODUCTORY REMARKS**
A. A. Benson

2:20 125. **MOLECULAR STRUCTURE AND FUNCTION OF CELLULAR MEMBRANE**
F. S. Sjostrand, University of California

2:40 **DISCUSSION**

3:00 126. **SELECTIVE REMOVAL OF MYCOPLASMA MEMBRANE COMPONENTS**
T. M. Terry, Albert Einstein College of Medicine

3:20 **DISCUSSION**

3:40 **INTERMISSION**

3:50 127. **BIOLOGICAL EFFECTS OF CHANGES IN FATTY ACYL GROUPS IN MEMBRANE POLAR LIPIDS**
M. Tourtellotte and R. N. McElhaney, University of Connecticut

4:10 **DISCUSSION**

4:30 128. **THERMAL PHASE TRANSITIONS IN BIOLOGICAL MEMBRANES**
J. M. Steim, Brown University

4:50 **DISCUSSION**

WEDNESDAY AFTERNOON—APRIL 23

2:00 P.M.—Ballroom 6

SESSION V—FLAVOR AND ODOR IN FATS AND OILS

Chairman—**Thomas H. Smouse,** Anderson, Clayton & Co., Richardson, Texas

2:00 129. **VOLATILES FROM HIGH TEMPERATURE OXIDATION OF CIS-7-TETRADECENE**
R. J. Horvat, Western Regional Research Laboratory, USDA

2:20 130. **ROASTED PEANUT FLAVOR**
George R. Waller, Bobby R. Johnson, Philip E. Koehler, George V. Odell and Michael E. Mason, Oklahoma State University

2:40 131. **OXIDATION AND FLAVOR DETERIORATION OF OILS AND MARGARINES DURING SHALLOW PAN FRYING**
Ulla Holm and Lillemor Fredholm, Margarinbolaget AB Technical Department, Sweden

3:00 132. **VOLATILE COMPOUNDS FROM THERMALLY OXIDIZED METHYL OLEATE**
D. A. Withycombe, Mead Johnson and Co., L. M. Libbey and R. C. Lindsay

3:20 **INTERMISSION**

3:30 133. **THE ROLE OF ESSENTIAL OILS OF RANGE AND FORAGE PLANTS IN INFLUENCING ANIMAL ACCEPTANCE**
George V. Odell, Charles J. Rudolph, Michael R. McGeehon, Winifred E. McMurphy and George R. Waller, Oklahoma State University

3:50 134. **THE GAS LIQUID CHROMATOGRAPHY OF NITRO AND CHLORO SUBSTITUTED PHENYLHYDRAZONES OF N-ALKANALS AND N-METHYL KETONES AND SYN-ANTI ISOMERS**
R. C. Tripp, T. Richardson, C. H. Amundson and J. H. von Elbe, University of Wisconsin

4:10 135. **REACTIONS OF FATTY ALDEHYDES WITH FATTY ALCOHOLS: FORMATION OF ACETALS, HEMIACETALS AND ALK-1-ENYL ALKYL ETHERS**
V. Mahadevan, VA Hospital

THURSDAY MORNING—APRIL 24

9:00 A.M.—Teakwood Room

SESSION W—SYMPOSIUM: POLLUTION CONTROL

Chairman—**Francis Scofield,** National Paint, Varnish & Lacquer Association, Washington, D.C.

9:00 **INTRODUCTORY REMARKS**
Francis Scofield

9:10 136. **REGULATIONS CONTROLLING SOLVENT EMISSIONS**
Milton Feldstein, Bay Area Air Pollution Control District

9:30 137. **PETROLEUM SOLVENTS CONFORMING TO AIR POLLUTION CONTROL REGULATIONS**
Bert Sipple, Shell Chemical Co.

9:50 138. **EMISSION CONTROL BY COMBUSTION**
L. C. Hardison, Robert B. Taylor and Otto M. Ikeda, UOP Air Correction Division

10:10 139. **WASTE TREATMENT**
James R. McFarland, Swift & Company

10:30 **PANEL DISCUSSION**

THURSDAY MORNING—APRIL 24

9:00 A.M.—Ballroom 4

SESSION X—NUTRITION II

Chairman—**George Vahouny,** George Washington University, Washington, D.C.

9:00 140. **THE EFFECT OF INHIBITORS OF CHOLESTEROL SYNTHESIS ON MYELIN FORMATION**
Marion E. Smith, Remo Fumagalli and Rodolfo Paoletti, VA Hospital

9:20 141. **IN VITRO STUDIES ON CHOLESTEROL METABOLISM IN THE BLOOD FLUKE SCHISTOSOMA MANSONI**
Thomas M. Smith and Thomas J. Brooks, Jr., University of Mississippi

9:40 142. **THE METABOLIC FATE OF CHOLESTEROL EPOXIDE IN THE RAT**
Joseph A. Fioriti, Marilyn N. George and Rex J. Sims, General Foods Corp.

10:00 143. **LIPASE, ESTERASE AND PHOSPHOLIPASE ACTIVITIES OF LYSOSOMES AND A SENSITIVE MEASUREMENT OF FATTY ACID RELEASE**
Cora J. Dillard, A. L. Tappel, K. Hayase, S. Mahadevan and A. Mellors, University of California

10:20 144. **STUDIES IN FATTY ACID SYNTHESIS BY CELL-FREE PREPARATIONS OF LACTATING MAMMARY GLAND OF THE MONGOLIAN GERBIL**
John G. Coniglio and Raymond B. Bridges, Vanderbilt University School of Medicine

10:40 145. **FATTY ACID METABOLISM OF COHO SALMON UNDER PENTACHLOROPHENOL AND SUBMAINTENANCE DIET**
David F.-Hanes, Hugo M. Krueger and Robert R. Lowry, Oregon State University

11:00 146. **ISOLATION OF 22:5ω6 AND 22:5ω3 ACIDS AND THEIR CONVERSION TO PROSTAGLANDINS VIA SHEEP VESICULAR GLANDS**
L. J. Nutter, O. S. Privett and W. O. Lundberg, The Hormel Institute

THURSDAY MORNING—APRIL 24

9:00 A.M.—Ballroom 7 and 8

SESSION Y—AFLATOXIN II

Chairman—**Walter A. Pons, Jr.,** Southern Regional Research Laboratory, New Orleans, La.

9:00 147. **AFLATOXINS IN FARMERS' STOCK PEANUTS: PEANUT QUALITY, MYCOFLORA AND CLIMATOLOGICAL CONDITIONS AS INFLUENCING FACTORS**
Ben Doupnick, Jr., University of Georgia

9:20 148. **AFLATOXINS IN COTTONSEED: DIFFERENTIAL ELABORATION BY ASPERGILLUS FLAVUS ISOLATES**
L. J. Ashworth, Jr., J. L. McMeans and C. M. Brown, U.S. Cotton Research Station, USDA

9:40 149. **MYCOTOXIN PROBLEMS IN TOBACCO**
A. I. Schepartz, D. G. Bailey and J. H. Cisle, Eastern Utilization Research and Development Division, USDA

10:00 150. **SIMPLE METHOD FOR DETERMINING AFLATOXIN-PRODUCING POTENTIAL OF FUNGI**
Jerry Kirksey, C. E. Holaday and Phillip Vincent, Market Quality Research Division, USDA

10:20 151. **THE EFFECTS OF AFLATOXINS ON GERMINATING SEEDS OF FIELD CRESS**
F. R. Roegner, Food and Drug Administration, Washington, D.C.

10:40 152. **A NEW METABOLITE FROM ASPERGILLUS PARASITICUS**
R. D. Stubblefield, Odette L. Shotwell and Gail M. Shannon, Northern Utilization Research and Development Division, USDA

11:00 153. **METABOLIC CONVERSION OF AFLATOXIN B_1 TO M_1 IN VITRO**
M. S. Masri, J. R. Page and V. C. Garcia, Western Utilization Research and Development Division, USDA

11:20 154. **REVIEW OF THE BIOLOGICAL EFFECTS OF AFLATOXINS ON SWINE, CATTLE AND POULTRY**
A. N. Booth, Western Utilization Research and Development Division, USDA

11:40 155. **MYCOTOXICITY OF ASPERGILLUS OCHRACEOUS TO CHICKS**
Ben Doupnick, Jr. and John C. Peckham, University of Georgia

THURSDAY MORNING—APRIL 24

9:00 A.M.—Ballroom 9

SESSION N5—SYMPOSIUM: MEMBRANE MODEL SYSTEMS SESSION IV—BIOLOGICAL PERSPECTIVES

Chairman—**W. Stoeckenius,** University of California, San Francisco, Calif.

9:00 **INTRODUCTORY REMARKS**
W. Stoeckenius

9:20 156. **MEMBRANE STRUCTURE EXPOSED IN HYDROPHOBIC FRACTURES**
D. Branton, University of California

9:40 **DISCUSSION**

10:00 157. **STEROID SPIN-LABEL OF MEMBRANE**
W. L. Hubbell and H. M. McConnell, Stanford University

10:20 **DISCUSSION**

10:40 **INTERMISSION**

10:50 158. **GENETIC CONTROL INVOLVED IN THE BIOGENESIS AND FUNCTION OF MITOCHONDRIA**
D. O. Woodward, Stanford University

11:10 **DISCUSSION**

11:30 159. **STRUCTURAL PROTEIN AND MEMBRANE ORGANIZATION**
S. Fleischer and W. L. Zahler, Vanderbilt University

11:50 **DISCUSSION**

THURSDAY MORNING—APRIL 24

9:00 A.M.—Ballroom 2 and 3

SESSION Z—ANALYTICAL II

Chairman—**Cameron K. Lyon**, Western Regional Research Lab., Albany, Calif.

9:00 160. EPOXIDIZED OILS AS EMULSIFIABLE PESTICIDE STABILIZERS
Keith L. Johnson, Swift & Co.

9:20 161. A RAPID GAS CHROMATOGRAPHIC METHOD FOR THE DETERMINATION OF BHA AND BHT IN VEGETABLE OILS
Kenneth T. Hartman and Lucien C. Rose, Frito-Lay, Inc.

9:40 162. COMPOSITIONAL ANALYSIS OF MANNIDE MONOOLEATE EMULSIFYING AGENT (ARLACEL A)
H. J. O'Neill and T. N. Yamauchi, IIT Research Institute

10:00 163. DEVELOPMENT OF IMPROVED BAKER'S SHORTENING THROUGH STATISTICAL UNDERSTANDING OF EMULSIFIER EFFECTIVENESS
D. T. Rusch and H. M. Truax, Atlas Chemical Ind., Inc.

10:20 164. DETERMINATION OF POLYSORBATE 60 IN FOODS
Charles F. Smullin, Frank P. Wetterau and Virginia L. Olsanski, Atlas Chemical Ind., Inc.

10:40 165. CONJUGATION OF POLYUNSATURATED FATTY ACIDS: METHYL LINOLENATE
T. L. Mounts and H. J. Dutton, Northern Regional Research Laboratory, USDA, and D. Glover

11:00 166. CONJUGATION OF POLYUNSATURATED FATTY ACIDS: METHYL LINOLEATE
T. L. Mounts and H. J. Dutton, Northern Regional Research Laboratory, USDA, and D. Glover

THURSDAY AFTERNOON—APRIL 24

2:00 P.M.—Ballroom 1

SESSION N6—SYMPOSIUM: MEMBRANE MODEL SYSTEMS

INTERNATIONAL MEMBRANE WORKSHOP: LIPID-PROTEIN ASSOCIATION IN MEMBRANES—HYDROPHOBIC OR ELECTROSTATIC?

WORKSHOP SESSION II: MEMBRANE PROTEINS

Chairman—**D. O. Woodward**, Stanford University, Stanford, Calif.

2:00 INTRODUCTORY REMARKS
D. O. Woodward

2:20 DISCUSSION

FRIDAY MORNING—APRIL 25

SYMPOSIUM—MEMBRANE MODEL SYSTEMS INTERNATIONAL . MEMBRANE . WORKSHOP: LIPID-PROTEIN ASSOCIATION IN MEMBRANES—HYDROPHOBIC OR ELECTROSTATIC?

WORKSHOP SESSION III: MEMBRANE MODELS—GENERAL DISCUSSION

Chairman—**D. Branton**, University of California

9:00 INTRODUCTORY REMARKS
D. Branton
DISCUSSION

12:00 CONCLUDING REMARKS
F. S. Sjostrand, University of California

ABSTRACTS OF PAPERS

1

NEW BUILDING HARD SURFACE CLEANING PROBLEMS. ALAN W. LEIPNITZ, Economics Laboratory, Inc., St. Paul, Minnesota.

An office building designed with cleanability in mind still may present unique hard surface cleaning problems. The Osborn Building, new home office of Economics Laboratory, Inc., presented two initial cleaning problems. Construction soil on the glass and stainless steel exterior required a new hard surface cleaning product and procedure for efficient removal. The combination of product and procedure was shown to be the key to success. One terrazo floor required only partial cleaning to obtain the desired appearance. This floor could be over-cleaned.

2

DETERGENTS—HARD WATER INTERACTION IN MACHINE DISHWASHING. R. F. VANCE, General Electric Company, AP5-249, Appliance Park, Louisville, Kentucky.

Insufficient detergent concentrations have recently been recognized as a major contributor to film formation on glassware in machine dishwashing. The film is shown to consist largely of calcium tripolyphosphate, $Ca_5(PsO_{10})_2$; evidence is cited for the presence of a silicate, probably calcium metasilicate, as well. Systematic variation of detergent concentration and of water hardness allows construction of a response surface describing conditions of maximum and minimum rates of film formation. The implications of this treatment to other detergency processes is briefly considered.

3

ANALYSIS OF DATA FROM DETERGENCY TESTS WITH A NATURAL SOIL. J. R. TROWBRIDGE, Colgate-Palmolive Company, Piscataway, New Jersey.

A method of applying soil to a small circular areas of a test fabric by rubbing it over the surface of the skin has been described previously. These test swatches have advantages over artificially soiled swatches because they have a real soil and can be resoiled after washing to provide a practical laundry simulation in which fabrics receive multiple soil-wash cycles. Recent experience shows that a considerable gain in precision can be obtained by using block designs in which swatches within a block are evenly soiled by the same individual. The standard deviation for replicate swatches is approximately proportional to the amount of residual soil as determined from reflectance measurements. A log transformation of the reflectance data provides a more homogeneous error variance and increased significance for differences amongst wash treatments.

4

A TRIPLY LABELED PARTICULATE SOIL FOR DETERGENCY STUDIES. B. E. GORDON and W. F. SHEBS, Shell Development Company, Emeryville, California.

The radioactive kaolinite described in an earlier report has undergone extensive testing to develop suitable padding and analytical methods for its application as a particulate component of a synthetic sebum. A large scale method for padding amounts of 500 swatches of a test fabric based on the technique of Rutkowski was developed. During the padding study it was found that the kaolinite (Spinks Bandy Black) remaining on the fabric was of different species activity than the starting clay. The cause was traced to the presence of a coarse high silica, low specific activity impurity and a very fine high specific activity component. Removal of these by wet screening yielded a more homogeneous clay. Gamma ray analysis of the swatches before and after washing leads to the per cent of clay removed. Beta ray analysis of the wash water leads to the per cent of the fatty soil removed. There is no interference by the fatty soil with the clay analyses. Interference by the radioactive clay in the fatty sebum analysis is avoided by control over the radioactivity of the various components and by careful spectrometric analysis.

5

STAINS—FABRICS—DETERGENTS. B. W. TERRY and W. L. GROVES, Continental Oil Company, Ponca City, Oklahoma.

Several common stains were applied to cotton, permanent press and soil release fabrics. The spot-stained fabrics were washed in eight detergent formulations, and in some cases at two temperatures. The formulation variables include LAS or nonionic actives, 45% or 35% tripolyphosphate, and with or without a commercially available enzyme. Results due to variations in response of stains to certain fabric finishes preclude broad generalizations. Cloth variations are probably the most significant parameters observed. Permanent press and soil release usually allow more complete removal of pea, ketchup, canned spinach and gravy stains. Permanent press fabrics will usually allow more complete removal of blood stains than soil release fabrics, while the reverse is true for fruit stains. High temperature (150 F) practically eliminates the enzyme effect; it sets some stains while rarely improving removal. Nonionic-enzyme combinations are usually as good as the comparable LAS formulation. In several cases the higher phosphate level shows an advantage in enzyme formulations. Some stains are attacked reasonably well by the enzyme formulations studied, whereas grass, fruit, chocolate syrup and blood stains will obviously need more extensive treatment.

6

CRITICAL MICELLE CONCENTRATION (CMC) OF DIHYDROXY AND TRIHYDROXY BILE SALTS—EFFECTS OF COUNTERION AND TEMPERATURE. MARTIN C. CAREY and DONALD M. SMALL, Boston University School of Medicine, Boston, Massachusetts.

Sodium taurocholate (NaTC) and sodium taurodeoxycholate (NaTDC) found in human bile and intestinal contents are detergents which on structural grounds should have both anionic and nonionic properties. Experiments were designed to study whether the extra hydroxyl group imparts more nonionic characteristics to NaTC micelles. The CMC of pure NaTC and NaTDC was determined from 10–20 C in water and NaCl solutions utilizing the shift in the absorption maximum of 2.5×10^{-6} M Rhodamine 6G. A sigmoid curve resulted when each spectral shift was plotted against the corresponding concentration of bile salt; the point of inflection was taken as an estimate of the CMC. The CMC minimum at 20–30 C of NaTC suggests the importance of hydrophobic interaction. At higher temperatures the CMC of both bile salts increases indicating that charge effects predominate. NaTC shows a smaller per cent increase with temperature—an effect of its three hydroxyl groups. An approximately linear relationship exists between log CMC and log NaCl. NaCl lowered the CMC of NaTDC but NaTC was resistant to salt effects indicating the small size of its micelles with charges well separated. The $-\Delta F$ of micellization was greater for NaTDC than NaTC. The enthalpy of micellization was calculated from the temperature dependence of the CMC and entropy from $\Delta F = \Delta H - T\Delta S$. With the exception of high temperatures, bile salt micellization was found to be an entropy directed process.

7

RAT LIVER LIPIDS: METABOLIC RELATIONSHIPS DERIVED FROM STRUCTURAL ANALYSES OF NEUTRAL GLYCERIDES AND PHOSPHOGLYCERIDES. RANDALL WOOD and R. D. HARLOW, Medical Division, Oak Ridge Associated Universities, Oak Ridge, Tennessee.

The distribution of fatty acids esterified at each position of glycerol in triglycerides (TG), phosphatidyl choline (PG), and phosphatidyl ethanolamine (PE) of rat liver was determined. The carbon number distribution of TG and diglyceride acetates derived from PC and PE was also determined. Each of the TG positions showed a distinct distribution of fatty acids that was not arranged randomly, indicating pairing of some acids. The composition of the fatty acids esterified at the 1 position of PC and PE was the same, but differed from the 1 position of TG; the 2 position differed in all three classes. The carbon number distribution of PC diglyceride acetates was different from that of PE diglyceride acetates and neither agreed with the random distribution values calculated from the experimentally determined compositions of the 1 and 2 positions of each class, indicating preferential pairing of some acids in these lipid classes also. Selectivity of diglycerides used for the biosynthesis of either TG or phospholipids or both is substantiated by the lack of agreement between the TG carbon number distribution determined experimentally and the distribution calculated from values of either PC of PE diglyceride acetates plus the values of the 3 position of the TG. These findings for rat liver are the opposite of those reported in a companion paper in which tumor cells did not show selectivity of diglycerides used for TG and PC biosynthesis.

8

TUMOR LIPIDS: METABOLIC RELATIONSHIPS DERIVED FROM STRUCTURAL ANALYSES OF ACYL-, ALKYL AND ALK-1-ENYL MOIETIES OF NEUTRAL GLYCERIDES AND PHOSPHOGLYCERIDES. RANDALL WOOD and FRED SNYDER, Medical Division, Oak Ridge Associated Universities, Oak Ridge, Tennessee.

The composition of the hydrocarbon moieties of the 1, 2 and 3 positions of triglycerides and glyceryl ether diesters (GEDE) and of the 1 and 2 positions of diacyl and alkyl acyl phosphatidyl cholines (PC) and diacyl, alkyl acyl and alk-1-enyl acyl phosphatidyl ethanolamines (PE) from Ehrlich ascites cells (EAC) were determined. The carbon number percentage distribution of triglycerides, GEDE, and the diglyceride-type acetates derived from each class of PC and PE was also determined by gas-liquid chromatography analysis of the intact lipids. The fatty acid compositions of the 1, 2 and 3 positions of the triglycerides are different and also differ from the composition of the corresponding positions of the GEDE, which are not the same. Both triglycerides and GEDE exhibit a 1-random-2-random-3-random type of distribution. The choline-containing phosphatides consist of approximately 66% diacyl PC and 33% alkyl acyl PC. Ethanolamine-containing phosphatides are composed of 55% diacyl PE, 30% alkyl acyl PE, and 15% alk-1-enyl acyl PE. The carbon chain of the 1 position of each PC and PE class is predominantly saturated and the 2 position is dominated by polyunsaturated fatty acids. All PC and PE classes except the alk-1-enyl acyl PE show a 1-random-2-random distribution. In EAC, randomly synthesized diglycerides are apparently used randomly for triglycerides and diacyl PC biosynthesis; the biosynthesis of GEDE and alkyl acyl PC from an alkyl acyl intermediate also occurs without selectivity. In contrast, we have shown in a companion report that selectivity of diglycerides for the biosynthesis of triglycerides PC and PE occurs in rat liver. The similarities in composition at both the 1 and 2 positions between triglycerides and diacyl PC and between GEDE and alkyl acyl PC suggest a loss of acyl CoA:lysophosphatide acyl transferase enzymes which are present in normal tissue. Metabolic relationships derived from structural analyses of acyl, alkyl and alk-1-enyl moieties of glycerides and phosphoglycerides and from established pathways for acylation reaction led to the proposal of a metabolic pathway for the biosynthesis of lipids containing glyceryl ethers.

9

METABOLISM OF LONG CHAIN FATTY ALCOHOLS: FORMATION OF WAXES AND GLYCERYL ETHERS. FRED

SNYDER, BOYD MALONE and JAMES SOODSMA, Medical Division, Oak Ridge Associated Universities, Oak Ridge, Tennessee.

Fatty alcohols, produced during cleavage of glyceryl ethers, are formed by the reduction of fatty aldehydes in the presence of NADPH. Recently, Friedberg and Greene described an enzyme system in mammalian and dogfish livers that esterifies the fatty alcohols with waxes. We have examined the oxidation and esterification of 1-^{14}C-fatty alcohols and their role as precursors of alkyl glyceryl ethers in liver, kidney, spleen, lung and bone marrow of rats and in neoplastic tissue. The metabolism of fatty alcohols was investigated in homogenates and subcellular fractions. The soluble protein fraction (34,000 *g* supernate) of cells contained the highest enzymatic activity for wax formation. The liver fraction formed more wax than analogous fractions from kidneys, spleens, lungs or bone marrow; similar results were obtained for both the $C_{16:0}$ and $C_{18:0}$ alcohols. Oxidation of the fatty alcohols to fatty acids was highest in livers and kidneys and $C_{16:0}$ alcohol was the most active substrate. In vivo experiments indicated that oxidation of the alcohol to the fatty acid and subsequent acylation with glycerol is the major metabolic route, but direct esterification with fatty acids to form waxes also occurs. In neoplastic cells, fatty alcohols and glyceraldehyde-3-PO_4 were readily converted to alkyl glyceryl ethers by microsomes in the presence of ATP, CoA and Mg^{++}.

10

SEED LIPIDS AND CHEMOTAXONOMY.

ARTHUR S. BARCLAY, QUENTIN JONES and IVAN A. WOLFF, New Crops Research Branch, Plant Industry Station, Beltsville, Maryland.

During the past 10 years, nearly 8000 seed samples from 170 plant families have been analyzed for gross chemical composition and more than 2,700 seed oils for fatty acid composition. This research has generated a wealth of chemical information on seed lipids, much of which appears to have taxonomic significance. In seed plants a positive correlation is evident between oil and protein content: families rich in seed oils tend also to be rich in protein; families with starchy seeds tend to be low in oil and protein. Compositional patterns observed for oil and protein percentage seem characteristic for certain families. Within some families, oil and protein content are positively correlated, whereas in others these constituents vary independently of one another. Examples of chemotaxonomic patterns below the family level are provided by comparative data on fatty acid composition of seed oils within the families Boraginaceae, Compositae and Cruciferae. The compositional data confirm taxonomic concepts synthesized from more traditional sources of evidence. Chemical characterization of major taxonomic categories within a family is illustrated by fatty acid profiles of the four widely recognized subfamilies of the Boraginaceae: Cordioideae, Ehretioideae, Heliotropioideae and Boraginoideae. The occurrence of high percentages of dimorphecolic acid in only the genus *Dimorphotheca*, a single section (*Blaxium*) of the genus *Osteospermum* and the genus *Castalis*, supports current concepts of relationship and phylogeny within the tribe Calenduleae (Compositae) based on comparative morphology. The relationships among three genera of the Cruciferae, *Cardamine*, *Leavenworthia* and *Selenia*, have long been uncertain. The fatty acid composition of representatives of these genera suggests an alliance between *Leavenworthia* and *Selenia* but not between these genera and *Cardamine*. On the basis of comparative morphology and cytology, *Physaria* is the genus most closely related to *Lesquerella* and now the discovery of lesquerolic acid in only these genera of Cruciferae corroborates this relationship. Also in seeds of *Lesquerella*, the distribution of C_{18} and C_{20} monohydroxy acids provides an example of the possible utility of chemical information in helping to elucidate relationships within genera. In the genus *Crepis*, the species so far analyzed agree markedly with the sectional classification of the genus with respect to the amounts of crepenynic and vernolic acid in their seed oils.

11

TAXONOMIC PATTERNS IN THE LIPIDS OF PHOTOSYNTHETIC TISSUE.

B. W. NICHOLS and R. J. B. WOOD, Unilever Research Laboratories, Colworth House, Sharnbrook, Bedford, England.

The relationship between the lipid and fatty acid composition of plant tissues and their taxonomic status has been intensively studied from the standpoint of seed and leaf cuticle analyses. The obvious limitations of such lines of approach are that they can only be applied to the higher plants in which these clearly defined organs occur. The question whether some form of lipid analysis can assist in the classification of lower forms of plant life has hitherto received only sporadic attention, but certain consistent features have now been established. Thin-layer chromatographic analyses of lipid extracts from photosynthetic tissues can readily determine whether the original tissue was that of a higher plant, a higher alga, a blue-green alga or a bacterium. In certain circumstances more specific information regarding the classification of the original cell can be obtained. In particular, a glycolipid unique to nitrogen-fixing blue-green algae has been characterized and this compound occurs only in cultures which have fixed molecular nitrogen during growth. In addition, a glyceride which contains nitrogen, but no phosphorus, has been isolated and appears to be characteristic of the *Ochromonas* family of phytoflagellates. Moreover, fatty acid compositions, particularly when related to the organelles and lipids in which they occur, can also assist in the taxonomic classification of the lower forms of plant life.

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THE DISTRIBUTION OF STEROLS, ALKALOIDS AND FATTY ACIDS IN SENITA CACTUS OVER ITS RANGE IN SONORA, MEXICO.

HENRY W. KIRCHER, Department of Agricultural Biochemistry, University of Arizona, Tucson, Arizona.

Senita cactus [*Lophocereus schottii* (Engelmann) Britton and Rose] is one of the large columnar cacti that grow in southwestern United States and northwestern Mexico. It ranges from the US-Mexico border south through the states of Sonora and Baja California partway into the state of Sinaloa. The morphology of the plant changes as one travels from the arid north to the wetter south. A Drosophila species (*D. pachea*) that uses senita exclusively as a habitat has a chromosomal inversion whose frequency in the population also shows a cline from north to south. The cactus contains a number of unusual sterols [including 4-α-methyl-Δ^7-cholesten-3β-ol (lophenol) and Δ^7-stigmasten-3β-ol (schottenol)], a triterpene fraction that includes lupeol, some isoquinoline alkaloids [including 1-isobutyl-2-methyl-6-methoxy-7-hydroxy-1,2,3,4-tetrahydroisoquinoline (lophocereine) and its trimer, pilocereine] and a fatty acid fraction composed mostly of palmitic, oleic, linoleic and linolenic acids. Plants were sampled over most of Sonora and analyzed for the title compounds by chromatographic methods to see if chemical changes accompanied the morphological changes and to determine whether the chromosomal inversion in *D. pachea* could be correlated with differences in the lipid fraction of the cactus. The results of this investigation showed greater differences between juvenile and mature branches of the same plant and between the epidermis and cortex of an individual branch than between plants collected over a 350 mile range and belonging to two distinct varieties. The younger branches contained a higher proportion of three unknown sterols, triterpenes, phenolic alkaloids and linolenic acid than the older branches. The latter contained much more of a fatty acid fraction having less than 16 carbon atoms. When the epidermis was compared to the cortex of a single branch, it contained more of the unknown sterols, triterpenes and linolenic acid. This may be a function of the photosynthetic apparatus, which lies entirely in the epidermis of the cactus. Except for a higher proportion of fatty acids of carbon number less than 16 in older arms of plants in southern Sonora, no chemical differences were noted that correlated with the morphological differences and the inversion frequencies noted in the giant chromosomes of *D. pachea*. These differences are more likely due to different climates than to the chemical characteristics of senita cactus.

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LIPID CONTENT AND FATTY ACID ANALYSIS OF THREE PURE VARIETIES OF OATS (*AVENA SATIVA*) GROWN UNDER VARIOUS AGRICULTURAL PRACTICES.

GEORGE C. POTTER and JOY CARY, The Quaker Oats Co., John Stuart Research Laboratory, Barrington, Illinois.

Three varieties of oats were grown using three seeding rates and three fertilization levels. The varieties included a low, moderate and high fat content type oat. The oats were dehulled and analyzed for crude fat content and fatty acid content of the neutral lipids of the groats. The effect of these cropping practices on crude fat and fatty acid content was negligible. These parameters are genetically controlled. By comparing the three varieties with the same oat varieties grown in a more southern latitude, some indication of a shift in the content of linoleic acid was indicated. However climate has very little effect on the total lipid content of the oat.

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CONTINUOUS EXTRUSION COOKING OF COTTONSEED KERNELS AND OF PARTIALLY DEFATTED MEAL.

S. P. CLARK, Texas A&M University, Cottonseed Products Research Laboratory, College Station, Texas.

Continuous extrusion cooking produces a short time pressure cooking of the material being processed. The Wenger extruder-cooker has been successfully applied by others to many materials including soybeans. This is a report of an investigation of the extruder-cooker applied to glanded cottonseed meats and to partially defatted cottonseed meal. The principal objective was lowering of free gossypol in these materials. The extruder was not effective in reducing residual gossypol to low levels, however the preconditioner ahead of the extruder was effective. Free oil, foam and fluidity in the extruder discharge was an operating problem which was controlled by the addition of water to the material ahead of the extruder. Extrusion cooked meats were screw pressed successfully, however the oily character of such meats would be a potential problem for a screw pressing operation.

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REACTION OF GOSSYPOL WITH AMINO ACIDS, PEPTIDES AND OTHER AMINO-COMPOUNDS—PREPARATION, PURIFICATION AND CHARACTERIZATION.

CARL M. CATER and CARL M. LYMAN, Cottonseed Products Research Laboratory, Texas A&M University, College Station, Texas.

Gossypol, the yellow pigment found in cottonseed, has long been known to form complexes with protein, the binding site thought to consist principally of the epsilon-amino group of lysine. However, the reactions of gossypol with free amino acids, small peptides and other amino-containing compounds are less well known. Studies have been conducted in aqueous solutions containing enough alcohol to keep the gossypol in solution, which demonstrate spectroscopic changes as the reaction proceeds, along with an increase in rate of reaction with increase in pH in the range from 5.7 to 7.5. The rate of reaction of gossypol with amino acids has been shown to be related to the distance of the amino group from the carboxyl group within the molecule. Reaction products of gossypol with amino acids, small peptides and amino-containing compounds were subjected to various purification procedures and analysis to determine combination ratios. In addition to the expected gossypol-to-amino-compound ratio of 1:2, dictated by the formation of Schiff-base type bonds with the two aldehyde groups of gossypol, compounds with ratios of 1:3 and 1:4 were isolated. These results indicate that each of the two aldehyde groups of gossypol can react with two amino-compounds.

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CHEMICAL INACTIVATION OF CYCLOPROPENOID FATTY ACIDS IN COTTONSEED MEALS.

H. J. O'NEILL, H. G. REILICH, J. PROCTOR and W. A. PONS, JR., IIT Research Institute, Chicago, Illinois.

Commercial cottonseed meals were subjected to chemical treatment with organic acids, anhydrous gases and sulfhydryl compounds to reduce the cyclopropenoid fatty acid (CPA) content. Selected meals were then incorporated at 20 wt % levels in the rations of laying hens and compared against a negative control containing 25 wt % soybean meal and a positive control consisting of 2% refined cottonseed oil of known CPA content (25 ppm CPA). During and after a four-week feeding period, eggs were collected and stored for three and six months at 35 F. Ration consumption, yolk and albumen discoloration, pH of yolks and albumen, and yolk fatty acid patterns were determined. Of the reagents used, sulfur dioxide eliminated practically all the residual CPA in the meal and yielded improved egg quality. Thioglycollic acid treated meal exhibited reduced albumen discoloration and normal yolk and albumen pH, but did exhibit high yolk discoloration after three months (32%) and six months (100%) storage. Oleic acid treated meals were less effective

than either sulfur dioxide or thioglycollic acid treatment under the conditions employed. The thioglycollic acid treated meal also exhibited a marked reduction in hen consumption as compared to the negative control (36%) and sulfur dioxide treated rations (50%).

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STRUCTURAL ANALYSIS OF FATTY ACIDS VIA OZONOLYSIS. O. S. PRIVETT, The Hormel Institute, Austin, Minnesota.

A 35 min, 16 mm color movie demonstrating the techniques of ozonolysis for the localization of double bonds in fatty acids will be presented. Theoretical aspects and basic principles of the reactions upon which the common methods are based are illustrated as well as consideration of factors that influence the reaction and cause spurious results. A method for the determination of specific positions of *cis* and *trans* double bonds in polyunsaturated fatty acids is demonstrated and applied to the analysis of liver fatty acids containing *trans* acids.

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MICRO-OZONIZER: AN APPARATUS FOR OZONOLYSIS. NICHOLAS PELICK, Supelco, Inc., Bellefonte, Pennsylvania.

A micro-ozonizer device is described for locating double bonds in organic compounds. Ozonization is conducted on as little as a microgram of methyl esters of fatty acids. The reaction is completed in a few minutes and gas chromatography is used for the analysis of the ozonolysis fragments.

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AUTOMATED METHOD FOR THE DETERMINATION OF TOTAL ENZYME IN PRESOAK DETERGENTS. L. M. PAIXAO, E. J. MAITHENY and O. BENZ, Colgate-Palmolive Research Center, Piscataway, New Jersey.

A modified Lowry color reaction has been adapted for the automated determination of total enzyme in pre-soak detergents. The detergent sample, in a buffered medium, is passed through an ion-exchange column and the effluent reacted with Folin-Ciocalteau phenol reagent. Reaction with the tyrosine groups in the enzyme results in the formation of a blue complex which is proportional to enzyme concentration. A novel approach to column chromatography is presented. It utilizes a Technicon Sampler II to automatically feed samples at the rate of 10 per hour into the ion-exchange column. The method is accurate and has been used in quality control work as well as in research. Interferences of some formulation ingredients and limitations of the method are discussed.

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FLUORESCENT WHITENING AGENTS FOR ENZYME-CONTAINING LAUNDRY PRODUCTS. PER S. STENSBY, WILLIAM R. FINDLEY and CHARLES W. LIEBERT, Geigy Industrial Chemicals, Ardsley, New York.

Fluorescent whitening agents (FWA), previously called optical brighteners, fluorescent bleaches, etc., have become standard ingredients in most laundry products. Since 1963 in Europe and since 1967 in the USA, the use of enzymes in pre-soaking agents and heavy duty detergents has increased tremendously. It appears that in the United States the usage of enzymes will continue to increase over the coming years. Eventually enzymes, like FWA, might become standard components of home laundry pre-soaks and detergents. The compatibility of currently available alkaline proteases (and amylases) on one hand, and surfactants, builders, fillers, etc., on the other, has been discussed in recent papers. Little information is available on the interaction of FWA and enzymes. This paper will discuss the whitener systems currently being used in pre-soaking agents and heavy duty detergents. Data on performance of commonly used FWA in formulations with and without enzymes will also be presented and interaction of FWA and enzymes discussed.

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AN AUTOMATED DETERMINATION OF LINEAR ALKYL SULFONATES IN SPRAY DRIED DETERGENTS. E. H. BRANDLI and R. M. KELLEY, Colgate-Palmolive Company, Piscataway, New Jersey.

An automated procedure has been developed for the determination of linear alkyl sulfonates in spray dried detergents. It is based upon the ultra violet absorbance of linear alkyl sulfonates at 224 mμ. A Technicon solid preparative sampler is employed to dissolve the sample, dilute to a fixed volume and remove an aliquot for analysis. The sample aliquots are continously fed through a Technicon proportioning pump to a flow cell in a Beckman DU spectrophotometer. The spectrophotometer is equipped with a spectral energy recording attachment and a strip chart recorder. All operations, excluding sample weighing, are automatic and analyses can be carried out continuously at the rate of 10 samples per hour. The method has been found to have a coefficient of variation of 3.5%.

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THE COURSE OF BIODEGRADATION OF ANIONIC DETERGENTS. T. C. CORDON, E. W. MAURER and A. J. STIRTON, Eastern Utilization Research and Development Division, ARS, USDA, Philadelphia, Pennsylvania.

Synthetic detergents derived from fats were subjected to the degradative action of sewage microorganisms in an aerobic system in which the detergent was the sole source of carbon and energy. Linear alkylbenzenesulfonate (LAS) was a reference standard. The breakdown of the detergents was followed by measuring the loss of carbon and methylene blue-active substance (MBAS) and the formation of sulfate ion. Alcohol sulfates were broken down within one or two days and the sulfur was recovered as sulfate ion almost quantitatively. Sulfate ion appeared concomitantly with loss of carbon. Ether alcohol sulfates were less readily attacked than alcohol sulfates. Three to five days were required for the MBAS to be reduced to zero compared to one to two days for the alcohol sulfates. Increasing the oxyalkyl groups from oxyethyl to oxypropyl or oxybutyl resulted in a longer time lapse before significant amounts of sulfate ion appeared although considerable carbon reduction had already occurred. Reduction in MBAS was quite rapid with the α-sulfo fatty esters. Loss of carbon and formation of sulfate ion was less rapid, possibly because of the intermediate formation of sodium sulfosuccinic acid or a related compound.

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N→O ACYL MIGRATION STUDIES OF N-(2-HYDROXYETHYL) LAURAMIDES. T. M. MUZYCZKO and J. A. LOBODA, The Richardson Company, Melrose Park, Illinois.

N-hydroxyethyl fatty acid amides are known surfactants. These compounds can rearrange to their ester-amine isomers. In our study, N-methyl-N-(2-hydroxyethyl) and N,N-bis-(2-hydroxyethyl) lauramides were prepared, purified and characterized. Acyl migrations were followed by nuclear magnetic resonance and infrared spectroscopy techniques as a function of temperature and ph. Kinetic data and activation energy calculations are presented. Probable mechanisms are discussed in light of these data.

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LIPIDS FROM THE MICROSOMAL, MITOCHONDRIAL AND MYELIN FRACTION OF MOUSE BRAIN. GRACE Y. SUN and LLOYD A. HORROCKS, Cleveland Psychiatric Institute, Cleveland, Ohio.

Microsomal, mitochondrial and myelin fractions were prepared from the whole brains of female C57BL/10 mice. The protein contents were 44% for microsomes, 72% for mitochondria and 32% for myelin (per cent of dry weight). Large differences were also observed for the lipid compositions. Myelin fractions were rich in cholesterol, galactolipids and alk-1-enyl acyl glycerophosphorylethanolamine (GPE). Mitochondrial fractions had much lower proportions of cholesterol, galactolipids and alk-1-enyl acyl GPE, a higher proportion of choline phosphoglycerides and contained an appreciable amount of cardiolipin. The composition of the microsomal fractions was intermediate between the mitochondria and myelin. The side-chain compositions of the major phosphoglycerides conformed generally to the usual patterns. However, pronounced differences were found when lipids from the myelin were compared with the same lipid class from the mitochondrial or microsomal fractions. All of the major phosphoglycerides from myelin were deficient in saturated and 22:6 side-chains and enriched in 18:1 and 20:1. The phosphoglycerides from the microsomes had side-chain compositions that were similar to those of the mitochondria. The mitochondrial cardiolipin contained unusually high amounts of several acyl groups including 18:1, 52%; 18:2, 6%; and 16:1, 4%. The major reservoir of 18:2 in the mouse brain seems to be in the mitochondrial cardiolipin.

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LIPID CONTENT AND FATTY ACID PATTERNS IN DEVELOPING STEELHEAD SAC FRY. LYLE HAYES, ROBERT R. LOWRY and GARY A. CHAPMAN, Department of Agricultural Chemistry, Oregon State University, Corvallis, Oregon.

The changes in total lipid and fatty acid composition of developing steelhead sac fry were examined in the period from fertilization of the egg to disappearance of the yolk sac (69 days). Fertilized eggs maintained in flowing water were sampled at intervals until hatching. After hatching, whole fish and sac fry whose yolk had been detached were extracted. Lipids were extracted by homogenization in chloroform-methanol 2:1 and aliquots were weighed. Fatty acid methyl esters were prepared by reacting the lipid extract with methanol and HCl. Identification of the fatty acid methyl esters was accomplished by combining the techniques of thin layer chromatography, hydrogenation, reductive ozonolysis and gas-liquid chromatography. Lipid content of the whole system (sac + fry) was found to remain relatively constant until the time of hatching when a rapid decrease was observed coincident with an increase in weight and lipid content of the fry. After 64 days the whole system retained 52% of its initial lipid, while the fry retained 27%. The fry were found to contain a larger percentage of fatty acids 16:0, 18:0 and 22:6, but less 14:0, 18:1, 22:1 and 22:5 than was found in the whole system. The presence of an effective partitioning of the fatty acids between yolk and developing fry is indicated.

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HYDROCARBON BIOSYNTHESIS BY COCKROACH INTEGUMENT. CHARLES W. CONRAD and LARRY L. JACKSON, Montana State University, Bozeman, Montana.

The three principal hydrocarbons of the American cockroach, *Periplaneta americana* (L.), were previously identified as *n*-pentacosane, 3-methylpentacosane and *cis,cis*-6,9-heptacosodiene. The principal site of hydrocarbon biosynthesis appears to be the cockroach integument. Both intact integument and integument homogenates are capable of incorporating activity from labeled precursors into the hydrocarbons. The incorporation of acetate, malonate, formate, palmitate, linoleate, isoleucine and other hydrocarbon precursors will be discussed. The implications of these experiments in relationship to suggested hydrocarbon biosynthetic pathways will be discussed.

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STUDIES ON THE METABOLIC FATE OF GOSSYPOL IN THE RAT USING ^{14}C LABELED GOSSYPOL. MOHAMED BAHI ABOU-DONIA and CARL M. LYMAN, Department of Soil and Crop Sciences, Texas A&M University, College Station, Texas.

Two groups of rats were used. One was fed a basal diet and the other the same diet plus 500 ppm of iron as ferrous sulfate. Five and 10 mg single doses of gossypol were administered; the animals were maintained in metabolic cages and killed after varous periods of time. ^{14}C activity was determined in the expired air, urine, feces and the various tissues. The data indicate that gossypol is poorly absorbed from the gastrointestinal tract and is rapidly eliminated from the animal body. Administration of larger doses of gossypol inhibited its elimination from the body, thus increasing tissue deposits. The data show that iron in the diet increased the elimination of gossypol via feces and expired air, and decreased tissue deposits and excretion in the urine. After 72 hr only 4.32% and 1.64% of the administered 5 mg dose was recovered from the tissues of rats fed basal and iron supplemented diets respectively, compared to 17.02% ond 11.35% when a 10 mg dose was given. It is proposed that iron protects the animals from gossypol toxicity by enhancing its elimination from the animal. Iron salts function in several ways: first, by forming insoluble iron gossypol compounds which are not easily absorbed from the intestinal tract; second, iron catalyzes the decarbonylation of gossypol to form CO_2 and presumably the unstable appogossypol. This reaction appears to take place in the intestinal tract but may also take place in the tissues.

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CHANGES IN FATTY ACID AND LIPID CLASS COMPOSITION OF SOYBEANS DURING MATURATION. O. S.

PRIVETT, R. A. GROSS and K. BEUTEL, The Hormel Institute, Austin, Minnesota.

Soybeans were harvested at nine intervals during maturation from 12 to 97 days after flowering for analysis of the lipid. The beans were extracted with chloroform-methanol and the fatty acid composition determined by GLC of methyl esters. Lipid class composition was determined by thin-layer chromatography (TLC) via the densitometry-charring technique. Specific color reactions and infrared spectral analysis in addition to TLC properties were used for identification of the lipid classes. There was a decrease in the percentage of linolenic and palmitic acids in the initial stages of development of the bean with a simultaneous increase in oleic and linoleic acids, after which the percentage composition of all fatty acids remained relatively constant. The composition of the lipid changed both quantitatively and qualitatively during maturation. In the early stages of development of the bean, the lipid consisted mostly of polar lipid; that of the mature bean mostly of triglycerides. The composition of the polar lipids of the immature bean was characterized by having a relatively large amount of cardiolipin-like compounds and glycolipids. No phosphatidylcholine (PC) or phosphatidylethanolamine (PE) were detected in the lipid until the later stages of development of the bean, but compounds believed to be precursors of these compounds were present in the immature bean. These compounds decreased during maturation and were virtually absent in the ripe bean.

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EFFECTS OF PARATHION ON LIPOLYSIS IN ISOLATED ADIPOSE CELLS.

RICHARD M. CALEY and ROBERT G. JENSEN, Storrs, Connecticut.

The effects of the pesticide, parathion, (0,0-diethyl 0-p-nitrophenyl thiophosphate) on lipolysis in vitro were studied. Isolated fat cells were prepared from epididymal fat pads by treatment with bacterial collagenase. Lipolysis was measured as the release of free fatty acids into an albumin-bicarbonate medium. Parathion was found to depress lipolysis at concentrations ranging from 10^{-2} to 10^{-8} M with its greatest inhibition at high concentrations. The effects of parathion on the action of various lipolytic and antilipolytic agents were also studied. At a concentration of 10^{-5} M, parathion depressed the lipolytic response to epinephrine (0.15 μg/ml) cyclic 3',5'-adenosine monophosphate (1.0 mM) and enhanced the antilipolytic response to nicotinic acid (33 μM).

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PHYLOGENERIC PATTERNS IN THE FATTY ACIDS OF AQUATIC ORGANISMS.

R. G. ACKMAN, Fisheries Research Board of Canada, Halifax, N.S., Canada.

Unicellular marine algae form the bulk of the aquatic biomass but only about two dozens out of thousands of species have been analyzed in detail for lipids or fatty acids. The typical unsaturated fatty acids include palmitoleic acid with very little oleic and *cis*-vaccenic acid and, significantly, no longer-chain monounsaturated acids, and conventional polyunsaturates with *cis*-methylene-interrupted double bond systems. In most cases there is an attempt to form the most highly unsaturated acid possible in each chain length. Accordingly, high proportions of 20:5ω3 and 22:6ω3 are found at this trophic level. Many marine life forms in the invertebrate classes do not lay down large amounts of depot fats as energy reserves. Instead they have mechanisms for slowing down their metabolism in times of food scarcity. The lipids are thus often essentially cellular and rich in phospholipids. There is a strong resemblance between the fatty acids of these lipids and an average fatty acid composition for marine phytoplankters, possibly reflecting direct adoption of fatty acids, or various intact or partially intact complex lipids, through phagocytosis. In higher marine life forms, the muscle tissue contains approximately 1% of phospholipids which again contain fatty acids in patterns resembling those in algae and filter feeders but with modest amounts of oleic acid added. Superimposed on this lipid are the depot fat storage systems. When these are triglycerides they are distinguished from the other lipids by high proportions of C_{20} and C_{22} monoethylenic fatty acids. There are extensive and involved recycling processes for fats in the aquatic environment, and triglyceride fatty acid composition is thus averaged out to a very large degree.

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TAXONOMIC PATTERNS IN THE TRIGLYCERIDE COMPOSITION OF NATURAL FATS.

CARTER LITCHFIELD, Department of Biochemistry and Biophysics, Texas Agricultural Experiment Station, College Station, Texas.

Natural triglyceride mixtures from different organisms frequently have similar fatty acid compositions but widely different triglyceride compositions. These differences in triglyceride composition often fall into clear-cut taxonomic patterns when the distribution of the fatty acids between the 1, 2 and 3 positions of the glycerol is considered. Plant and animal triglyceride exhibit widely different positional distribution patterns in their fatty acids. Palmitic, stearic and fatty acids with chain lengths longer than 18 carbons are esterified almost exclusively at the 1,3-positions in plant triglycerides, whereas animal triglycerides have these acids esterified at all three positions. Seed triglycerides can be divided into at least two categories according to the positional distribution of oleic, linoleic and linolenic acids. In the first category (Gramineae, Juglandaceae, Compositae, and many other plant families), 18:1 and 18:3 are almost equally distributed between the 1, 3 and 2 positions, while 18:2 is somewhat more concentrated at the 2 position. In the second category (Cruciferae), 18:1, 18:2 and 18:3 follow more complex positional distribution patterns which can be described by mathematical equations. Other categories of 18:1/18:2/18:3 distribution may exist, but further work will be necessary to establish this. Characteristic taxonomic patterns have been recognized in the triglycerides of a few groups of animals. Polyunsaturated C_{20} and C_{22} fatty acids are preferentially esterified at the 2 position in fish and invertebrate triglycerides and at the 3 position in marine mammal blubber triglycerides. The distribution patterns for 22:6 and 22:5 are so regular that their positional distribution can be accurately predicted by simple proportionality equations. Bird triglycerides generally have equivalent fatty acid compositions at the 1 and 3 positions, whereas most other animals exhibit unsymmetrical distributions. General tendencies have been noted for the positional distribution of various fatty acids in other animal triglycerides, but clear-cut taxonomic patterns have not yet been defined.

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PHYLOGENETIC RELATIONSHIPS IN LIPID METABOLISM.

JAMES F. MEAD, Laboratory of Nuclear Medicine and Radiation Biology, University of California, Los Angeles, California.

In lipid metabolism, as in other areas, certain basic pathways are common to most groups of organisms. However, superimposed on these are alterations in the basic patterns that are characteristic of the species and may show relationships between species and provide some evolutionary information. Fatty acid synthesis has been studied thoroughly in mammalian and avian liver, in other organs, in yeast and higher plants, and in certain microorganisms, all of which show some revealing differences. In some cases, closely related to synthesis, desaturation takes three main pathways, depending on the particular organism—the aerobic, anaerobic and plant systems. Among the microorganisms, the position of desaturation is species dependent as is the effect of temperature on the extent of the reaction. The beta oxidation system appears to be common to most species in its basic form, but the alpha or one carbon oxidation has an interesting distribution in higher plants and animals. A presently very confusing field is that of plasmalogen and glyceryl ether formation. Depending on the species chosen, the biosynthetic pathway appears to take several different, even opposite routes.

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A COMPARATIVE STUDY OF SPHINGOSINE BASES IN CENTRAL NERVE TISSUE.

KATHLEEN M. GILLILAND and EZIO A. MOSCATELLI, University of Texas Southwestern Medical School, Dallas, Texas.

Total polar lipids, less gangliosides, were prepared from whole brain of rat, chicken, turtle and frog, and from brain plus ventral cord of crayfish. Turtle and fish polar lipids were further fractionated to isolate sphingomyelin, cerebroside and sulfatide. Folch extraction, silicic acid column chromatography and thin layer chromatography (TLC) were used for preparation and identification of ganglioside-free polar lipids. Mild alkaline hydrolysis, various column chromatographic methods, and preparative TLC were used to isolate individual sphingolipid classes. Acid-catalyzed methanolyses were used to prepare sphingosine bases, which were purified and cleaved to aldehydes by periodate oxidation for compositional analysis, using gas-liquid chromatography on diethylene glycol-succinate coated, packed columns. Aldehyde identification was accomplished by standard techniques, including chemical and catalytic reduction and conversion to bisulfate derivatives. In the direction of lower species, the following generalizations can be made for polar lipid sphingosine bases: greater diversity, shorter average chain length, and some increase in saturation. Crayfish is exceptional in that two of its major sphingosine bases are C_{16} sphingosine and C_{14} sphingosine. The latter is a novel compound, essentially peculiar only to crayfish among the species investigated. Turtle whole sphingolipids, which contain C_{18} sphingosine, C_{18} dihydrosphingosine, C_{16} sphingosine, and C_{16} dihydrosphingosine in the ratio 86:6:7:1, reveal different distributions among the individual sphingolipid classes. An analogous situation was encountered in the case of fish sphingolipids. Sphingosine base composition will be compared by sphingolipid class and species, and possible biological significance will be discussed. Also discussed will be limitations of the methodology used and the application of newer analytical techniques.

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PHOSPHOLIPIDS AND GLYCOLIPIDS OF ANIMAL CELL MEMBRANES, ORGANELLES AND ORGANS: SPECIES AND AGE VARIATIONS.

GEORGE ROUSER, City of Hope Medical Center, Duarte, California.

Procedures for accurate, precise determination of the molar amounts of each polar lipid class and the fatty acid composition of each lipid will be reviewed. Data for various species of vertebrates will be shown to demonstrate little or no species variation. The qualitative and quantitative variations in polar lipid composition of invertebrate tissues will be presented. The data show that some invertebrate species substitute either ceramide phosphorylethanolamine or ceramide aminoethylphosphonate for sphingomyelin which is found in all vertebrates. The large changes with age of lipids of human aorta and human brain will be presented.

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BACTERIAL HYDROCARBONS: STRUCTURE AND BIOSYNTHESIS.

PHILLIP W. ALBRO and JOHN C. DITTMER, St. Louis University Medical School, St. Louis, Missouri.

Interest and speculation on the possible role of bacteria in the formation of petroleum deposits date back to the 19th century. Only recently has any definitive studies been done on the characterization of bacterial hydrocarbons and their biosynthesis. We have now established the detailed structure of the major hydrocarbons of a strain of *Sarcina lutea*. These structures and the composition of the lipid fatty acids are most consistent with a biosynthetic mechanism in which the aliphatic groups to either side of the center of the hydrocarbon molecules are derived from fatty acids with decarboxylation of one of the fatty acids. Studies with ^{14}C labeled intermediates both in vivo and in vitro confirm this. However, contrary to Chibnall and his coworkers in the 1930's and others who have proposed for plants that the hydrocarbons may be synthesized by a mechanism in which head-to-head condensation of two molecules of fatty acids with decarboxylation of one of them occurred and in which a long chain ketone and secondary alcohol served as intermediates, the condensation occurs only after one of the fatty acids has been reduced to the oxidative level of an aldehyde and neither ketones or secondary alcohols serve as intermediates. The cofactor requirements and exact nature of the intermediates in this biosynthetic pathway will be discussed.

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PLANT WAXES.

P. E. KOLATTUKUDY, Connecticut Agricultural Experiment Station, New Haven, Connecticut.

The surface of plants is covered with a complex mixture of lipids, often in crystalline form, called plant waxes. The most common components of this wax are paraffins, waxy esters, free fatty alcohols and acids. Ketones, secondary alcohols, diols, aldehydes, terpenes and flavones are also found. The major function of these waxes appears to be protection of the organism from water loss and other hazards of the environment. Young rapidly expanding leaves of tobacco (*Nicotiana tabacum*),

brocholli, cabbage (*Brassica oleracea*), pea (*Pisum sativum*) and spinach (*Spinacea oleracea*) readily incorporated labeled precursors into the waxes. The mechanism of synthesis of paraffins was examined with specifically labeled fatty acids. The carbon chains of palmitic and stearic acid were incorporated intact into nC_{29} paraffin of *B. oleracea* and nC_{31} paraffin of pea and spinach, stearic acid being a much better precursor. Thus, if a head-to-head condensation of these fatty acids results in paraffin synthesis, the other condensing acid must lose its carboxyl carbon. However, doubly labeled C_{12} acid was incorporated into the paraffin without decarboxylation, showing that it does not condense with C_{18} acid to produce C_{29} paraffin. Thus either the exogenous fatty acids can condense only with endogenous acids which specifically lose their carboxyl carbon, or fatty acids produce paraffins by undergoing elongation and decarboxylation. The carbonyl group of the C_{29} ketone, which is supposedly an intermediate in the condensation route, has been shown to be derived not from the carboxyl carbon of the precursor fatty acid but is formed on a specific carbon of the precursor fatty acid but is formed on a specific carbon atom of a preformed saturated carbon chain. Furthermore, labeled C_{18} acid did give rise to C_{20}–C_{28} fatty acids of high specific activity in all paraffin synthesizing plant tissues tested. The formation of these elongation products was affected by light, 3-(4- chlorophenyl-1,1-dimethyl urea) and trichloroacetic acid in the same manner as they affected paraffin synthesis. This and other evidence supports the elongation-decarboxylation mechanism for paraffin synthesis in plants. The fatty acids are reduced to alcohols presumably via the aldehydes. The fatty alcohols are then esterified with fatty acids to give waxy esters. The mechanisms of this esterification are analagous to those involved in cholesterol esterification in animals; namely, acyl transfer from phospholipids and acyl CoA.

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THE CUTICULAR LIPIDS OF INSECTS. LARRY L. JACKSON and GRAEME L. BAKER, Department of Chemistry, Montana State University, Bozeman, Montana.

Knowledge of the constituents of insect cuticular lipid is basic to an understanding of the role of the cuticle in maintaining water balance. Since in most of the insects investigated to date hydrocarbons are the most abundant constituent, characterization of the hydrocarbons is of interest. Detailed analysis of naturally occurring hydrocarbon mixtures is now possible with the use of analytical and preparative gas-liquid chromatography, coupled with mass spectral techniques. In conjunction with hydrocarbon characterization, the metabolic pathways available for hydrocarbon biosynthesis are being investigated. Hydrocarbon biosynthesis is an area of lipid metabolism in which little has been done. Since more information is available on hydrocarbon biosynthesis in plants, the studies on insect hydrocarbon biosynthesis will be compared to the systems found in plants. Chemical taxonomy utilizing hydrocarbon patterns will also be discussed.

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DETERMINATION OF THE COMPOSITION OF UNHYDROLYZED BEESWAX. A. P. TULLOCH, National Research Council of Canada, Ottawa, Canada.

Beeswax has been fractionated on a silicic acid column into hydrocarbons (15%) and esters A (35%), esters B (12%), esters C (4%), esters D (8%), esters E (10%) and free acids (8%). Esters A are composed of C_{40} to C_{50} monoesters and esters B of diesters containing acylated 15-hydroxypalmitate. Esters of 15-hydroxypalmitic acid in which the hydroxyl group is not acylated are present in esters C, D and E. The fatty acid composition of the different esters will be discussed. The configurational relationship of the 15-hydroxypalmitic acid of beeswax to the acid of the same strucure previously isolated from natural sources has been investigated.

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THE OCCURRENCE, FUNCTION AND BIOSYNTHESIS OF WAX ESTERS IN MARINE ORGANISMS. JUDD C. NEVENZEL, University of California, Los Angeles, California.

The long-chain monohydric alcohol esters of fatty acids (i.e., the wax esters) occur widely among marine organisms. They are the principal lipid type in at least some calanoid copepods (crustaceans), several families of fishes, and the sperm whale. The wax esters found in the eggs of the mullets and gouramis, which contain predominantly polyunsaturated fatty acids C_{16}–C_{22} esterified with saturated and monounsaturated *n*-alkanols C_{14}–C_{18}, probably are primarily reserve energy stores from the developing embryos and larvae. In a second group of fishes and in the shales the wax esters are present in massive amounts (up to 16.5% of the fresh weight) in the muscle tissue, and are in the range C_{28}–C_{40}, predominantly mono- and diunsaturated. The alcohol moieties again are C_{14}–C_{20} saturated and monounsaturated, but the acids contain at most a few per cent of polyunsaturated components. In these species (many, perhaps all of which are mesopelagic fishes which make extensive diurnal vertical migrations in the open ocean) the wax esters are primarily a buoyant agent, the presence of large amounts of this lipid type correlating with deeper living and vertical migration, not with taxonomic relationships. Preliminary investigation of the biosynthetic of wax esters in fishes has established that they incorporate exogenous acetate preferentially into the alcohol as compared to the acid moieties of the wax esters, and in the same ratio (90:10) as from labeled 1-dexadecanol. Palmitic and oleic acids are handled differently, with the former labeling equally the acid and alcohol moieties, but the latter being incorporated almost exclusively (95%) into the fatty acid portion of the wax esters.

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WAXES OF ANIMAL SKIN SURFACES. N. NICOLAIDES, HWEI C. FU and M. N. A. ANSARI, University of Southern California, Los Angeles, California.

A review of the literature of the lipid composition of animal skin surfaces will be presented. New data on the types of wax ester classes and their analysis will be reported with special emphasis on the diester wax content. Possible functions of these lipids will be discussed.

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SULFONATE FLOTATION OF PHOSPHORITE AND COMMONLY ASSOCIATED MINERALS. M. C. FUERSTENAU, R. O. JUAREGHI and J. D. MILLER, Department of Metallurgy, University of Utah, Salt Lake City, Utah.

Electrokinetic and micro-flotation experiments were conducted to establish the mechanisms involved in the adsorption of sulfonate on apatite, calcite, dolomite and quartz. High molecular weight sulfonates chemisorb on apatite, calcite and dolomite. Sulfonates adsorb on quartz only when contained in a basic aqueous complex of metalsulfonate, such as $Ca(OH)(RSO_3)_{aq}$. Flotation of natural phosphate rock ores is also presented. The molecular weights of sulfonate and oil required have been determined. Minimum molecular weight of sulfonate lies between the range of 360 and 420. The effect of oil additions is most pronounced in the cleaning stages of flotation. Reasons why high oil viscosity is beneficial in these systems are presented.

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THE ROLE OF SURFACTANTS IN THE FLOTATION OF MOLYBDENITE AT CLIMAX. RICHARD A. RONZIO, Climax Molybdenum Company, Mines Park, Golden, Colorado.

The use of petroleum hydrocarbon as a collector for molybdenum sulfide improved metal recovery but necessitated the investigation of surfactants to produce a suitable froth for flotation at the mill of American Metal Climax, Inc. Up to December 1958, 346 surfactants had been tested and evaluated. A sulfated monoglyceride of coconut oil is presently employed at Climax. The study of the role of surfactants for molybdenum sulfide flotation is continuing. One of the better materials evaluated in the last 10 years is ethoxylated, esterified and neutralized lauryl alcohol. Longer and shorter chain alcohols ethoxylated, esterified and neutralized have also been evaluated with encouraging results.

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THE EFFECTS OF SURFACTANTS AND ALCOHOLS ON THE FLOTATION COLLECTION PERFORMANCE OF AN ALKYL THIONOCARBAMATE. D. J. COLLINS and T. F. IZZO, The Dow Chemical Co., Walnut Creek, California.

An investigation of the effects of various alcohols and surfactants on the performance of isopropyl ethyl thionocarbamate for the flotation of copper minerals from two porphyry ores from the Southwestern USA will be presented. The surfactants tested were the sodium and calcium salts of dodecyldiphenyl ether disulfonic acid, and the sodium salt of petroleum sulfonate. Methyl, ethyl, isopropyl and methyl amyl alcohols as well as propylene glycol methyl ester were used as diluents to promote spontaneity of emulsification of the collector. The results indicate that thionocarbamate diluted with methyl alcohol produces superior metallurgical results to either undiluted thionocarbamate or to thionocarbamate diluted with other solvents. Petroleum sulfonate was indicated to be the superior surfactant.

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SOME OBSERVATIONS ON THE EFFECTS OF SURFACTANTS IN THE MICROBIOLOGICAL LEACHING OF LOW-GRADE SULFIDE ORES. J. P. KASS, Technical Counsel for the Food and Chemical Industries, Milwaukee, Wisconsin.

The economies inherent in the microbiological leaching of low-grade copper, uranium and other metal-sulfide ores have focused attention on possible methods of improving the rate and efficiency of this empirically established procedure to the levels characteristics of the more costly conventional chemical hydrometallurgical extractive processes. Since the rate of proliferation of the effective sulfide-oxidizing organisms and their elaboration of an essential surface active gap-bridging substance are among the basic rate-governing factors, the addition of extraneous synthetic surfactants presents one logical approach, among others, to the potential acceleration of the necessarily slow biological process. Published laboratory studies have indicated that selected hydrophilic surfactants, among which Tween 20 was found most effective, do indeed appear to have a beneficial effect under specific conditions. However, this effect is limited to only very few minerals and exhibits anomalies, so that the approach remains technically and economically questionable. This paper will attempt to correlate the experimental data with considerations of the fundamental structures of the added surfactants, the mineralogical aspects of the substrate ores, and the cultural characteristics of the autotrophic organisms as regards size and charge of the inoculum, oxygen requirements, and degree of agitation determined by extraction in shake flasks, stationary flasks or percolators. Certain types of surfactants with potentially less erratic behavior than those previously reported will be suggested for future investigation.

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EFFECT OF SURFACTANTS ON THE RHEOLOGY OF HEMATITE SLURRIES. HARLEY Y. JENNINGS, JR., Chevron Oil Field Research Co., La Habra, California.

The rheological behavior of aqueous hematite slurries has been studies by laboratory viscosity and flow measurements. Data are presented for slurries with volume concentrations of hematite ranging from 28% to 40%. Solids concentration and particle size distribution were found to be important variables in rheological behavior of hematite slurries as contrasted to temperature which was found to have only a small effect. Chemical dispersant additives and viscosity control additives used in relatively small concentrations improved the flow characteristics of the hematite slurries. Specifically, a lignosulfonate dispersant markedly increased the flow of slurries containing finely divided hematite, −325 mesh, and a viscosity control additive increased the carrying capacity of other slurries for course particles.

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SURFACTANTS FOR OIL RECOVERY. G. P. AHEARN, Esso Production Research Co., Houston, Texas.

In recent years the high cost of searching and drilling for oil has emphasized the importance of increasing production efficiency from known petroleum reserves. Currently, it is technically possible to recover only 47% of this country's estimated total oil in place. It is anticipated that steady improvement in technology may eventually bring crude oil recovery up to 65% of the oil in place. From these facts it is apparent that oil recovery is a relatively inefficient process. It is estimated that there are about 250 billion barrels of oil in this country currently deemed economically unrecoverable. The recovery of a small fraction of this oil provides an enormous economic incentive for the oil industry. Water injection has been the most successful additional recovery process developed in the last century. It is predicted that by 1975, 41% of the nation's crude output will be recovered by this technique. For years the idea of developing a chemical that could be added to the injected water to increase ultimate oil recovery has intrigued the entire industry. This paper deals with recent attempts by the industry to develop surfactants to recover the oil that remains after conventional recovery operations.

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N-PHENYLAMINOMETHYLATION: A ONE-STEP ROUTE TO N-SUBSTITUTED ANILINES FROM UNSATURATED FATTY DERIVATIVES. ROBERT A GRIMM, Ashland Chemical Company, Bloomington, Minnesota.

Unsaturated fatty materials are found to react with carbon monoxide, hydrogen and aniline to give N-monoalkylanilines. The alkyl group is derived from the unsaturated fatty material plus the group H-CH_2 added across the double bond. Ester and nitrile groups do not interfere with the reaction but carboxyl groups do. Rhodium complexes are the catalysts of choice for this reaction.

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OLEYL ALCOHOL FROM ANIMAL FATS BY CATALYTIC HYDROGENOLYSIS. R. S. KLONOWSKI, T. W. FINDLEY, C. M. JOSEFSON and A. J. STIRTON, Swift & Co., Oak Brook, Illinois.

Catalysts composed of ternary and quaternary combinations of chromium, zinc, cadmium, aluminum, barium and copper were used in a study of the hydrogenolysis of fatty esters and acids. High selectivity (per cent hydrogenolysis/per cent hydrogenation) was obtained with aluminum, cadmium and barium modified zinc chromites and catalysts composed of zinc, cadmium and copper. Parameters studied were temperature (350 to 450 C), pressure (2500 to 3500 psi) and the ratio of metals in the catalyst. Catalysts found useful for the reduction of unsaturated fatty methyl esters were generally found to be even more effective for the corresponding acid. Triglycerides, however, gave a product which was interior to that obtained with the methyl ester or acid. Products were analyzed by gas liquid chromatography and shown to contain in addition to the expected products mono- and diunsaturated hydrocarbons and fatty ethers, both saturated and unsaturated. All reaction products were accompanied by an unchromatographical product of higher molecular weight than the starting material. Studies to identify these by-products will be discussed.

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HF CATALYSIS I—A NOVEL SYNTHESIS OF GLYCEROL MONOESTERS. EUGENE J. MILLER, HARLAN E. TIEFENTHAL and AGO MAIS, Armour Industrial Chemical Company, McCook, Illinois.

Reaction of triglycerides with glycerol has, in the past, always resulted in an equilibrium distribution of mono-, di- and triesters. It has now been demonstrated that glycerol monoesters may be prepared directly using liquid, anhydrous hydrogen fluoride as a unique solvent-catalyst system. Glycerol monoesters of greater than 95% activity have been prepared consistently from virtually stoichiometric proportions of hydrogenated tallow and glycerol. Alternatively, direct esterification of glycerol with free acids in anhydrous hydrogen fluoride may also be used. Work with other polyols and acids has demonstrated the usefulness of this unique solvent-catalyst as a versatile tool for preparation of a broad range of polyol monoesters.

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COMPETITIVE HYDROGENATION RATES OF ISOMERIC METHYL OCTADECENOATES. C. R. SCHOLFIELD, T. L. MOUNTS, R. O. BUTTERFIELD and H. J. DUTTON, Northern Regional Research Laboratory, ARS, USDA, Peoria, Illinois.

Hydrogenation rates of isomeric methyl octadecenoates were compared by reducing mixtures containing a small weight of one radioactively labeled isomer and a much larger weight of another unlabeled isomer. As the hydrogenation proceeded, samples were removed for analysis by a gas chromatograph coupled with an ionization chamber so that the mass peaks measured the reduction of the unlabeled isomer and the radioactive peaks of the labeled isomer. The ratio of hydrogenation rate best fitting these data was calculated with a digital computer. Reductions were made at atmospheric pressure mostly with either nickel at 140 C or platinum at 50 C. A few were made with palladium at 50 C. In typical experiments methyl oleate hydrogenated more rapidly than methyl elaidate and the *cis*-15-isomer more rapidly than oleate. The *cis*-12 isomer reduced more slowly than oleate with platinum or palladium but more rapidly with nickel. The *cis*-6-isomer reduced more rapidly than oleate with platinum but at about the same rate with palladium or nickel.

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HOMOGENEOUS CATALYTIC CONJUGATION OF POLYUNSATURATED FATS BY METHYL BENZOATE-Cr(CO)$_3$. E. N. FRANKEL, Northern Regional Research Laboratory, ARS, USDA, Peoria, Illinois.

Previous work showed that arene-Cr(CO)$_3$ complexes are effective homogeneous catalysts for the hydrogenation of polyunsaturates in soybean oil. Now methyl benzoate-Cr(CO)$_3$ has been found to be one of the most effective soluble catalysts for the conjugation of polyunsaturated fats. The conjugation procedure involves heating fatty esters or oils in an autoclave under N_2 pressure at 175–185 C with 5–10 mole % catalyst in solution (cyclohexane) or without a solvent. By this treatment methyl linoleate was 65% conjugated and methyl linolenate 50% conjugated. Up to 60% of the polyunsaturates in soybean oil were conjugated. Conjugated dienes were principally *cis,trans*. Trienes contained approximately 30% diene conjugation and 20% triene conjugation. Formation of monoenes and conjugated trienes from ethyl linoleate indicates that hydrogenation and dehydrogenation are side reactions that limit the yield of conjugated dienes. With chromium carbonyls conjugation proceeds apparently by a hydride shift from substrate to catalyst.

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REDUCTIVE AMINATION OF 12-KETOSTEARIC ACID. BERNARD FREEDMAN and GLENN FULLER, Western Regional Research Laboratory, ARS, USDA, Albany, California.

The preparation of 12-aminostearic acid by reductive amination of 12-ketostearic acid has been studied. Two steps are involved in the process. The keto group is contacted with ammonia under pressure and the resulting intermediate is hydrogenated to give the product. Variables such as time and temperature of reaction, hydrogen pressure, and amount and type of catalyst were examined to find optimum conditions for high yield and purity of 12-aminostearic acid. By proper choice of reaction variables the product was obtained in greater than 90% purity and yield. Gas liquid and thin layer chromatography were used to determine purity of the reaction products.

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THERMAL POLYMERIZATION OF SAFFLOWER SEED OIL. YESHWANT K. PURANDARE, State University A&T College, Farmingdale, New York.

Thermal polymerization of safflower seed oil was studied to investigate its rate, extent and nature of polymerization. It was polymerized in an inert atmosphere of carbon-dioxide at different temperatures ranging from 200 to 300 C for 12 hr. In order to know the progress of reaction, a sample was drawn out every 3 hr, and its various physical and chemical characteristics were determined. The partially polymerized oil samples were subjected to solvent fractionation. The polymerized oil samples were tested for their film properties, such as drying time, resistance toward water, solvent and alkali; it was found that the samples showed betterment in these properties. The oleo-resinous varnishes prepared from them showed properties comparable to those of dehydrated castor oil and raw linseed oil.

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CATALYTIC ISOMERIZATION OF SAFFLOWER SEED OIL. YESHWANT K. PURANDARE, State University A&T College, Farmingdale, New York.

Safflower seed oil is a good drying oil (I.No. = 145.0) and contains 64.6% linoleic acid as its main constituent. Its utility as a drying oil can be increased by catalytic isomerization. In the present investigation, safflower oil was isomerized using the following catalysts: iodine in absence of a solvent; iodine in presence of a solvent; hydriodic acid (55%); iodic acid; nickel-carbon; sodium hydroxide; iodoform; iodoacetic acid; periodic acid; anthraquinone; phenanthraquinone; metallic iodides; metallic oxides. Of these the first six catalysts were found to bring about substantial isomerization in safflower seed oil. The various isomerized oil samples were tested for their film properties to estimate the improvement in the oil due to conjugation. Oleo-resinous varnishes were also prepared by copolymerizing these samples with Bedesol A66. The isomerized oil samples compare very well with dehydrated castor oil and raw linseed oil in their drying properties.

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URETHANE FOAMS FROM ANIMAL FATS: V. FLAME RESISTANT FOAMS FROM HYPOHALOGENATED GLYCERIDES. F. SCHOLNICK, E. J. SAGGESE, A. N. WRIGLEY and G. R. RISER, ARS, USDA, Philadelphia, Pennsylvania.

A series of urethane foams has been prepared using hypohalogenated derivatives of triolein, monoolein, lard and tallow as the polyol ingredient. Hypochlorination was effected by liberation of hypochlorous acid from calcium hypochlorite. Hypobromination was obtained by use of N-bromoacetamide. Hypohalogenation could also be achieved by expoxidation of the glyceride followed by treatment with HX. The polyols, which varied from viscous liquids to semi-solids, were adjusted in equivalent weight with triisopropanolamine. Urethane foams were prepared from the adjusted polyols using PAPI as the isocyanate, triethylene diamine catalyst and Freeon 11 as blowing agent. Additional foams were made with 2% antimony oxide as an added fire retardant. Rigid foams of density 1.50–2.30 lb/ft^3 were obtained from each glyceride. Fire retardant properties were measured using a modification of ASTM Method D1692-59T. In each case, the foams exhibited greater flammability resistance than those obtained from glycerides containing on halogen atoms. It was noted that the presence of animony oxide was necessary in order to attain non-burning foams but was accompanied by a lowering of compressive strength.

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EFFECTS OF DIET ON CHOLESTEROL METABOLISM IN RABBITS. K. K. CARROLL, Collip Medical Research Laboratory, University of Western Ontario, London, Ontario, Canada.

Studies with rats have shown that liver slices from animals fed commercial diet incorporate more $1\text{-}^{14}C$-acetate into cholesterol than those from animals fed semisynthetic diet, and that addition of unsaturated fats to either diet stimulates incorporation into cholesterol more than addition of saturated fats. Similar experiments have now been carried out with young male rabbits. Diets were fed for two-week periods and plasma cholesterol levels were also measured when the animals were killed. The average plasma cholesterol was 200 mg/100 cc in rabbits fed fat-free semisynthetic diet. When 15% by weight of different saturated fats was added to this diet the cholesterol levels ranged from 190 mg/100 cc for beef tallow to 375 mg/100 cc for coconut oil. Olive oil gave a level of 160 mg/100 cc while addition of polyunsaturated oils gave values varying from 75 mg/100 cc for corn oil to 115 mg/100 cc for cottonseed oil and rapeseed oil. Rabbits on ether-extracted commercial diet had plasma cholesterol levels of only 50 mg/100 cc and addition of either saturated or unsaturated fats raised the level above 100 mg/100 cc only in a few cases. The highest level, 125 mg/100 cc, was obtained with coconut oil. Acetate incorporation into cholesterol by liver slices was generally lower on semisynthetic than commercial diet but the results were quite variable and there seemed to be no consistent differences between diets containing saturated and unsaturated fats. Acetate incorporation into fatty acids, on the contrary, tended to be higher on the semisynthetic diets. With both semisynthetic and commercial diets, incorporated into fatty acids was highest when the diets contained no added fat. Addition of casein to commercial diet in the ratio of one to three inhibited acetate incorporation into cholesterol and caused an elevation of plasma cholesterol levels when the diet contained no fat or saturated fats but not when it contained polyunsaturated fats. Adding dextrose to the commercial diet in the same proportions seemed to have no effect on either plasma cholesterol or acetate incorporation.

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DIETARY INDUCED CHANGES IN CANINE ERYTHROCYTE FATTY ACID COMPOSITION. ANTANAS BUTKUS, L. ALLEN EHRHART, LENA A. LEWIS and F. MERLIN BUMPUS, Cleveland Clinic Foundation, Cleveland, Ohio.

An atherogenic diet containing 16% of hydrogenated coconut oil and 5% of cholesterol was fed to dogs for several months. Changes in the fatty acid composition of red blood cell (RBC) lipids were compared to those in the hyperlipemic plasma. Concentration and relative per cent distribution of fatty acids (FA) were determined in various lipid fractions. The per cent distributions of phospholipid and triglyceride FA were similar in RBC and plasma but free FA patterns differed. The percentage of free linoleic acid decreased in hyperlipemic plasma but increased in the RBC whereas the opposite occurred with palmitoleic. The total FA concentration in RBC lipid was unaffected by hyperlipemia but lauric acid, which comprised nearly 40% of the dietary total FA, increased in the free FA fraction, while in the triglycerides, palmitoleate increased. Phospholipids FA were the most affected by the diet. Monounsaturated FA (oleate and palmitoleate) were significantly elevated in RBC

phospholipids. Increases in laurate and myristate and reductions in palmitate and stearate were also observed. The net effect was an overall decrease in the phospholipid saturated as well as total FA to 69% of their control RBC concentrations. Polyunsaturated FA were reduced; linoleate fell to 50% of control concentrations and arachidonate to 25%. Although total lipid and total FA concentrations in the hyperlipemic plasma rose 10-fold, there were no increases in the corresponding fractions in RBC from dogs on the atherogenic diet. The main change in RBC lipids resulting from the diet, therefore, may possibly be an alteration of membrane FA patterns as evidenced by the striking changes primarily in phospholipid FA.

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EFFECT OF DIETARY CHOLESTEROL ON THE COMPOSITION, MORPHOLOGY AND FUNCTIONS OF GUINEA PIG RED CELLS. R. OSTWALD, W. YAMANAKA, J. KROES and M. LIGHT, Department of Nutritional Sciences, University of California, Berkeley, California.

The hemolytic anemia produced in guinea pigs by dietary cholesterol presents a good tool to study the interrelationships between composition, function and morphology of cell membranes. The composition of red blood cells of cholesterol-fed, anemic guinea pigs was changed drastically with a doubling of the amount of unesterified cholesterol, a decrease of the ratio of phospholipids to cholesterol, increased proportions and amounts of lysolecithin compensated by decreases in sphingomyelin and changes in the fatty acid pattern of the individual phospholipid classes. Light- and electronmicroscopy showed that the morphology of these cells was greately altered. Cell sizes ranged from relatively small burr cells to large reticulocytes. The burr cells showed a few large spicules and had a roughened and pebbly surface. Microelectrophoretic mobility data showed that the surface charge concentration from pH 1–10 remained unaltered. The measurement of sedimentation rates and osmotic fragility indicated a great heterogeneity in the population of cells from cholesterol-fed guinea pigs. Efflux of sodium and permeability to glycerol, thiourea and monoacetin was decreased. The increased amounts of lipids and the changes in the relative proportions of cholesterol to phospholipid, of the individual phospholipid classes and of their fatty acids may have a profound effect on both the function properties and on the morphology of these cells.

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EFFECT OF DIETARY FAT LEVEL AND CALORIC INTAKE ON BODY WEIGHT, DISTRIBUTION OF FAT DEPOTS AND FATTY ACID COMPOSITION OF CARCASS, LIVER AND ADIPOSE TISSUE OF RATS. MILDRED J. BENNETT and SHIRLEY BARBER, Children's Hospital Medical Center, Oakland, California.

Weanling rats of the Long-Evans strain were fed purified diets providing 0%, 11% or 45% of the calories as hydrogenated coconut oil. Linoleic acid was provided by the 7–8 drops of safflower oil fed per week. After growth rate reached a plateau, the rats were grouped and fed isocaloric amounts of the different diets in unrestricted amount or in amounts restricted to 66% or 79% of the full complement of calories. The amount of total fat deposits were similar for similar caloric intake regardless of fat content in the diet. The relative amounts of abdominal, epididymal and mesenteric fat deposits did not change with the decrease in total amount of adipose tissue when restriction of calories was imposed. Caloric restriction produced no changes in fatty acid composition of liver, carcass or adipose tissues. The fatty acid composition of adipose tissue and carcass readily reflected differences in fat intake of the rat, but changes in liver were minimal. There were sex differences in the effect that the level of dietary fat had on the fatty acid composition of adipose tissue.

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NUTRITIONAL EVALUATION OF FILLED MILKS IN MONKEYS AND RATS. HANS KAUNITZ and JAIME SANYEE, Department of Pathology, College of Physicians and Surgeons, New York, New York.

The present experiments were carried out with samples of filled milks prepared by the research laboratories of a dairy company, and careful attention was given to preventing the deterioration of the samples. The following filled milks were studied: (a) skim milk + 3.2% of milk fat; (b) skim milk + 3.1% of hydrogenated coconut oil; (c) samples b + 0.2% of cottonseed oil; (d) sample c + 0.8% of buttermilk solids; (e) sample c + 5% of sample a; and (f) skim milk + 3.1% of soybean oil. Matching groups of rats were given the milk samples ad lib and 3 g of rat chow daily for six months and then killed. Their daily milk intake was measured. Weight gain and general appearance was equally good in all groups. Average serum and liver cholesterol and liver total lipid values were somewhat higher in the group fed milk fat than in those fed coconut oil. The highest values occurred in the group given soybean oil. Eighteen cynemologus monkeys were given unlimited amounts of the milk samples (after weaning at two to three months) and they were offered 5–10 g of monkey chow once each day. They were observed for general appearance, occurrence of diarrhea, milk intake, weight gain and intestinal flora (stool cultures). All filled milks gave equally good results.

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SEX DIFFERENCES IN INTESTINAL PROTEIN SYNTHESIS AND ABSORPTION OF LIPIDS. GEORGE V. VAHOUNY, M. ITO and C. R. TREADWELL, School of Medicine, The George Washington University, Washington, D.C.

The incorporation of 1-^{14}C-leucine into mucosal protein of everted intestinal sacs was higher in tissues from female animals than from males. Two weeks after castration, protein synthesis was significantly reduced, and was returned to normal by injection of testosterone in vivo or by its addition to the sac preparation in vitro. Similar results were obtained with female rats after ovariectomy and estradiol administration in vivo and in vitro. Ovariectomy also resulted in marked depression of both cholesterol and fatty acid absorption in lymph duct cannulated animals. When puromycin was used to inhibit protein synthesis in vivo, the absorption of cholesterol and oleic acid into lymph in female rats was markedly depressed. However, under identical conditions with male rats, only the absorption of cholesterol was inhibited. Measurement of lipid accumulation in the mucosa of these animals substantiated the apparent differences between sexes in the response to puromycin. The data suggest a possible sex difference in mucosal protein synthesis, which may explain the differences between sexes in alterations of lipid transport due to puromycin administration.

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EFFECT OF DIETARY STERCULIC ACID ON LIPID COMPOSITION OF RAT TISSUES. BARRY J. BURNS, ROSLYN B. ALFIN-SLATER and JAMES F. MEAD, University of California, Center for Health Sciences, Los Angeles, California.

Cyclopropenoid fatty acids (e.g., sterculic acid) found in cottonseed and *Sterculia foetida* oil have been reported to increase the amount of stearic acid at the expense of oleic acid in the lipids of animal tissues. Since the polyunsaturated fatty acids in fat-free animals are derived from oleic acid, our investigation was undertaken to determine whether the C20 triene normally found in tissue lipids of rats fed fat-free diets would be decreased in the presence of sterculic acid. Male weanling albino rats were placed on a fat free diet and allowed to eat ad lib. During the 2–12 week feeding period, groups of animals were dosed with 20, 40 or 60 mg/day per rat of sterculic acid as the methyl ester. The growth rate of the animals decreased as the amount of sterculic acid in the diet was increased from 0–60 mg/day. The animals receiving the 60 mg/day gained only two thirds of the weight gained by the control animals. Animals receiving 20 mg/day showed only slight differences in total lipid content of the liver compared to the fat free-fed animals. Within two weeks the liver and adipose tissue fatty acids showed increased amounts of stearic acid and decreased amounts of oleic, palmitoleic and 5,8,11 eicosatrienoic acids. After 12 weeks of daily sterculate rations, the amount of stearic acid in the liver increased two- to threefold over the control animals with corresponding decreases in oleic and palmitoleic acids. In adipose tissue the differences observed were similar but more pronounced.

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LIPID MONOLAYERS: INFLUENCE OF CHEMICAL STRUCTURE ON SURFACE PROPERTIES. GIUSEPPE COLACICCO, City University of New York, Flushing, New York.

Structural differences among lipids are responsible for differences in the properties of their films at the air/water interface: Surface pressure (π), surface potential (ΔV), surface viscosity (η_s), film penetration ($\Delta\pi$), and accessibility to enzymes. Some physical meaning is derived from the surface potential equations, in which ΔV is proportional to π/dielectric constant. At a given temperature and area/molecule: Hydrophobic chains, salt linkages (of phosphoryl choline), ester and amide bonds produced low dielectric constant and large ΔV values. OH groups which are surface bound and parallel to the interface (as in sphingomyelin and glycosphingolipids) caused copolymerization structuring of uppermost water layers, thus high η_s, high dielectric constant of film, and low ΔV. OH groups which extend perpendicularly into subphase (as in cholesterol and long chain alcohols), produce little surface viscosity but large surface potential. Unsaturation in the middle of hydrophobic chain causes increased contact with water, greater surface valence, thus greater π, but lower ΔV because of increased dielectric contant of electron rich film. Regions of structured water within hydrophobic molecular contours raise dielectric constant and lower ΔV but cause little change in π or η_s; replacement of ester bond by ether or $\alpha\beta$-unsaturated ether causes marked decrease of ΔV, typical of plasmalogens. In accord with a previous observation at the oil/water interface, the appearance of a negative or positive charge at the water interface in contact with the negative pole of the electrometer causes respectively a decrease or increase of ΔV. This finding is now used to detect ionic impurities in neutral lipids. It is demonstrated that the observations which Shah and Shulman have attributed to dipole effects are actually due to these ionic impurities. In the interaction with proteins, at pH 7.0 and 25 C, rate and extent of lipid film penetration ($\Delta\pi$) by serum albumin vary: cholesterol > lactoside > phosphatidyl choline > sphingomyelin > ganglioside. The mechanisms of penetration are: Free for phosphatidyl choline, binding-mediated for cholesterol and lactoside, and binding-inhibited for sphingomyelin and ganglioside.

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BLM AS BIOLOGICAL MEMBRANE MODELS: AN EVALUATION AND A SUGGESTION FOR ITS SPONTANEOUS FORMATION IN NATURE. H. T. TIEN, Michigan State University, East Lansing, Michigan.

For the past several years an experimental model of the cellular membrane known usually as BLM (biomolecular, bilayer, or black lipid membrane) has been extensively studied by several groups of workers. Among the properties investigated, these include chemical composition of lipid solutions for the BLM formation, optical measurement of thickness, electrical parameters including transient phenomena, water and other solute permeability, bifacial tension and excitation of BLMs by light. The present paper attempts a critical evaluation of the experimental data on the BLM in view of our current knowledge of natural membranes. In addition, a scheme for the spontaneous generation of a BLM in nature is proposed.

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EFFECT OF CHOLESTEROL ON PERMEABILITY AND ELECTRICAL PROPERTIES OF PHOSPHOLIPID MODEL MEMBRANES. D. PAPAHADJOPOULOS and S. OHKI, State University of New York at Buffalo, Buffalo, New York.

The presence of cholesterol has a significant effect on the permeability of phospholipid model membranes. Generally it improves the stability of such membranes and decreases their permeability to ions and water. Using sonicated liquid-crystalline vesicles, it was found that cholesterol mixed with lecithin in 1:1 molar ratio reduces the self-diffusion rate for Cl^- 14-fold. Using planar bilayer membranes, it was found that the presence of cholesterol increased the capacitance from 0.37 to 0.65 $\mu F/cm^2$. In addition, it has been found that when cholesterol is mixed with phosphatidylserine in 1:1 molar ratio, it abolishes the ability of Ca^{2+} to initiate permeability changes. The above results will be discussed in terms of molecular orientation and their possible implications on the structure and function of biological membranes.

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FRACTURE SURFACES IN BULK-PHASE LIPID SYSTEMS. DAVID W. DEAMER, University of California, Berkeley, California.

Various bulk-phase lipid systems were examined by freeze-fracture surfaces will be discussed with reference to images synthetic and natural phospholipids typically fractured along lamellar planes which presumably represent cleavages of weakly bonded hydrophobic areas within the crystal. Lipid mixtures

such as total chloroplast lipid presented fracture surfaces which could best be interpreted as fractures through hexagonal crystalline phases. Surface textures of fractured phospholipids depended on whether saturated or unsaturated hydrocarbon chains were present. Synthetic saturated lecithin had relatively smooth fracture surfaces, whereas unsaturated natural lecithin was rougher in appearance. Particles were apparent on the fractured surfaces of several lipids, such as cholesterol, dipalmitoyl lecithin and fractured stearic acid bilayers. The significance of these fractures surfaces will be discussed with reference to images obtained from freeze-fracturing and freeze-etching of biological membrane systems.

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FILTRATION IN THE FATS AND OILS INDUSTRY. ROBERT J. ZILLI, Johns-Manville Corporation, New York, New York.

Filtration is one of the most necessary and important steps in the processing of fats and oils. Unfortunately, it is still more an art than a science and therefore, frequently much abused. The selection of the proper filter and filter septum depending upon where it will be used in the process is just the beginning in insuring good filtration results. The grade and quantity of filter aid used for precoating and body feeding are extremely important factors for proper filter operation in guaranteeing adequate flow rates and clarities. For example, in the removal of nickel catalyst, the use of faster flow rate filter aids is sometimes false economy depending upon the type of filter used. These problems can also appear during bleaching clay removal if the particle size of the filter aid is too large compared to that of the clay. Understanding of the variables involved can lead to better filtration, improved economies and insured high product quality.

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DEODORIZATION OF FATS AND OIL IN COMMERCIAL PRACTICE. GEORGE KREUTZER, Swift & Co., Chicago, Illinois.

Abstract not available at press time.

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HYDROGENATION OF SOYBEAN OIL WITH COPPER-CHROMITE CATALYST: WINTERIZATION OF LOW-LINOLENATE OILS. K. J. MOULTON, R. E. BEAL and E. L. GRIFFIN, JR., Northern Utilization Research and Development Division, ARS, USDA, Peoria, Illinois.

Soybean oil hydrogenated in the presence of copper-chromite catalysts to a low linolenate level must be winterized to pass the cold test. Winterization yields were therefore studied. Soybean oil was sampled at intervals during partial hydrogenation on a pilot-plant scale with a commercial copper-chromite catalyst. Samples were post-bleached under vacuum in the laboratory to remove dissolved copper, and then placed in a low-temperature box at 5 C for 48 hr before being filtered. These filtered winter oils passed the standard cold test. Partially hydrogenated oils having less than about 2% linolenate, as analyzed by ultraviolet spectrophotometery, required winterization. At a linolenate level of 0.4% the winter oil yield was about 85%. By comparison, soybean oil hydrogenated to 3% linolenate in the presence of a nickel catalyst yielded only 66% winter oil. The data show that after linolenate is substantially eliminated from soybean oil by hydrogenation with a copper-chromite catalyst, the yield of winterized oil is high.

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THE EFFECTS OF HYDROGENATION PROCESS VARIABLES ON THE SELECTIVITY AND ISOMERIZATION CHARACTERISTICS OF NICKEL CATALYSTS. ROBERT R. ALLEN, M. C. MOORE and J. E. COVEY, JR., Anderson, Clayton & Co. Foods Division, Richardson, Texas.

The selectivity ratio (SR) of a catalyst depends on the inherent characteristics of the catalyst and also the temperature, pressure, concentration and agitation of the hydrogenation used to determine the SR. A study of the SR of catalysts over a wide range of process conditions resulted in a second degree polynomial equation developed over the response surfaces calculated from the SR of many experimental hydrogenations of soybean oil. These response surfaces show the interactions of the four process variables and their effect on the SR.

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POSITIONAL AND GEOMETRICAL ISOMERIZATION DURING PARTIAL HYDROGENATION OF TRILINOLEIN—A COMPARISON OF COPPER AND NICKEL CATALYSTS. EHUD KIRSCHNER and E. R. LOWREY, The Procter & Gamble Co., Cincinnati, Ohio.

A comparison of a nickel with a copper type of catalyst is made in their effects on some positional and geometric isomers formed during the potential hydrogenation of trilinolein. Differences in the amounts and type of isomers formed are found between the two types of catalyst. These differences are described. A postulated mechanism for hydrogenation of polyunsaturated fatty acids with copper catalyst is presented based on literature, the trilinolein data, and other data from hydrogenation of soybean oil.

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PRESERVATION OF LIPIDS WITH MALIC ACID. S. EDMUND BERGER, CASIMIR V. KROLEWSKI and LESLIE C. WIZEMANN, Allied Chemical Corporation, Buffalo, New York.

Published literature on the use of malic acid as a preserving agent in lipids shows this acid to have a wide range of effectiveness. Whereas some authors reported the performance of malic acid to be excellent, others, working with different lipids and under different conditions, did not. The present paper deals with an evaluation of malic acid in six different lipids of animal and vegetable origin under comparable conditions. An AOM procedure was used, with citric acid as the standard of comparison. In most cases, the performance of the acids was determined in the presence and absence of added iron salts and antioxidants (BHA, BHT). Mathematical evaluation of the data shows malic acid to be significantly more effective than citric acid in most cases and equal to citric acid in some. In very few instances, a reversal of performance was observed depending on experimental conditions used. In a few experiments, malic acid was also tested against phosphoric acid and found to be equal to or better than the latter.

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STUDIES ON ANTIOXIDANT TREATMENTS OF CRUDE VEGETABLE OILS. E. R. SHERWIN and B. M. LUCKADOO, Eastman Chemical Products, Inc., Kingsport, Tennessee.

Treatments of crude safflower seed, soybean, sunflower seed and cottonseed oils with the antioxidant compounds butylated hydroxyanisole (BHA), propyl gallate (PG) and tertiary butylhydroquinone (TBHQ) have been investigaed. PG and TBHQ were effective in inhibiting oxidative degradation of the crude oils subjected to long-term storage as determined by measurement of peroxide formation in the oils during storage and by determination of AOM and oven (145 F) stabilities of the oils before and after storage. Of particular interest were the oxidative stability characteristics of these oils after they had been stored for relatively long periods in crude form (with and without the antioxidants) and then alkali refined, bleached and deodorized. The data from stability tests on these refined oils indicate that vegetable oils protected with potent antioxidants, such as PG or TBHQ, during storage in the crude form might yield refined oils with somewhat higher initial oxidative stability, and better response to further antioxidant treatment.

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EFFECTS OF HUMIDIFICATION ON ACTIVITY OF CATALYSTS AND ANTIOXIDANTS IN MODEL SYSTEMS. K. H. TJHIO, T. P. LABUZA and M. KAREL, Massachusetts Institute of Technology, Cambridge, Massachusetts.

Studies on freeze-dried model systems containing methyl linoleate and various additives have shown that water content plays an important role in controlling activity of antioxidants and catalysts. It was shown previously that water can counteract catalytic activity of certain metals, and that it enhances the antioxidant effect of some chelating agents. Different additives, however, show different response to the effect of humidification, especially at high water activities. Data are presented which show that complexes of manganese with histidine show enhanced prooxidant activity at high water contents, but those of cobalt become more antioxidant. Effects on other antioxidant systems are also discussed.

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EFFECTS OF NITROGEN BASES OF PHOSPHATIDYL CHOLINE AND PHOSPHATIDYL ETHANOLAMINE ON AUTOXIDATION OF METHYL LINOLENATE EMULSION. LLOYD M. SMITH and LEE-SHIN TSAI, Department of Food Science and Technology, University of California, Davis, California.

It has been suggested that the synergistic activity of phosphatidyl choline and phosphatidyl ethanolamine, when these compounds are added to menhaden oil, resides in the nitrogen-containing moiety. However, the role of the nitrogen-containing groups of these glycerophospholipids in the autoxidation of fatty materials in aqueous dispersions has not been elucidated. Methyl linoleate, emulsified in borate buffer with 0.002 M sodium lauryl sulfate, was used to study the pro- or antioxidant effect of 0-phosphocholine, choline, 0-phosphoethanolamine and ethanolamine. The amounts of oxygen uptake were calculated from rate data obtained at 37 C with an oxygen electrode. Compared to the corresponding controls with no added nitrogen base, 0-phosphoethanolamine and ethanolamine significantly increased the oxygen uptake by the methyl linoleate emulsion at pH 7.9, but significantly decreased the uptake at pH 10.2. Also, the accelerating effect of both compounds appeared only at the early stage of autoxidation. In contrast, 0-phosphocholine and choline had very little effect at either pH. The increase in pH from 7.9 to 10.2 had little effect on oxygen uptake of the control samples. Since the pK value of the amino group of ethanolamine in aqueous solution is about 9.4, the predominant chemical form of the amino group at pH 7.9 is $-\overset{+}{N}H_3$, and at pH 10.2 is $-\ddot{N}H_2$. The above results suggest that the $-\overset{+}{N}H_3$ group of 0-phosphoethanolamine and ethanolamine accelerates oxygen uptake by the methyl linoleate emulsion, whereas the $-\ddot{N}H_2$ group decelerates oxygen uptake. Thus the number of protons bonded to the nitrogen atom determines whether the amino group acts as a pro- or antioxidant. It is postulated that since the quaternary amine group of 0-phosphocholine and choline do not have any protons bonded to the nitrogen atom, they do not affect the autoxidation of the methyl linoleate emulsion.

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THE INHIBITION OF OXIDATION BY AROMATIC AMINES. K. U. INGOLD, K. ADAMIC and D. F. BOWMAN, National Research Council of Canada, Ottawa, Ontario.

Inhibition of hydrocarbon oxidation by aromatic amines will be described in terms of the kinetics, mechanisms and products of the reaction. The rate of hydrogen atom abstraction from the amine by peroxy radicals can be correlated with the polar properties of the substituents on the aromatic ring. The efficiency of formation of nitroxide radicals in the reaction between peroxy radicals and amino radicals has been determined for a number of amines and for a variety of peroxy radicals. For a particular amine, the efficiency of this reaction is greater with tertiary than with secondary peroxy radicals. Six products of the oxidation of the N-phenyl-2-naphthylamine have been identified. Some of our results may have application to the inhibition of oxidation by the aliphatic amines present in certain lipids.

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ANTIOXIDANT PROPERTIES OF α-TOCOPHEROL DERIVATIVES AND RELATIONSHIP OF ANTIOXIDANT ACTIVITY TO BIOLOGICAL ACTIVITY. W. A. SKINNER and R. M. PARKHURST, Stanford Research Institute, Menlo Park, California.

A series of 5-methyl substituted derivatives of α-tocopherol and the α-tocopherol model, 6-hydroxy-2,2,5,7,8-pentamethylchroman, were synthesized and evaluated for their antioxidant activities in preventing the air oxidation of β-carotene in corn lard. In addition, the compounds were evaluated for biological activity in preventing vitamin E-deficiency induced muscular dystrophy in the rabbit and dietary liver necrosis in the rat. The relationships between the antioxidant effects, chemical structure and biological activities will be discussed.

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FERRIC IRON-CATALYZED REACTIONS OF BIOLOGICAL ANTIOXIDANTS WITH PREFORMED LIPID HYDROPEROXIDES. E. H. GRUGER, JR. and A. L. TAPPEL, Bureau of Commercial Fisheries, Seattle, Washington.

Reactions of several biological antioxidants were measured in the presence of ferric iron-catalyzed dissociations of preformed lipid hydroperoxides. Spectrophotometric evidence supports a unimolecular dependence of antioxidant concentration. Rates of α-tocopherol oxidation were compared in reactions with the hydroperoxides prepared from methyl linoleate, methyl linole-

nate, ethyl arachidonate, methyl eicosapentaenoate, and a highly unsaturated fraction of ethyl esters of fatty acids obtained from menhaden oil. The degree of unsaturation of hydroperoxides of homogeneous chain length fatty acids had little effect on the rates of α-tocopherol oxidation. The hydroperoxides from the menhaden fatty acids gave faster rates of α-tocopherol oxidation than did the hydroperoxides of homogeneous chain length. The antioxidants and their relative rates of reactions with methyl linoleate hydroperoxides at 37 C were found to be as follows: ubiquinol-6, 4.52; α-tocopherol hydroquinone, 2.86; ubichromenol-6, 1.05; ubichromenol-10, 1.01; and -tocopherol, 1.00. In model systems composed of homogeneous media, the rates of oxidation of antioxidants vary inversely with respect to pH and water content of the media. A mechanism is proposed for α-tocopherol oxidation, which implicates acid catalysis and hydration steps in addition to a quinone methine intermediate.

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ANTIOXIDANT PROPERTIES OF TOCOPHERAMINES. H. S. Olcott and J. Van der Veen, Institute of Marine Resources, University of California, Berkeley, California.

Some of a group of amine derivatives of the tocopherols have recently been shown by others to have vitamin E activities in the rat and chick apparently not dependent upon their conversion to tocopherols. Several of these tocopheramines have not been assayed for antioxidant activity in vitro in squalene at 37 C. It was found that none of the tocopheramines were as effective as their parent compounds. The mono- and dimethylamines in general were less effective than the non-substituted tocopheramines. The data will be presented and discussed in the light of other observations with amines as antioxidants.

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ALIPHATIC NITROGEN DERIVATIVES: I. ADDITION OF N,N-DIBROMOSULFONAMIDES TO INTERNAL OLEFINS. T. A. Foglia, E. T. Haeberer and G. Maerker, Eastern Utilization Research and Development Division, ARS, USDA, Philadelphia, Pennsylvania.

The purpose of this investigation was the preparation of heterocyclic nitrogen compounds (aziridines) derived from readily accessible olefinic materials. Toward this goal the addition reaction of N,N-dibromobenzenesulfonamide (NNDBS) to a variety of internal olefinic compounds was investigated. Addition of NNDBS to the olefin in carbon tetrachloride solution was found to be an exothermic reaction. The initial adducts formed were β-bromo-N-bromosulfonamides which were not isolated but converted directly to their corresponding β-bromosulfonamides by reduction with aqueous sodium bisulfite. The reaction was investigated with pairs of *cis-trans*-olefins and proceeded in a stereoselective fashion. Major side products identified were the corresponding dibromides and bromohydrins of the olefin being studied. The yields of isolated β-bromosulfonamides were in the range of 35–80%. The purified β-bromosulfonamides were chemically cyclized by treatment with alkali to their corresponding N-sulfonylaziridines in almost quantitative yield. The stereochemistry of the aziridines has been shown to possess the same configuration as that of the starting olefins. Accordingly, a *cis*-olefin on reaction with NNDBS yields a *threo*-β-bromosulfonamide adduct which on cyclization yields a *cis*-aziridine.

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ALIPHATIC NITROGEN DERIVATIVES: II. REACTION OF *CIS*-9,10-EPIMINOOCTADECANE WITH CARBOXYLIC ACIDS. G. Maerker, E. T. Haeberer, E. T. Donahue and T. A. Foglia, Eastern Utilization Research and Development Division, ARS, USDA, Philadelphia, Pennsylvania.

The addition of carboxylic acids to *cis*-9,10-epiminooctadecane were studied further. The principal isolable products were 9-carboxamido-10-hydroxyoctadecanes (hydroxyamides) and 4,5-di-n-octyl-2-alkyl (or aryl)-Δ^2-oxazolines. Yields of isolated pure hydroxyamides varied in the 69–89% range while oxazolines were obtained in trace amounts to 30%. The ratio of the amounts of hydroxyamides to oxazolines could not be related to the ionization constant of the acid, but aromatic acids yielded more oxazoline than aliphatic acids under identical conditions. Yields of hydroxyamides and of oxazolines were both affected by the solvent used. The mechanism for hydroxyamide formation seems to involve protonation of the heterocyclic nitrogen atom and nucleophilic attack by carboxylate at one of the carbon atoms of the three-membered ring. The amino ester so formed promptly rearranges to the hydroxy amide by O → N acyl migration. Evidence will be presented to demonstrate that the oxazolines formed are true by-products rather than secondary products resulting from the dehydration of hydroxyamides.

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FRIEDEL CRAFTS REACTIONS OF *N*-ALKENOIC ACIDS. M. F. Ansell and G. F. Whitfield, Queen Mary College, University of London, London, E.1. England.

The reaction of pent-2-enoic acid with benzene and aluminium trichloride has been reported to give phenylpentanoic acid and a methyl tetralone and an ethyl- or dimethylindanone. Our work has revealed that 3-phenylpentanoic and 4-phenylpentanoic acids are formed, together with 4-methyltetral-1-one and 3-ethylindan-1 one. Pent-3- and pent-4-enoic acids both gave rise to both cyclic ketones, but in different proportions. The Friedel-Crafts reactions on non-2-enoic acid have been reported to give solely 8-phenylnonanoic acid. We have found that phenylation occurs at other positions in the aliphatic chain to yield a mixture of x-phenylnonanoic acids, with x = 3 — 8 inclusive. The neutral products comprise 3-n-hexylindan-1-one and 4-n-pentyltetral-1-one, together with a mixture of 1,y-diphenylnonan-1-ones (y = 8 —5 inclusive). Treatment of non-8-enoic acid with benzene and aluminium trichloride has also yielded a mixture of x-phenylnonanoic acids (x = 8,7,6,5, and possibly 4), together with neutral products. The identity of the Friedel-Crafts products has been established by comparison of the spectral characteristics with those of authentic samples and gas liquid chromatography peak enhancement experiments. The reaction of octadec-9-enoic (oleic) acid with benzene and AlCl$_3$ will be discussed. The application of mass spectrometry to the analysis of mixtures of methyl phenyl-n-alkanoates will also be reported.

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DIOL DIRICINOLEATES FROM DIHALOALKANES. C. K. Lyon and V. H. Garrett, Western Regional Research Laboratory, ARS, USDA, Albany, California.

The general esterification procedure of Mills et al. has been applied to the preparation of alkanediol diricinoleates. These diricinoleates were prepared by heating ricinoleic acid with triethylamine and dihaloalkanes such as: 1,2-dichloroethane, 1,4-dichlorobutane, 1,5-dibromopentane or 1,10-dibromodecane. As expected, the dibromides react more rapidly than the dichlorides. This method of esterification avoids the side reaction of estolide formation which occurs when ricinoleates are prepared by direct esterification or transesterification. The products are high molecular weight diols suitable for use in the preparation of polyurethanes.

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ACRYLATE ESTERS OF LONG CHAIN HYDROXY ACYL CHLORIDES. M. J. Diamond, Western Regional Research Laboratory, ARS, USDA, Albany, California.

Treatment of either 10-hydroxydecanoic acid or 12-hydroxystearic acid with acryloyl chloride yields the acrylate ester of the corresponding hydroxy acid. The ester acids have been converted to ester acyl chlorides by reaction with oxalyl chloride. Acrylate esters of low molecular weight homopolymers of long chain hydroxy acids previously have been copolymerized with vinyl chloride. The resultant resins are useful internally plasticized products. Potential uses for the more high reactive acryloyl acyl chlorides described herein will be discussed.

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PREPARATION OF SULFATE ESTERS BY CARBODIIMIDE-MEDIATED SULFATION. Ralph O. Mumma, F. J. Vastola and C. P. Hoiberg, Pesticide Research Laboratory, The Pennsylvania State University, University Park, Pennsylvania.

Dicyclohexylcarbodiimide, sulfuric acid and certain nucleophiles react in dimethylformamide to produce sulfated products. Alkyl sulfates, phenolic sulfates, and alkylthiol sulfates have been synthesized in good yields. Mass syntheses of sulfate esters of primary and secondary alcohols, phenols, steroids, polyhydroxyl molecules and various alkylthiols will be discussed. Reaction conditions have been found that will selectively sulfate various polyfunctional molecules. For example, alcohols can be sulfated under reaction conditions that do not sulfate phenols or alkylthiols. In some cases the carbodiimide-mediated sulfation produces unwanted side products. Techniques have been developed, for the purification of the sulfate esters, using ion-exchange columns. The sodium, potassium and cesium salts of these organic sulfate esters have been successfully analyzed by laser ionization mass spectrometry.

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STEREOSPECIFIC HYDRATION OF UNSATURATED FATTY ACIDS BY BACTERIA. L. L. Wallen and E. N. Davis, Northern Regional Research Laboratory, ARS, USDA, Peoria, Illinois.

Stereospecific addition of water by bacteria to a 9,10 double bond has been reported as a means for preparing 10-hydroxystearic acid from oleic acid in 71 mole % yield. Similarly, 10-hydroxypalmitic acid has been prepared in 53 mole % yield from palmitoleic acid. This work has been extended to the microbial hydration of three other substrate acids—ricinoleic, linoleic and linolenic. Products from the microbial hydration of *cis*-9,10 bonds of these substrates are new hydroxy fatty acids, previously unreported in the literature. The hydration was accomplished by first growing cells of a *Pseudomonas* sp. under aerobic conditions; the substrate fatty acid then was added and conversion occurred readily under anaerobic conditions. When ricinoleic acid was the substrate, we isolated 41 mole % of 10,12-dihydroxystearic acid having an unexpectedly low melting point of 66.0–66.5 C. Gas chromatographic retention time and spectral analyses confirmed its structure. Linoleic acid gave 10-hydroxy-*cis*-12-octadecenoic acid, whereas linolenic acid gave 10-hydroxy-*cis*,*cis*-12,15-octadecadienoic acid. The physical, spectral and chemical characteristics of these new fatty acids are described.

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ALUMINUM CHLORIDE-CATALYZED ACYLATION REACTIONS USING ISOPROPENYL ESTERS AS ACYLATING AGENTS. E. S. Rothman and G. G. Moore, Eastern Utilization Research and Development Division, ARS, USDA, Philadelphia, Pennsylvania.

The enol ester isopropenyl stearate forms stearophenone with benzene under mild Friedel-Crafts reaction conditions. The same enol stearate reacts similarly at the olefinic bond of 9-octadecene and of methyl oleate to form an α-β unsaturated ketone and an α-β unsaturated keto-ester respectively. Unlike the most complex product mixtures obtained in France by Perron and co-workers from the reaction of methyl oleate with aluminum chloride, under our mild conditions in the presence of the enol ester simple products are obtained. A by-product in these reactions is heneicosane-2,4-dione which becomes the sole product when olefin is omitted from the mixture. The β diketone easily formed a copper chelate derivative.

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THE CONTRIBUTIONS OF DR. LAURANCE W. KINSELL AND CO-WORKERS TO ATHEROSCLEROSIS RESEARCH. Hugh Sinclair, Magdalen College, Oxford University, England.

Abstract not available at press time.

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CHOLESTYRAMINE AND POLYUNSATURATED FAT: SIMILARITY OF EFFECTS IN MAN. Peter Wood, Institute of Metabolic Research, Highland General Hospital Oakland, California.

Investigations have been made of the effects of cholestyramine and polyunsaturated fat upon human cholesterol metabolism, other than their well-known hypocholesterolemic action. In metabolic ward studies both cholestyramine and polyunsaturated fats have been shown to increase fecal steroid excretion rate in normal subjects. Construction of specific activity-time curves for plasma free-cholesterol in subjects injected intravenously with 4-^{14}C-cholesterol suggested that both agents studied result in increased rate of addition to plasma of newly-synthesised cholesterol and increased synthesis rates of bile acids. An increase in the ratio of primary (cholic and chenodeoxycholic) bile acids to secondary (deoxycholic) bile acid is also observed, accompanied by an increase in the ratio of glycine to taurine conjugates in bile. Studies on daily output in bile of bile acids, free-cholesterol and phospholipids have been made (in the case of polyunsaturated fats only) in three subjects. It is suggested that output of all these components is increased on diets containing relatively polyunsaturated fat as compared with saturated fat diets.

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EFFECT OF HYDROGENATED FATS ON BLOOD CHOLES-

TEROL IN MAN. F. H. MATTSON, The Procter & Gamble Co., Cincinnati, Ohio.

The hydrogenation of an edible fat can result in (a) an increase in saturated fatty acids, (b) a decrease in polyunsaturated fatty acids, (c) an increase in both *cis* and *trans* monounsaturated acids, and (d) an increase in the isomers of polyunsaturated fatty acids. The dominant products are determined by the hydrogenation conditions selected. The conditions currently used result mainly in b and c, and some d. There is a considerable literature on the effect on blood cholesterol of feeding hydrogenated fats to man. Unfortunately in many of these reports the experimental fats are not adequately described. The fatty acid composition, either supplied by the authors or estimated by us where it was not given, has been correlated with the effect on the blood cholesterol level. The hypo- or hypercholesterolemic effects of the hydrogenated fats are not different from that of any other fat or oil. All of the observations can be explained on the basis of fatty acid composition and no unique property of hydrogenated fat need be invoked.

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DIETARY MANAGEMENT OF HYPERLIPEMIC STATES IN MAN. EDWIN L. BIERMAN, VA Hospital and University of Washington School of Medicine, Seattle, Washington.

Abnormal accumulation of triglyceride (TG) in the circulation (hyperlipemia) may be associated with a variety of disorders. Rational dietary management of these hyperlipemic states depends on an assessment of the specific abnormality and on an understanding of its pathogenesis. Endogenous lipemia is usually seen in overweight adults, commonly associated with mild glucose intolerance, and is characterized by an increase in concentration of TG-rich avery low density plasma lipoproteins (pre β or α_2 mobility on electrophoresis; $S_f > 20$ on ultracentrifugation). The degree of lipemia is directly related to dietary carbohydrate content (carbohydrate-induced lipemia) and is also extremely sensitive to total caloric balance. Hence caloric restriction is the treatment of choice, and reduction of body weight to ideal levels may be associated with complete amelioration of hyperlipemia. Exogenous lipemia, characterized by the presence of dietary fat particles (chylomicrons) in fasting plasma, usually reflects impaired assimilation of plasma triglyceride associated with low levels of post-heparin lipolytic activity, and may be primary or secondary to other metabolic disorders, e.g., diabetes. The degree of lipemia is directly related to dietary fat content (fat-induced lipemia). Hence dietary fat restriction minimizes chylomicronemia in this disorder and medium chain triglycerides (MCT) provide a helpful dietary supplement. Mixed hyperlipemic states are often associated with moderately severe diabetes and also can be managed by caloric restriction. A more rare form of hyperlipemia (Broad-Beta disease) is associated with the accumulation of a plasma VLDL (S_f 12–100) rich in both TG and cholesterol with an unusual β electrophoretic migration. Dietary management of this disorder, in combination with drugs, is particularly effective and involves both caloric and cholesterol restriction.

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GRAPHIC PRESENTATION AND ANALYSIS OF COMPUTER-DERIVED SCHLIEREN LIPOPROTEIN DATA. LIN C. JENSEN, THOMAS H. RICH and FRANK T. LINDGREN, Donner Laboratory, University of California, Berkeley, California.

A method is described for the graphical presentation of Computer-derived schlieren lipoprotein data. Lipoprotein spectra are fully corrected to standard conditions and are plotted by a cathode ray tube (CRT) or a Cal-Comp plotter. The plot-producing computer programs are flexible in providing many options to appropriately represent many types of spectra, including difference spectra. Plots can correspond to montaged tracing of schlieren photographs (useful for error detection) or to a single pattern on a continuous log scale. Special application includes analysis of very low-density lipoproteins and other lipoprotein spectra in chyle or in plasma very rich in chylomicrons.

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THE METABOLISM OF UNSATURATED FATTY ACIDS IN RATS FED DL-ETHIONINE. R. L. LYMAN, C. GIOTAS, M. A. FOSMIRE and P. MILJANICH, Department of Nutritional Sciences, University of California, Berkeley, California.

Ethionine, the ethyl analogue of methionine, has been known for many years to affect lipid metabolism in experimental animals. The most characteristic and widely studied lesions produced by ethionine in the rat is a fatty liver, characterized by infiltration of the organ with triglyceride. Recently we have observed that rats fed 0.25% DL-ethionine with a 9% casein diet had more and higher proportions of linoleic and oleic acids and less arachidonic and docosapentaenoic acids in their liver phospholipids than did pair-fed controls. When rats fed the same diets were given 1-^{14}C-linoleic acid, the animals fed the ethionine retained much more of the label in the linoleic acid and had much less labeling in the arachidonic acid than did the controls, indicating that ethionine interfered in some way with the conversion of linoleic acid to arachidonic acid. When rats were fed essential fatty acid deficient diets, with or without supplemental ethionine, the animals receiving the analogue had much less eicosatrienoic acid in their hepatic phospholipids than did controls, but the proportions of oleic acid were maintained or increased. Administration of 18-^{14}C-stearic acid indicated that the synthesis of oleic acid by desaturation of stearic acid was severely depressed. It appeared, therefore, that small quantities of ethionine fed to rats interfere with the conversion of linoleic acid to arachidonic acid as well as with the conversion of stearic acid to oleic acid, suggesting that desaturation reactions may be impaired by the analogue. The maintenance of high levels of oleic acid in the essential fatty acid deficient animals fed ethionine indicates that alternate pathways for its synthesis are operative or even enhanced.

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SOME RELATIONSHIPS OF POLYUNSATURATED FATTY ACID METABOLISM TO ATHEROSCLEROSIS. J. F. MEAD and D. F. HAGGERTY, JR., University of California, Los Angeles, California.

The relationships of the polyunsaturated fatty acids to atherosclerotic disease have been demonstrated in many studies involving both dietary and metabolic factors. However, the mechanism of their function and many aspects of their metabolism are still obscure. In attempting to solve some of these problems in this laboratory, cells in culture are grown in the presence or absence of polyunsaturated fatty acids and the metabolism of these acids is studied. It has been found that some types of cells can affect the conversion of linoleic acid to arachidonic whereas others seem to be incapable of carrying out the desaturation steps, although quite capable of elongation. In one case, a cell line was grown in the complete absence of polyunsaturated acids in the medium and desaturation was prevented with sterculic acid. Although these cells grew, they showed certain abnormal features that will require further elucidation. It is evident that one of the most important functions of these fatty acids is mainenance of the properties of the various cellular membranes.

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USE OF OPTICAL ROTATION IN DETERMINING CONFORMATION OF PROTEIN WITHIN MEMBRANES. D. W. URRY, American Medical Association, Chicago, Illinois.

Essential to the application of the spectroscopic method of optical rotation to the study of particulate or membranous systems is an understanding of the types of distortions which are inherent in such systems. Basic to an avowed understanding is a mathematical formalism capable of calculating the distortions from spectra on molecularly dispersed systems. Ultimately what is desired in the formalism is the ability to correct the distorted curve by using readily available experimental data. It has been demonstrated that two factors, absorption flattening and light scattering, when suitably formulated can reproduce a set of experimental curves on poly-L-glutamic acid in varying degrees of aggregation as measured by varying turbidity. The mathematical formalism and means of correcting the distorted curves will be presented. The results of the above considerations indicate that there is a higher degree of ordered protein in membranous systems than previously thought and that the protein exhibits a characteristic α-helical optical rotation pattern.

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PROTEIN CONFORMATION AND MEMBRANE STRUCTURE. S. J. SINGER, University of California, San Diego, California.

Abstract not available at press time.

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TEMPERATURE DEPENDENCE OF THE CONFORMATION OF HUMAN SERUM LOW AND HIGH DENSITY LIPOPROTEINS. A. SCANU, University of Chicago, Chicago, Illinois.

Circular dichroic (CD) spectra of human serum low and high density lipoproteins (LDL and HDL) were recorded as a function of temperature (0 → 80 C) under a variety of experimental conditions. Similar studies were carried out in the lipid-free products, apo LDL and apo HDL, dissolved in aqueous buffers. Both LDL and HDL exhibited reversible thermal transitions which were only modestly influenced by changes of the medium or chemical modification of either of the two lipoprotein species. In comparison, apo HDL and apo LDL exhibited a more marked temperature-dependent conformational instability and had spectra significantly affected by the procedure of solubilization employed. In the case of apo HDL, spectral differences were also noted among the non-identical subunits; re-lipidation with aqueous dispersion of HDL lipids produced restoration of the spectral properties of the original lipoprotein under given thermal conditions. The data appear to support the view that the conformation of apo HDL and apo LDL is to a large extent independent of lipids. The latter appear to have a stabilizing role in the conformation of these protein moieties, presumably through protein-phospholipid interactions.

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PHYSICAL STUDIES OF LIPID-POLYPEPTIDE AND LIPID-PROTEIN INTERACTIONS. D. CHAPMAN, Unilever Research Laboratory, The Frythe, Welwyn, Hearts, England.

Lipid-protein interactions may be of considerable importance in the organization and function of cell membranes. We shall describe some of our recent studies using various physical techniques, e.g., x-ray, calorimetric and spectroscopic techniques, which have been designed to provide information about the nature of these interactions. Model systems have been studied using lipids and pure polypeptides and the natural serum lipoproteins have also been examined. The information obtained from these studies has been compared and contrasted with studies of natural membranes.

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PREPARATIVE FRACTIONATION BY FRONTAL COUNTERCURRENT DISTRIBUTION. R. A. BARFORD, R. J. BERTSCH and H. L. ROTHBART, Eastern Utilization Research and Development Division, ARS, USDA, Philadelphia, Pennsylvania.

When solute was added to the first tube of a countercurrent distributor in a large number of increments, the output profile was no longer Gaussian but was a step function or frontal which could be described in terms of the integral of the Gaussian curve, the partition coefficients, the volumes of upper and lower phases, and the retention volume of the solutes. With these relationships, idealized output profiles of the solutes, as well as such information as the number of inputs required to produce a frontal, could be determined. This newer countercurrent technique was found to be useful for the fractionation of large amounts of material. For example, in a typical experiment, a trivernolin fraction of 64 g (ca. 95% purity) and a 10 g divernoyl-acyl triglyceride concentrate (86% purity) was obtained when 107 g of Vernonia oil was distributed. This was considerably more product that would be produced from CCD operated in the convention manner. In these frontal experiments conditions were not usually ideal and deviations from predicted profiles were observed. Data from phase equilibria studies on model systems explained these deviations.

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AN UNUSUAL NITROGEN-CONTAINING LIPID FROM *CORDIA VERBENACEAE* SEED OIL. K. L. MIKOLAJCZAK, D. S. SEIGLER, C. R. SMITH, JR. and I. A. WOLFF, Northern Utilization Research and Development Division, ARS, USDA, Peoria, Illinois.

Cordia verbenaceae (Boraginaceae) seed oil consists of normal triglycerides and 35% of an optically active nitrogenous fraction, which liberates HCN when treated with dilute base. This nitrogen-containing lipid is a mixture of esters. The esters are composed of two ordinary fatty acid moieties of varying chain lengths esterified with a dihydroxy nitrile. Experimental evidence (especially NMR) suggests that this nitrile is a five-carbon compound with a terminal methylene group. However, all attempts to isolate it in an unesterified form resulted in mixtures of unstable products. The detection of formaldehyde in the prod-

ucts from mild alkaline oxidation of the diesters supports the NMR evidence for a terminal methylene grouping. When a palladium catalyst is used, hydrogen uptake by the diesters is erratic and usually incomplete. Hydrogenation in the presence of a large excess of a platinum catalyst results in hydrogenolysis of the ester groups along with partial reduction of the double bonds and the nitrile group. Under mild conditions, partial hydrogenolysis to monoesters occurs, but longer reaction times yield a mixture containing an isopentyl amine and isovaleronitrile. Hydrolysis of the diesters with barium hydroxide gives a mixture of unstable products. Infrared and NMR spectra of this mixture are not inconsistent with hydroxy lactam structures. γ-Lactones with one fatty acyl group still attached are formed by treatment of the diesters with refluxing glacial acetic acid containing sulfuric acid as a catalyst.

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NEW SOURCES OF 9-D-HYDROXY-*CIS*-12-OCTADECENOIC ACID. R. G. POWELL, R. KLEIMAN and C. R. SMITH, JR., Northern Utilization Research and Development Division, ARS, USDA, Peoria, Illinois.

9-D-Hydroxy-*cis*-12-octadecenoic acid has been isolated from three new seed oils of the family Apocynaceae: *Holarrhena antidysenterica* (73%), *Nerium oleander* (11%), and *Nerium indicum* (8%). Previously, the known occurrence of this acid was limited to the genus *Strophanthus* (9–15%). Acetate-containing lipids are present in seed oils from *N. oleander* and *N. indicum* but are not evident in oils of *H. antidysenterica* or *Strophanthus hispidus*. An unusual tetra-acid glyceride was isolated from *N. oleander* oil by thin-layer chromatography. Pancreatic lipase hydrolysis of the glyceride shows that 9-D-acetoxy-*cis*-12-octadecenoic acid is esterified exclusively at an outer glycerol position and that normal fatty acids occupy the remaining two glycerol positions.

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OXYGENATED *TRANS*-3-OLEFINIC ACIDS OF *STENACHAENIUM* SEED OIL. R. KLEIMAN, G. F. SPENCER, L. W. TJARKS and F. R. EARLE, Northern Utilization Research and Development Division, ARS, USDA, Peoria, Illinois.

Stenachaenium macrocephalum (Compositae) seed oil contains among its minor components two classes of oxygenated fatty acids. One class has an epoxy group at the 9,10 position (6.5%); the other, a hydroxyl group adjacent to conjugated *cis-trans* double bonds (5.6%). The major epoxy acid (4.0%) is the previously unknown 9,10-epoxy-3,12-octadecadienoic acid. The conjugated dienols include two additional new acids with Δ3 unsaturation (2.5%): 9-hydroxy-*trans*-3,*trans*-10,*cis*-12-octadecatrienoic and 13-hydroxy-*trans*-3,*cis*-9,*trans*-11-octadecatrienoic acids. The other acids, except for the large amount (40%) of *trans*-3,*cis*-9,*cis*-12-octadecatrienoic, are those that commonly occur in seed oils. Functional groups were identified by infrared, ultraviolet and nuclear magnetic resonance spectroscopy, and by gas-liquid and thin-layer chromatography. Oxidative cleavage established the positions of unsaturation and oxygenation. The epoxy function was located by cleavage of milligram quantities of ester with periodic acid and identification of the oxidation products by GLC. Ozonolysis of the conjugated dienols in methanol produced fragments which served to establish the positions of the hydroxyl groups and the double bonds.

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CORRELATION OF SOLUBILITY DATA: III. THE ISOPLETH REFERENCE METHOD FOR PREDICTING SOLUBILITY DATA FOR LONG CHAIN HOMOLOGOUS AND ANALOGOUS COMPOUNDS. AUGUST V. BAILEY, JAMES A. HARRIS and EVALD L. SKAU, Southern Utilization Research and Development Division, New Orleans, Louisiana.

A new graphical method for correlating and predicting solubility data for homologous and analogous compounds is described. It complements the isotherm and isopleth methods which are applicable only to long chain homologous compounds. The method is based upon the linear relationship,

$$\frac{1}{T} = \frac{\Delta H_f'}{\Delta H_f}\left(\frac{1}{T'}\right) + C,$$

derived from the approximate freezing point depression equation in which T and T′ are the primary freezing points of two analogous compounds at the same molar concentration in a given solvent. The basic assumption that $\Delta H_f'/\Delta H_f$ remains essentially constant for homologous and analogous compounds in the same solvent over wide ranges of temperatures was validated by construction of isopleth reference plots using both published and new experimental solubility data for a number of long chain fatty acids and fatty acid derivatives in a variety of solvents. Complete solubility data are reported for behenic, stearic, palmitic, heptadecanoic, brassidic, erucic, petroselaidic and petroselinic acid in acetone, methanol, toluene and isopropyl ether. Complete solubility data are also included for elaidic acid in acetone, toluene and isopropyl ether and oleic acid in isopropyl ether.

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KINETIC RATE CONSTANTS DETERMINED BY A DIGITAL COMPUTER. R. O. BUTTERFIELD, Northern Regional Research Laboratory, ARS, USDA, Peoria, Illinois.

The difficulty of determining rate constants for complex reactions has been overcome with the development of a general digital computer program that can determine up to 10 rate constants in any reaction scheme having as many as 10 components. A given reaction to which the experimental data are to be fitted is described to the computer in a short one-step integration subroutine, which solves the differential equations representing the scheme. Only this sub-routine needs to be rewritten to change the reaction scheme. Special features of the program and length of calculations are discussed. A copper-chromite hydrogenation of a mixture of linolenate and conjugated linoleate demonstrates what the program does and what typical output is.

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SOME PARAMETERS OF CUPRIC SALTS OF FATTY ACIDS WITH REFERENCE TO ANALYTICAL USAGE. ROBERT R. LOWRY and IAN J. TINSLEY, Department of Agricultural Chemistry, Oregon State University, Corvallis, Oregon.

The procedure of Baker utilizing the cupric salts of free fatty acids has been examined in several areas in an attempt to broaden its applicability for fatty acid analysis. The level of sensitivity was increased through the use of an additional step using diethyldithiocarbamate as the detecting reagent. Analysis by atomic absorption was also used as another approach to increase the sensitivity. Some conditions for the formation of the copper salts were examined, namely the effect of pH and the substitution of other solvents. Solvents not only affected the color development, but also the solubility of the salts. Examination of the dependence of solubility upon chain length and unsaturation in a particular solvent was explored.

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ACOUSTIC CHARACTERISTICS OF SOME FATS AND OILS. G. O. HUSTAD, T. RICHARDSON, W. C. WINDER and M. P. DEAN, Department of Food Science and Industries, University of Wisconsin, Madison, Wisconsin.

Triggering frequency (cps), an indirect and highly accurate indicator of sound velocity, was measured at 65 C in a number of fatty acids, triglycerides, vegetable oils and animal fats. Each value was obtained after 3 to 5 min of temperature equilibration with a solution analyzer manufactured by Chesapeake Instrument Corp. Increased chain length and unsaturation of fatty acids resulted in increased triggering frequency values. Butter oil, being highly saturated and containing large percentages of short-chained fatty acids, exhibited a frequency that was approximately 265 to 365 cps less than those of corn, soybean, cottonseed, peanut, safflower or olive oils. Frequency values obtained with oils from margarines and shortenings were slightly less than those in the above vegetable oils, but at least 193 cps above butter oil. Coconut oil, on the other hand, was the only oil that gave a value lower than butter oil. Values from beef tallow and lard were approximately 190 cps above butter oil. A linear relationship existed between various ratios of individual vegetable oils or animal fats to butter oil and the corresponding frequency value at 65 C. This relationship may be of value in detecting adulteration of butter oil with other fats and oils.

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A SIMPLIFIED PREPARATIVE THIN LAYER CHROMATOGRAPHY OF PHOSPHOLIPIDS. JAMES M. IACONO and TERRY T. ISHIKAWA, Cincinnati General Hospital, Cincinnati, Ohio.

To isolate a sufficient quantity of pure phospholipids for fatty acid analyses, individual bands containing mixtures of phospholipids were rechromatographed following unidimensional TLC. A lipid extract was applied as a streak along the origin of a plate (Silica Gel HR), developed with chloroform-methanol-water (65:30:5 v/v/v). Phosphatidyl inositol (PI) and sphingomyelin (SPH) were mixed together as one band above the origin; phosphatidyl choline (PC) and phosphatidyl serine (PS) were mixed together to form another band above the band containing SPH and PI. The outlined areas were removed from the plate by scraping with a spatula into a beaker or test tube. A slurry of the silica gel was made for each scraped sample with chloroform-methanol (5:1 v/v) with the silica gel in a ratio of 1:1 (v/v). Each slurry was applied from a capillary pipette or syringe along the origin to a second plate so that a smooth, low mound formed, without sharp peaks on the surface. The solvent was evaporated from the mound by applying a gentle stream of nitrogen over the surface. When the bottom surface of the glass above the mound returned to ambient temperature, the plate was carefully placed in a tank containing the second solvent system (chloroform-methanol-acetic acid-water, 80:40:5:7, v/v/v/v). A separation was obtained of the mixture of PC and PS and of PI and SPH. Separations have been achieved with mixtures of known standards and extracts of lipids from platelets and erythrocytes.

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THE PHOSPHATIDES OF SAFFLOWER SEEDS AND THEIR CONTRIBUTION TO PIGMENT FORMATION OCCASIONALLY OCCURRING IN EXTRACTED OILS. H. J. BURKHARDT, Western Regional Research Laboratory, ARS, USDA, Albany, California.

The dark color occasionally found in extracted safflower oils is formed from colorless precursors. The three responsible precursors were isolated and identified as phosphatidyl ethanol amine, phosphatidyl choline and phosphatidyl inositol. These phosphatides were completely separated on microcrystalline DEAE-cellulose by gradient elution column chromatography. The fatty acids of each lipid were qualitatively and quantitatively determined as were the molar ratios of phosphorous to glycerol and inositol or ethanol amine. Paper chromatography and thin layer chromatography were also used for confirmation of structure of the intact and deacylated unknowns by comparison to reference phosphatides. The color forming potential of each of the three phosphatides and its relative abundance in safflower oils were determined. In most cases a direct relationship was found between total phosphatide content of a vegetable oil and color formed upon heating. A test was developed which allows determination of phosphatide content and composition as well as color formation of safflower seeds and oils. Several varieties of safflower seeds were compared.

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GAS CHROMATOGRAPHIC EQUIVALENT CHAIN LENGTHS OF ISOMERIC METHYL OCTADECENOATES AND OCTADECYNOATES. C. R. SCHOLFIELD and H. J. DUTTON, Northern Regional Research Laboratory, Peoria, Illinois.

Equivalent chain lengths (ECL) have been determined for the methyl *cis*-2-, 5-, 6-, 8-, 9-, 10-, 11-, 12-, 15-; *trans*-2-, 3-, 5-, 6-, 8-, 9-, 10-, 11-, 12-, 15-; and 17-octadecenoates and for methyl 6-, 8-, 9-, 10-, 11-, 12-, and 15-octadecynoates. These esters were mixed with appropriate saturated reference esters and run on the following capillary columns: diethylene glycerol succinate (DEGS) at 165 C, polyphenyl ether at 190 C and a 100% β-cyanoethylmethylsiloxane at 200 C. Although the levels of ECL values differed, the patterns were similar for the different substrates. For monoenoic esters, ECL generally increased with distance of double bond from the ester group but with little change from the 6 to the 9 position. ECL values for 2-isomers were larger probably because of conjugation of the double bond with the carbonyl group. Differences were smaller among isomeric *trans* esters than among *cis*. In methyl octadecynoates there was a greater and more regular increase for ECL as the triple bond moved away from the ester group.

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DETECTION OF UNUSUAL COMPONENTS BY DIRECT GAS LIQUID CHROMATOGRAPHY OF SEED OILS. R. KLEIMAN, J. W. HAGEMANN, F. R. EARLE and I. A. WOLFF, Northern Utilization Research and Development Division, ARS, USDA, Peoria, Illinois.

Direct gas liquid chromatography (GLC) of crude vegetable oils reveals much about their composition. For oils that contain only triglycerides of common acids and their homologs, the range

of peak retention times provides an excellent indication of fatty acid chain lengths. In some oils the quantity of a component present can be approximated; for example, the percentage of erucic acid in crucifer oils such as those from rapeseed and crambe. Components more volatile than the usual triglycerides are evident in many oils, occasionally in substantial amounts. Typical of these are acetotriglycerides, esters of triterpene alcohols, sterols, sterol esters, diol esters, free fatty acids, partial glycerides, wax esters, essential oils and hydrocarbons. Peaks midway between those of the usual triglycerides suggest the presence of acid constituents with an odd number of carbon atoms. Abnormally shaped peaks may result from triglycerides that contain conjugated unsaturation, whether it be preformed or created in the chromatograph. Correlation of GLC data on a seed oil with other analytical results aids in definitive characterization of components and is particularly useful in demonstrating whether constituents under study occur originally as glyceridic esters.

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QUANTITATIVE DETERMINATION OF MONO- AND DIGLYCERIDES BY GAS LIQUID CHROMATOGRAPHY. J. BLUM and W. R. KOEHLER, Lever Brothers Company, Edgewater, New Jersey.

Current methods for determining mono- and diglycerides in commercial products, such as emulsifiers and shortenings, usually make use of column chromatography on silicic acid followed by gravimetric determination or gas chromatographic analysis of the silyl derivatives obtained from each fraction. Several workers have described the gas chromatography separation of the silyl derivatives of mono- and diglycerides. This paper describes the application of this technique to the determination of mono- and diglycerides in mixtures that may contain variable amounts of other components. A known weight of sample is dissolved in a solution of cholesteryl acetate (used as internal standards) in pyridine; the solution is then silylated by the usual procedure using HMDS and TMS. The mixture is chromatographed on 1 foot columns packed with 3% OV-1 on Chromosorb W with temperature programming. This procedure is simple and rapid and has been applied to determine levels of mono- and diglycerides in commercially available emulsifiers as well as margarines, shortenings, intact oils, etc. Experimental data will be presented illustrating the precision and accuracy of the procedure.

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COMBINED ULTRAMICRO DRY COLUMN CHROMATOGRAPHY AND MASS SPECTROMETRY OF LIPID CLASSES. A. J. BAUMAN and HEINZ G. BOETTGER, California Institute of Technology, Pasadena, California.

Model mixtures of lipid classes, such as sterols, were chromatographed on dry microcolumns of porous glass 0.2 mm ID by 3 cm long. Peaks were visualized by iodine vapor and the column broken into segments, each of which held one peak. The segments were then characterized by means of mass spectrometry. The technique is very fast and it appears to be a useful adjunct for monitoring the purity of fractions developed in other types of chromatography.

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TECHNIQUES AND QUANTITATION OF PROGRAMMED TEMPERATURE GLC FOR DETECTING FATTY ACIDS PRESENT IN SUB-MICROGRAM AMOUNTS. JOHN L. IVERSON, Department of Health, Education & Welfare, Washington, D.C.

The value of varying the rate of temperature increase during programmed temperature gas chromatography (PTGC) of methyl esters is illustrated by its application to reference mixtures, fractions of cod liver oil and cotton seed oil and unfractionated butter oil. Column overloading and changing programming rates during an analysis have been found necessary to detect fatty acids present in parts per million. Since specific methods cannot be detailed for analysis of complex oils, guidelines are presented for obtaining optimum results. In isothermal analysis, with the emphasis shifting from precision to accuracy, the inherent error in measuring early eluting (spike) and late eluting (pancake) peaks becomes increasingly important. In isothermal analysis, the detection of late eluting components is surprisingly difficult. By the use of PTGC techniques, qualitative and quantitative data can be obtained on early eluting and late eluting fatty acids present in sub-microgram amounts in milk fats, animal fats and marine oils. By using modified PTGC techniques, short and long chain esters are eluted under optimum conditions for separation and quantitation. In using PTGC techniques with increasing heating rate for late eluting esters, there is little loss in resolution with decreased tailing actually improving the separations.

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ISOLATION AND DETERMINATION OF TRACE AMOUNTS OF FATTY AMINES AND RELATED COMPOUNDS. L. D. METCALFE, R. J. MARTIN and W. A. WAGNER, Armour Industrial Chemical Co., McCook, Illinois.

Long chain amines and quaternary ammonium compounds are used in coatings, packaging and agricultural chemicals. Because of these and other industrial uses, the fatty nitrogen derivatives are possible residues in food, tobacco and other materials. It was desirable for us to develop analytical methods for trace amounts of these chemicals. It was decided to use the great sensitivity of gas chromatography to help resolve the problem. Most analytical schemes tried depended on evaporating solutions of the amine salts to dryness or a small volume. It was found on analyzing residues treated in this way that the amines were lost entirely or recoveries were very low. Eventually the well known reaction of cationics with bromophenol blue was used in this work. The dye-cationic complex formed was partitioned between chloroform and water. The chloroform solution of the complex was separated and evaporated. The resulting residue was dissolved in a known volume of solvent. An aliquot of this amine-dye complex solution was injected into the GLC instrument. Using conditions of high sensitivity, almost quantitative recovery of the nitrogen bases in the nanogram range was achieved. This technique was used with various amines and quaternary ammonium compounds. A procedure for the determination of long chain amines on tobacco was developed in which this technique is a key step after an extraction and initial cleanup. The method appears to have wide application in determining trace amounts of nitrogen-bases in many systems. Furthermore, it may be applicable to important biological amines and quaternary ammonium compounds.

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DETERMINATIN OF AFLATOXINS IN PEANUT SOAPSTOCKS. A. F. CUCULLU, L. S. LEE, W. A. PONS, JR. and L. A. GOLDBLATT, Southern Utilization Research and Development Division, U.S.D.A., New Orleans, Louisiana.

A method is proposed for the estimation of aflatoxins in alkaline or acidulated peanut soapstocks. An aqueous suspension of the sample is adjusted to pH 6 with acetic acid for alkaline soapstocks, or neutralized to pH 7 and readjusted to pH 6 in the case of acidulated soapstocks. Lead acetate solution and acetone are added, and the mixture diluted with water to contain 30% acetone. After addition of filter aid, and filtration through paper, an aliquot of the filtrate is partitioned into chloroform, and purified by silica gel column cleanup. Aflatoxins in the purified extract are estimated on thin layer plates, either visually or densitometrically. Neutralization of soapstocks with acetic acid to pH 6 prior to extraction yielded higher recoveries of added aflatoxins than did neutralization to pH 3 with either hydrochloric or sulfuric acids. Recovery of aflatoxin B_1, but not of G_1, was essentially quantitative.

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RECOVERY OF AFLATOXINS FROM ARTIFICIALLY INOCULATED TOBACCO. C. Y. YANG and FRANKLIN F. FANNIN, University of Kentucky, Lexington, Kentucky.

An aflatoxin producing strain of *Aspergillus parasiticus* is one of the major components of tobacco foliar microflora. Morphologically, this strain resembles *Aspergillus flavus* strain NRRL 2999; however, its aflatoxin producing capacity is greater. By coupling solvent extraction and chromatographic fractionation procedures, a rapid and simplified separation method was developed for obtaining purified aflatoxins in large quantities. Using a sucrose-yeast extract solution as the culture medium, 1.89 mg of crude extract was obtained per gram of substrate. The crude extract consists of 43.7% aflatoxins in the ratio G_1-B_1-G_2-B_2 (100:24.6:17:14.1). The major aflatoxin component were characterized by thin layer chromatography. Ultraviolet absorption, fluorescence, infrared spectra and melting points and their properties were identical with known standards. Duckling bioassay of B_1, G_1, G_2 were positive. B_2 could not be tested. The unusually high ratio of G_1 to B_1 provides a source for further study of the physiological significance of G_1 on many biological systems. Experimental results indicate that aflatoxin components were partially recovered from *Aspergillus parasiticus* inoculated tobacco leaves under controlled environmental conditions.

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DECOMPOSITION OF AFLATOXINS IN AQUEOUS SOLUTIONS: EFFECTS OF pH AND HEAT. HARRY W. SCHROEDER and HUGO HEIN, JR., Market Quality Research Division, ARS, USDA, College Station, Texas.

Aflatoxin decomposition and degradation were studied in aqueous solutions buffered to pH's of 2.0, 4.0, 6.0, 7.0, 8.0, 10.0 and 12.0. Aflatoxins were most stable in mildly acidic to neutral solutions. At pH 2, B_1 and G_1 were degraded rapidly to fluorescent products tentatively identified as aflatoxin M_1 and G_{2a}, respectively (R_f's 0.18 and 0.12 on thin-layer chromatographic plates developed with chloroform plus acetone at a ratio of 85:15). Aflatoxins were degraded, primarily to non-fluorescent products, at an increasing rate as the solutions were made increasingly basic. A light-blue fluorescent material (R_f 0.35) accumulated to detectable levels in the basic solution; it was tentatively identified as aspertoxin. Also, in basic solutions, some reversal of loss of fluorescence by the aflatoxins was observed, following acidification of the solutions. In the more alkaline solutions or after the application of heat, fluorescence was regained to a lesser degree. Aflatoxin G_1 appeared to be essentially more reactive than B_1. All reactions proceeded more rapidly when the solutions were autoclaved 20 min at 15 lb/sq in. (121 C).

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OXIDATION OF 6-METHOXYDIFUROCOUMARONE. MABRY WILEY and ANTHONY C. WAISS, JR., Western Utilization Research and Development Division, ARS, USDA, Albany, California.

In attempts at a chemical synthesis of 3-hydroxy-6-methoxydifurocoumarone (aflatoxin M_1) from 6-methoxydifurocoumarone (aflatoxin B_1), several oxidation reactions were carried out. The products of these reactions and their implications will be discussed.

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STABILITY OF AFLATOXIN STANDARDS IN SOLUTION AND DRY FILM. J. A. ROBERTSON, W. A. PONS, JR. and L. A. GOLDBLATT, Southern Utilization Research and Development Division, ARS, USDA, New Orleans, Louisiana.

The stability of aflatoxins B_1, B_2, G_1 and G_2 was evaluated by ultraviolet absorptivity, solution fluorescence and solid state fluorescence on thin layer plates following storage in chloroform, in benzene, and as dry film for 6, 9 and 12 months at —18 C and 28 C. In general, at —18 C, the relative stability of the aflatoxins in benzene solution, in chloroform solution and in dry film was practically the same. In all cases the ultraviolet molar absorptivities of the aflatoxins at this temperature did not change significantly after storage for one year; however, the total solid state fluorescence decreased by 18% in chloroform solution, by 22% in benzene solution and by 26% in dry film storage. At 28 C the molar absorptivities, the solution fluorescence, of the aflatoxins all decreased significantly after 6–12 months of storage. There was a greater decrease in all three indexes for dry film storage than for storage in benzene or chloroform solution.

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A STUDY OF THE VARIABILITY ASSOCIATED WITH SAMPLING PEANUTS FOR AFLATOXIN. PETER J. TIEMSTRA, Derby Foods, Inc.

Peanuts known to contain aflatoxin were extensively sampled to study sources of variability. A nested design was used where sections (50 bag units), sub-samples and analytical variation were studied. Sample size was the most critical factor in characterizing this lot. The variability from section to section was not significant, indicating random distribution of the contaminant. The extent of damage in each sample was determined by visual examination and the fraction of the damage which was moldy or decayed was determined further. There was no correlation between these factors and the aflatoxin content. The variability of these particular quality factors were very close to theoretical values. One section was sampled in smaller units. The moldy and decayed fraction was analyzed separately as was the remainder of the damaged portion and the sound kernels.

The aflatoxin was definitely attributed to damage, particularly the moldy and decayed fractions.

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AFLATOXINS IN COTTONSEED HULLS. M. E. WHITTEN, Market Quality Research Division, ARS, USDA, Beltsville, Maryland.

Previous studies indicate that aflatoxins were found in laboratory-prepared hulls from cottonseed which contained aflatoxins. However, no aflatoxins were found in cottonseed hulls commercially processed from cottonseed containing aflatoxins, crops of 1964, 1965 and 1968, which were assayed in our laboratory. On the other hand, some lots of cottonseed hulls prepared from contaminated cottonseed in our laboratory, simulating commercial conditions as nearly as possible, were found to contain small but measurable quantities of aflatoxins. No aflatoxins were detected on other laboratory-prepared lots of hulls. Since no mechanical means to remove linters were available in the laboratory, the approximately 11% to 12% total linter content remained on the hulls or was only partially removed during dehulling. In normal oil-mill operation, the linter content of the seed is reduced 80% to 90% before dehulling. In the laboratory operation small quantities of fine meats containing aflatoxins may have been trapped in the linters on the laboratory-prepared hulls, resulting in a positive test for aflatoxins.

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SURVEY OF CORN FOR AFLATOXIN, ZEARALENONE AND OCHRATOXIN. ODETTE L. SHOTWELL, C. W. HESSELTINE and MARION L. GOULDEN, Northern Regional Research Laboratory, ARS, USDA, Peoria, Illinois.

Samples of corn from Grades 2–5 and from Sample Grade (SG) were analyzed for the presence of aflatoxins, zearalenone and ochratoxins. The 157 samples were extracted and assayed by a procedure that the Food and Drug Administration developed for detecting the three mycotoxins simultaneously. Extracts were assayed by thin-layer chromatography and results were confirmed by chemical derivatives. Ochratoxin A was found for the first time as a natural contaminant in one sample. Thin layer chromatography indicated that zearalenone could be present in one sample. Five samples contained aflatoxin B_1, the highest level being 26 ppb. All of the positive samples were in SG. Included in the 157 samples were 7 in Grade 2, 28 in Grade 3, 33 in Grade 4, 29 in Grade 5, and 60 in SG.

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CHEMICAL INACTIVATION OF AFLATOXINS IN OILSEED MEALS. G. E. MANN, L. P. CODIFER, JR., H. K. GARDNER, JR., F. G. DOLLEAR and S. P. KOLTUN, Southern Utilization Research and Development Division, New Orleans, Louisiana.

A variety of organic and inorganic reagents have been tested for their ability to destroy or inactivate the aflatoxins present in peanut and cottonseed meals containing these toxins. The reagents included acids, bases, oxidizing agents and various amines. Other reagents also were tested. The treatments were performed in a special laboratory-scale reactor, and were evaluated by determination of the aflatoxins in the products by TLC. In some instances, a large pilot-plant scale reactor was used. Ammonia, methylamine, sodium hydroxide and formaldehyde have proved effective in reducing aflatoxin levels and appear practical for large scale treatments. The effects of various reaction parameters including time, temperature and moisture content on the efficiency of these reagents are presented.

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ALTERNATIVES IN AFLATOXIN METHODOLOGY. M. S. MASRI and JON R. PAGE, Western Utilization Research and Development Division, ARS, USDA, Albany, California.

Alternative procedures in analytical work with aflatoxins will be described. One novel method for aflatoxin B_1 analysis which is based on extraction from agricultural products with mixtures of dimethylsulfoxide-water or dimethylformamide-water followed by partitioning with benzene to transfer the aflatoxin into benzene. This procedure yields exceptionally clean extracts, thus obviating in many instances the need for column cleanup steps prior to thin layer chromatography. Also described are several versatile variations of column cleanup steps with elution programs aimed at obtaining aflatoxin B_1 and M in separate fractions in which interfering substances with R_f values close to either of the aflatoxins are minimized.

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MOLECULAR STRUCTURE AD FUNCTION OF CELLULAR MEMBRANE. FRITIOF S. SJOSTRAND, University of California, Los Angeles, California.

When studying by means of electron microscopy the protein components of cellular membranes, methods must be applied which do not introduce too drastic changes of the native conformation of the protein molecules. After preservation of tissues with methods that take this aspect into account, the cellular membranes appear structurally different from their appearance after conventional preparatory procedures. This difference can be shown to be due to an extensive denaturation of the membrane proteins after conventional fixation and dehydration of the tissue. The new observations made on cellular structures after preparing the tissues according to a new technique have made it justifiable to propose a new model describing the molecular architecture of cellular membranes.

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SELECTIVE REMOVAL OF MYCOPLASMA MEMBRANE COMPONENTS. THOMAS M. TERRY, Albert Einstein College of Medicine, Bronx, New York.

Information on the structure of purified plasma membranes from *Mycoplasma laidlawaii B* has been sought by studying how this structure is affected morphologically and chemically by the selective removal of lipid and protein components. Membranes depleted of over 95% lipid by aqueous acetone extraction have a higher isopycnic density (1.25 g/cm^3) than native membranes (1.18 g/cm^3); trilamellar fine structure and vesicular character are preserved in these lipid-depleted membranes. Membranes can be depleted of up to 80% protein by prolonged incubation with proteolytic enzymes. Trilamellar fine structure becomes difficult to observe, but the protein-depleted membranes retain vesicular morphology. Membrane-bound hexosamine (galactosamine and glucosamine) remains associated with sedimentable vesicular material following either lipid or protein depletion, and can be substantially purified by combining these techniques. These data suggest a possible structural role for hexosamine in these membranes. The protein species of these membranes have been analyzed by detergent-polyacrylamide gel electrophoresis. The relative amounts of different proteins and of cysteine-containing proteins have been quantitated by gel analysis of appropriate radioisotope-labeled amino acids incorporated into membranes. Preliminary evidence for differences in the localization of different protein species within the membrane will be discussed.

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BIOLOGICAL EFFECTS OF CHANGES IN FATTY ACYL GROUPS IN MEMBRANE POLAR LIPIDS. M. E. TOURTELLOTTE and R. N. MCELHANEY, University of Connecticut, Storrs, Connecticut.

Unlike that of most cells, the fatty acid composition of membrane polar lipids of *Mycoplasma laidlawii* can be profoundly varied. For example, if grown in high levels of exogenous palmitic acid (16:0), the polar lipids of the cell membrane contain 90% saturated fatty acids. On the other hand, when grown in excess palmitoleic (16:1), the polar lipids now contain over 80% unsaturated fatty acids. In general, incorporation into polar lipids of a large number of unsaturated, branched chain, cyclopropanoid and saturated odd and even carbon numbered acids has little or no effect on growth rate or viability. These drastic changes in fatty acid composition are, however, accompanied by morphological changes; cells grown in unsaturated, branched chain and cyclopropane fatty acids grow as long filaments while those in long-chain saturated acids become coccoid and eventually swell and lyse. In the series of saturated acids containing 12 to 20 carbon atoms, no lysis occurs with 12 to 16 carbon acids; the 17 carbon acid shows some lysis while 18 carbon and longer acids show complete lysis at 37 C. As the growth temperature is lowered from 37 C to 25 C, the critical chain length resulting in lysis, as measured by optical density of cell suspensions and loss of viability decreases from 17 carbon at 37 C to 16 at 30 C and to 15 at 25 C. The interpretation of this data in terms of hydrophobic lipid-lipid interactions and liquid crystalline-crystalline transitions of smectic bilayers will be discussed.

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THERMAL PHASE TRANSITIONS IN BIOLOGICAL MEMBRANES. JOSEPH M. STEIM, Brown University, Providence, Rhode Island.

Isolated membranes and whole cells of *Mycoplasma laidlawii* and several bacteria undergo a reversible thermal phase transition which has been detected by differential scanning calorimetry and electron spin resonance. The transition temperature of the membranes can be changed by changing the fatty acids composition, and always is the same as the transition temperature of the extracted membrane lipids dispersed in water. Biological effects such as swelling and leakage are observed when the temperature of the cells is lowered below the transition, but the cells remain viable when they are again raised to the normal growth temperature. Neither the circular dichroism nor fluorescence emission spectra are affected by the transition, which appears to arise from melting of the fatty acid chains within the membrane bilayer.

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VOLATILES FROM HIGH TEMPERATURE OXIDATION OF *CIS*-7-TETRADECENE. R. J. HORVAT, Western Regional Research Laboratory, ARS, USDA, Albany, California.

Oxidations of fatty acids are characterized by formation of complex mixtures of products. To simplify the analysis of products, to minimize their number, and to help understand mechanisms of formation of these volatile products, a model compound, *cis*-7-tetradecene, was synthesized and then oxidized with air at 180 C. Oxidation of this olefin should be characteristic of oxidation under similar conditions of monoenoic fatty acids. The olefin was prepared by hydrogenation of 7-tetradecyne using Lindlar catalyst and characterized by IR, MS and ozonolysis. The volatile products (organic and aqueous phase) were removed from the effluent gases by means of a trap immersed in dry ice-acetone. GLC analysis of the organic phase with an open tubular column and a preparative column revealed the presence of four major peaks possessing retention times corresponding to pentanal, hexanal, heptanal and 2-nonenal, in addition to many other components. Some discussion of the origin of these aldehydes and other compounds will be presented.

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ROASTED PEANUT FLAVOR. GEORGE R. WALLER, BOBBY R. JOHNSON, PHILIP E. KOEHLER, GEORGE V. ODELL and MICHAEL E. MASON, Agricultural Experiment Station, Oklahoma State University, Stillwater, Oklahoma.

The current state of knowledge concerning the compounds which contribute to roasted peanut flavor will be discussed. The presence of the oil is required for full flavor development and all isolated compounds are oil soluble. Emphasis will be placed on pyrazines and carbonyls, which have been identified as the major volatile constituents of roasted peanuts. Early efforts resulted in the identification of a number of carbonyl compounds. They were isolated as their 2,4-dinitrophenyl-hydrazones and regenerated for analysis on a combination gas chromatograph-mass spectrometer (GC-MS). Seven monocarbonyl compounds were identified. Recent work in a number of laboratories indicates that alkylated pyrazines are important components of the flavor of several roasted food products. Five major pyrazine constituents of the volatiles from roasted peanuts have been reported from our laboratory. Recent studies on the identification of the numerous quantitatively minor pyrazines have been partially successful with the result that nine pyrazines were identified and a number were tentatively identified. Identification techniques used include gas-liquid chromatography retention times, infrared, ultraviolet, GC-MS, and accelerating voltage switching GC-MS. The accelerating voltage switching GC-MS technique was used to determine the components in unresolved or partially resolved mixtures. The mechanism for the formation of alkylated pyrazines has been investigated using ^{14}C-labeled sugars and amino acids. The results indicated that sugars were the principal source of the carbon atoms while amino acids mostly furnished nitrogen to the pyrazine molecule. Ammonium ions were not the common intermediate through which nitrogen entered the pyrazine ring.

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OXIDATION AND FLAVOR DETERIORATION OF OILS AND MARGARINES DURING SHALLOW PAN FRYING. ULLA HOLM and LILLEMOR FREDHOLM, Margarinbolaget AB, Technical Dept., Bromma, Sweden.

Oils and margarines with different content of linoleic and

linolenic acid were thermostatically heated in a frying-pan in a layer of 1,6 mm. The temperature of the fat-layer was measured and continously recorded. The influence of polyunsaturated fatty acid, frying temperature and frying time on the formation of oxidation products and flavor deterioration was studied. The oxidation was followed by peroxide-values and by measuring their decomposition products as aldehydes (benzidine test). The oxidation products formed in linoleic oils were proportional to their content of linoleic acid. Linolenic oils as soybean and rapeseed were oxidized to a higher extent under the same conditions. In the range of 155 C–185 C a temperature increase of 10 C approximately doubled the content of oxidation products. In prolonging the frying time from 5 to 10 min a 100% increase in oxidation products was noted. The development of oxidation products in margarines and butter compared with cooking oils was much slower in relation to the content of polyunsaturates. Oxidation values corresponding to 0,5–1% oxidized linoleic acid (absolute per cent) were found when frying vegetable oils containing 25–60% linoleic acid 10 min at 165 C. In margarines with same content of linoleic acid the oxidation values were five to seven times lower. The water phase had a marked inhibiting effect which was due to the skim milk. The flavors of the fried oils and margarines have been evaluated by a trained panel and a correlation between the limit of acceptance and the analytical value of aldehydes has been pointed out. The volatile oxidation products in the fried oils have been fractionated by means of gas chromatography and the amount of the most intense off flavors has been semi-quantitatively estimated.

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VOLATILE COMPOUNDS FROM THERMALLY OXIDIZED METHYL OLEATE. D. A. Withycombe, L. M. Libbey and R. C. Lindsay, Department of Food Science and Technology, Oregon State University, Corvallis, Oregon.

The thermal oxidation of methyl oleate was studied over a spectrum of heat treatments ranging from 50 to 150 C for periods of time up to 30 min. The degradation was quantitatively followed by gas-liquid chromatography (GLC) and liquid scintillation counting of the products of methyl oleate-U-^{14}C heated under a stream of compressed air. Hexane, octane, 2-decanone, benzene, *o*-xylene, methyl hexanoate, methyl heptanoate and methyl octanoate were identified by GLC and mass spectrometry. Mass spectral evidence also was obtained for the semi-aldehyde methyl esters of pimelic, suberic and azelaic acids. Most of the products formed suggested that autoxidation was responsible for the degradation occurring at the temperatures studies.

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THE ROLE ESSENTIAL OILS OF RANGE AND FORAGE PLANTS IN INFLUENCING ANIMAL ACCEPTANCE. George V. Odell, Charles J. Rudolph, Michael R. McGeehon, Winifred E. McMurphy and George R. Waller, Agricultural Experiment Station, Oklahoma State University, Stillwater, Oklahoma.

A possible role for the essential oil or steam distillable volatiles of range and forage plants in influencing animal acceptance or selection will be discussed. It is well established that animal (range) show preferences for certain forages and that they can detect differences between varieties within a species without tasting these plants. A specific problem of animal grazing preference of a single grass produced on different types of soil indicates the quantitative composition of this oil may be important. Further, the specific feeding preference of many organisms may be influenced by the qualitative composition of the essential oil fraction of grasses and forbs. Recent studies of the essential oil composition are based on the following: Samples of 24 major range and forage plants were collected at an active stage of growth before and during flowering. This collecting was made from several sites within 10 miles of Stillwater in late June and early July of 1967 and included *Andropogon gerardi, A. scoparius, Panicum virgatum, Sorghastrum nutans, Sorghum halepense, Tripsacum dactyloids, Bothriochloa ischaemum, B. intermedia* and *B. caucasica*. The yield of essential oil, obtained by steam distillation, was determined and gas chromatographic-mass spectral analyses used to characterize the major constituents of each oil. Animal (bovine) preference data for these and other range plants has been determined and will be related to the composition of the isolated essential oils. Recognition of the groups of compounds which may affect the palatability of a plant has been reviewed by many researchers and this present work suggests a means of plant selection by the animal.

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THE GAS LIQUID CHROMATOGRAPHY OF NITRO AND CHLORO SUBSTITUTED PHENYLHYDRAZONES OF *N*-ALKANALS AND *N*-METHYL KETONES AND SYN-ANTI-ISOMERS. R. C. Tripp, T. Richardson, C. H. Amundson and J. H. von Elbe, Department of Food Science and Industries, University of Wisconsin, Madison, Wisconsin.

Normal alkanals of carbon number 1–10 or *n*-methyl ketones of carbon number 3–9 were reacted with the following substituted phenylhydrazines: 2,4-dinitrophenylhydrazine (2,4-DNP), 2-nitrophenylhydrazine (2-NP), 4-nitrophenylhydrazine (4-NP) and 2,4,6-trichlorophenylhydrazine (TCP). Gas liquid chromatography (GLC) of the substituted phenylhydrazones was accomplished on a 6 ft × ⅛ in. column packed with 5% SE-30 coated on DMCS treated Chromosorb W. Column temperature was programmed from 100 C at rates of 6°/min to 15°/min depending on the derivative. A linear relationship between carbonyl carbon number and retention temperature of the derivatives was demonstrated. Methyl ketone derivatives exhibited slightly lower retention temperatures than the corresponding alkanal derivatives. Symmetrical carbonyl derivatives of 2-NP and 2,4-DNP (methanal, dimethyl ketone) chromatographed on a 6 ft × ⅛ in. column containing 1% OV-1 on Chromosorb W exhibited a single peak. Unsymmetrical carbonyl derivatives exhibited two peaks. The two peaks were concluded to represent syn-anti-isomers. Acetaldehyde 2,4-DNPH was recrystallized several times and shown to exist as the pure syn isomer by nuclear magnetic resonance spectroscopy. Upon chromatography, two peaks were observed. Isomerization was concluded to occur during GLC.

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REACTIONS OF FATTY ALDEHYDES WITH FATTY ALCOHOLS: FORMATION OF ACETALS, HEMIACETALS AND ALK-1-ENYL ALKYL ETHERS. V. Mahadevan, VA Hospital, Minneapolis, Minnesota.

Reaction of fatty aldehydes with alcohols in the presence of acid catalysts yields symmetrical acetals. The acetals are decomposed to a mixture of *cis* and *trans* alk-1-enyl alkyl ethers in the vapor phase by aluminum columns and aluminum containing solid supports during gas chromatography. The results of cleavage of alcohols from unsymmetrical acetals under the same conditions will be described. Hemiacetal formation between palmitaldehyde and various fatty alcohols was shown to occur as evidenced by infrared spectra obtained by the potassium bromide pressed-disk technique, but they were highly dissociated in solution. Although the oxidation of hemiacetals of fatty aldehydes and alcohols to the corresponding esters is known, their dehydration to the alk-1-enyl alkyl ethers has not hitherto been described. Results of dealcoholization of acetals and dehydration of hemiacetals to the alk-1-enyl alkyl ethers will be discussed as a possible biosynthetic pathway for the naturally occurring aldehydogenic lipids.

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REGULATIONS CONTROLLING SOLVENT EMISSIONS. M. Feldstein, Bay Area Air Pollution Control District; San Francisco, California.

Regulation 3 of the Bay Area Air Pollution Control District was designed to control the emission of reactive organic compounds, i.e., olefins, substituted aromatics and aldehydes, from stationary sources. These substances are generally most reactive in the formation of photochemical smog. All stationary sources except incineration are subject to the Regulation. However, this paper is concerned with its application to the solvent-using industries. The major control effort is directed towards the reformulation of materials, to limit the quantity of reactive solvents present. Three categories of complying solvents and surface coatings have been established: complying solvents which contain less than 8% reactive materials plus an allowable additional quantity of 12% monosubstituted aromatics; complying surface coatings which contain less than 8% reactive compounds plus an allowable additional quantity of 12% mono-substituted aromatics; and complying industrial surface coatings which contain less than 20% reactive compounds. The basic emission limitation in the Regulation is 50 ppm hydrocarbon or 300 ppm carbon. All sources meeting this requirement are exempt from control. Sources exceeding this limitation may still be in compliance if one of the following situations applies: less than 20 lb/day total organics are emitted; less than 10 lb/day reactive organics are emitted; the emission contains less than 5% reactives in the organic fraction of the emission; the overall reduction of reactives by an abatement device is 85% or more; and complying materials only are used, and no heat is applied. Several case histories are described concerning the application of the Regulation to specific emissions.

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PETROLEUM SOLVENTS CONFORMING TO AIR POLLUTION CONTROL REGULATIONS. H. E. Sipple, Shell Chemical Company., Petrochemicals Product Application, Downey, California.

Rule 66 and Regulation 3 are designed to control emission of photochemically reactive volatile organic substances into the atmosphere. A limited amount of non-exempt solvents may be used, but it is expedient to utilize exempt solvents wherever possible. Primary attention is focused on the aromatic content of the solvents. Exempt mineral spirits, extraction solvents and VM & P naphthas have been prepared and are commercially available. High solvency naphthenic base stock help offset the solvency of displaced aromatics. These exempt solvents appear to be entirely adequate for certain types of pain resins. Where greater solvency is needed in other paint resin systems, polar compounds such as alcohols, esters, ketones and nitroparaffins have been incorporated in the solvents. Thus, most solvent consuming industries can now be supplied with materials which qualify under Rule 66 and Regulation 3.

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EMISSION CONTROL BY COMBUSTION. L. C. Hardison, Robert B. Taylor and Otto M. Ikeda, UOP Air Correction Division, Darien, Connecticut.

Abstract not available at press time.

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WASTE TREATMENT. James R. McFarland, Swift & Company, Oak Brook, Illinois.

Abstract not available at press time.

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THE EFFECT OF INHIBITORS OF CHOLESTEROL SYNTHESIS ON MYELIN FORMATION. Marion E. Smith, Remo Fumagalli and Rodolfo Paoletti, VA Hospital, Palo Alto, California.

Three inhibitors of cholesterol synthesis, AY 9944, 20,25 azacholesterol, and Atromid were fed to suckling rats at the time of rapid myelination (15–21 days of age). At 22 days the rats were injected with 1-C^{14}-acetate and their brains and spinal cords removed on the 23rd day. Subcellular fractions and purified myelin were prepared from the brains and spinal cords, the lipids extracted, and the composition and specific activities of the lipids determined. The phospholipid-galactolipid-total sterol ratio was normal in the myelin from the treated animals, although a decreased yield of myelin was usually obtained. Myelin from animals treated with Atromid contained normal amounts of cholesterol although the specific activity of the cholesterol was somewhat decreased. The sterol in myelin from animals fed AY 9944 contained large amounts, up to 38% of the total sterol as 7-dehydrocholesterol, while desmosterol comprised up to 50% of the total sterol in myelin from 20,25 diazacholesterol-fed rats. The specific activity of the myelin sterol of the azacholesterol fed rats was only 60–70% of the control, therefore, some of the myelin cholesterol must have been derived from the mothers' milk. These experiments suggest that sterol is an absolute requirement for myelin synthesis, and when this is limiting, myelination is slowed. Certain sterols may substitute cholesterol when available, but the phospholipid-galactolipid-sterol ratio remains fixed.

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IN VITRO STUDIES ON CHOLESTEROL METABOLISM IN THE BLOOD FLUKE *SCHISTOSOMA MANSONI*. Thomas M. Smith and Thomas J. Brooks, Jr., University of Mississippi Medical Center, Jackson, Mississippi.

Three groups of blood flukes dissected from mesenteric venules of mice were axenically maintained in tissue culture medium NCTC 109 containing 0.05 μ of sodium 1-C^{14}-acetate ml for periods of four days. These trematodes were recovered from culture and tissue lipid extracted with chloroform-methanol (2:1 v/v). Thin layer chromatography (TLC) of the free sterol components in two hexane-diethyl ether-acetic acid solvent systems (90:10:1 v/v/v and 30:70:1 v/v/v) resulted in the iso-

lation of two 200 μg and one 300 μg sample of cholesterol. Separation of the dibromide derivatives of these cholesterol samples by TLC in benzene-ethanol (95:2 v/v) followed by counting in a liquid scintillation spectrometer revealed an absence of significant radioactivity in all samples. Under the above conditions *S. mansoni* adults do not synthesize cholesterol from labeled acetate.

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THE METABOLIC FATE OF CHOLESTEROL EPOXIDE IN THE RAT. JOSEPH A. FIORITI, MARYLIN N. GEORGE and REX J. SIMS, General Foods Corp., White Plains, New York.

The metabolic fate of cholesterol epoxide has been studied using Sprague-Dawley cesarean derived male rats. The epoxide was intubated as a 10% solution in monoolein. The animals were killed at 0, 1, 3 and 5 hr after intubation, and their livers and gastrointestinal tracts were extracted with 2:1 benzene-methanol; the extracts were water washed and analyzed by thin layer chromatography and, after silylation, by gas liquid chromatography. Quantitation of the TMS derivative of the sterols was best carried out using a 10% UC-W98 silicone column which separates the sterols of primary concern. The results show that the disappearance of cholesterol epoxide is accompanied by the appearance of a sterol which has been tentatively identified as cholestane trial. To shed further light on the metabolism of this epoxide, the thoracic duct was cannulated, and the collected lymph was analyzed. The results of these and other related experiments will be discussed.

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LIPASE, ESTERASE AND PHOSPHOLIPASE ACTIVITIES OF LYSOSOMES AND A SENSITIVE MEASUREMENT OF FATTY ACID RELEASE. CORA J. DILLARD, A. L. TAPPEL, K. HAYASE, S. MAHADEVAN and A. MELLORS, Food Science, University of California, Davis, California.

This paper reviews studies of the lipid-hydrolyzing enzymes of lysosomes. Subcellular distribution studies in rat liver have shown lysosomes to be the subcellular organelle which contains the highest specific activities of the lipid-hydrolyzing enzymes lipase, esterase and phospholipase. These organelles appear to have the complete complement of hydrolytic enzymes for lipid degradation, including sphingolipids and other glycolipids. Lipase and esterase are membrane bound; phospholipase being in both soluble and membrane fractions. Typical of other lysosomal hydrolases, optimal activities are found in the acid range of pH 4 to 5; optimal esterase and lipase activity are at a lower pH when Triton X-100 is used as substrate dispersing medium. Glyceryl-tri-, 1,2-di-, and 1-monodecanoate are hydrolyzed by lipase in decreasing order of activity. Of the *p*-nitrophenol fatty acid esters tested, containing between 8 to 18 C atoms, the esterase was least active on caprylate and more active on the higher fatty acid esters. Phospholipase cleaves both fatty acid ester linkages of lecithin and of phosphatidyl ethanolamine and releases free fatty acids from both positional isomers of lysolecithin. The distributions of *p*-nitrophenyl esterase and lipase are the same in hydroxylapatite column chromatography. Sulfhydryl reagents, iodoacetate and PCMB, inhibit 80–100% of their activities. These two activities may be attributed to a single enzyme. A sensitive method was used for estimation of fatty acids released. Copper soaps were prepared of the enzymatically released fatty acids which were previously extracted from the reaction mixtures by petroleum ether, and after resuspension of a dried aliquot in $CHCl_3$. Diphenyl carbohydrazide was used as color complexing agent; absorbance being measured at 540 mμ.

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STUDIES IN FATTY ACID SYNTHESIS BY CELL-FREE PREPARATIONS OF LACTATING MAMMARY GLAND OF THE MONGOLIAN GERBIL. JOHN G. CONIGLIO and RAYMOND B. BRIDGES, Vanderbilt University School of Medicine, Nashville, Tennessee.

Cell-free preparations of lactating mammary gland of the Mongolian gerbil incorporate significant amounts of ^{14}C from ^{14}C-acetyl CoA or ^{14}C-acetate into fatty acids having retention times equivalent to those of 18:2 and 20:2, respectively, as well as into fatty acids of shorter chain and with a higher degree of saturation. These radioactive compounds have been isolated in pure form by techniques of gas-liquid and $AgNO_3$ silica gel thin layer chromatography and the location of the radioactivity determined by ozonolysis and gas liquid radiochromatography of these products. The radioactivity was thus shown to be in $\Delta^{9,12}$ 18:2 and in $\Delta^{11,14}$ 20:2, respectively. Results of Dauben degradation of the latter compound indicated that it had been formed by elongation of $\Delta^{9,12}$ 18:2. Since these gerbils had been maintained on a diet composed of equal proportions of guinea pig chow, oats and sunflower seed (which made it rich in linoleic acid), other cell-free preparations were made using mammary gland from gerbils maintained on a diet composed of equal proportions of oats and guinea pig chow, or a purified diet containing 5% corn oil. With either ^{14}C acetate or ^{14}C acetyl CoA as substrate there was greater incorporation of ^{14}C into both 18:2 and 20:2 in preparations made from gerbils on the sunflower seed diet compared to those on either of the other two diets. This effect may be due to the greater dietary intake of linoleic acid by the gerbils on the diet containing sunflower seed.

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FATTY ACID METABOLISM OF COHO SALMON UNDER PENTACHLOROPHENOL AND SUBMAINTENANCE DIET. DAVID F. HANES, HUGO M. KRUEGER and ROBERT R. LOWRY, *Department of Agricultural* Chemistry, Oregon State University, Corvallis, Oregon.

Under specified conditions, the fraction of any specific fatty acid (FA) destroyed is greater than or less than the fraction of the total mass of all FA destroyed. Fatty acids can be characterized by the algebraic difference in their rates of destruction from the rate of destruction of the total FA mass. One set of poisoned (0.1 mg/1 PCP) and control salmon was fed daily a ration of tubifex worms twice with maintenance level and another set was fed one half the maintenance level. The fractions of the total FA lost (mg lost/mg available) over a 14 day period were 0.25 and 0.47 (PCP) on the high dietary intake and 0.34 and 0.50 (PCP) on the low intake. (Mass of FA available is the mass in salmon on day 0 plus the total FA mass contributed by the diet.) Of the 13 major FA present, seven FA (14:0, 18:0, 18:2, 20:2, 20:5, 22:5 and 22:6) in all four groups lost similar fractional amounts above or below the total FA fraction lost in their respective groups; the metabolism of these acids did not change relative to total FA metabolism thus these acids were similarly characterized quantitatively under four different conditions. Four acids (16:0, 16:1, 18:1 and 18:4) lost higher fractional amounts, relative to the total FA fractions lost, in the two groups on low dietary intake; the submaintenance diet induced a selectively increased rate of catabolism of these acids. Two acids (20:1 and 20:4) had lower relative fractional amounts only in the low intake control, but not in the poisoned group; the low intake required a selective retention of these acids, a retention which PCP poisoning abolished. Generally, many fatty acids are randomly destroyed in proportions to their concentrations. On a meager diet (one half maintenance), the relative rates of catabolism of some individual FA may increase or decrease and PCP poisoning can abolish selective decreases.

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ISOLATION OF 22:5ω6 AND 22:5ω3 ACIDS AND THEIR CONVERSION TO PROSTAGLANDINS VIA SHEEP VESICULAR GLANDS. L. J. NUTTER, O. S. PRIVETT and W. O. LUNDBERG, The Hormel Institute, Austin, Minnesota.

Docosapentaenoic acids were isolated in highly purified form from tuna oil and testicular lipids of rats fed a corn oil diet via a combination of liquid-liquid partition and argentation chromatography. Reductive ozonolysis of the fish oil 22:5 showed that the double bonds resided in the 7,10,13,16,19 positions; the acid isolated from rat testicular lipid had the double bonds in the 4,7,10,13,16 positions. In contrast to arachidonic acid and 5,8,11,14,17–20:5 which gave single homogeneous prostaglandins PGE_2 and PGE_3, respectively, when incubated with sheep vesicular glands, both 22:5 acids gave a mixture of prostaglandins. These prostaglandins were isolated by argentation chromatography. Studies on their formation, isolation and structure are described.

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AFLATOXINS IN FARMERS' STOCK PEANUTS: PEANUT QUALITY, MYCOFLORA AND CLIMATOLOGICAL CONDITIONS AS INFLUENCING FACTORS. BEN DOUPNIK, JR., Georgia Coastal Plain Experiment Station, University of Georgia, Tifton, Georgia.

Samples of farmers' stock peanuts were collected from six locations at weekly intervals for five weeks during the 1967 and 1968 harvesting seasons. Seventeen of 228 samples collected in 1967 and 23 of 356 samples collected in 1968 contained aflatoxins. In 1967, 2.5% of Segregation I (Seg I), 12.1% of Seg II, and 25.7% of Seg III samples contained aflatoxins and averaged 22, 264, and 324 ppb total aflatoxins respectively. In 1968, 3.2% of Seg I, 6.3% of Seg II, and 11.9% of Seg III samples contained aflatoxins and averaged 28, 61, and 583 ppb total aflatoxins respectively. Thus, in both years, significant relationships were found between the presence and concentration of aflatoxins and grading factors. Weekly incidences of aflatoxin contaminated samples were related to climatological conditions during the 14-day period to the collection dates in both years. Mycofloral studies of 73 selected samples in 1967 and of all samples in 1968 showed a relationship between the mean number of fungi/sample, grading factors and aflatoxin contamination. These findings support pervious observations that factors which influence peanut quality also influence aflatoxin contamination in farmers' stock peanuts. A model showed the possible relationships of these factors will be presented.

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AFLATOXINS IN COTTONSEED: DIFFERENTIAL ELABORATION BY *ASPERGILLUS FLAVUS* ISOLATES. L. J. ASHWORTH, JR., J. L. MCMEANS, and C. M. BROWN, U.S. Cotton Research Station, ARS, USDA, Shafter, California.

A total of 193 isolates of *Aspergillus flavus* collected throughout the cotton producing areas of California, were tested for pathogenicity to cotton and for in vivo aflatoxin producing potential. These isolates were expected to be pathogenic because they were isolated from acid delinted, surface disinfested cotton seeds. Each isolate was injected, as a water suspension of conidia, into each of five near-mature bolls. Observations were made on the fuzzy and decorticated seeds harvested from these bolls. All isolates were pathogenic, as characterized by the greenish yellow fluorescence of fibers in ultra violet light, and 98.4% were toxigenic (toxin concentration in seeds varied from trace to 260 ppm, with a mean of 29.4 ppm). Aflatoxins B_1 and B_2 were observed, which agrees with results of approximately 2,000 analyses made of naturally infected seed in this laboratory. Both aflatoxins B_1 and B_2 were detected when the total toxin content of seeds exceeded 40 ppb (88.6% of the cases). Absence of aflatoxin B_2 in 11.4% of the cases was not surprising, because it ordinarily occurs in lower concentrations than aflatoxin B_1. In these experiments, the mean ratio of aflatoxin B_1 to B_2 was 12.2:1. These results, together with results of earlier reported observations, suggest that the possibility of aflatoxins G_1 and G_2 occurring in cotton seed is nil. This is because strains of *A. flavus*, with a potential for producing aflatoxins G_1 and G_2, do not appear to occur in naturally infected cotton seeds. In addition, these results diminish the importance of the non-aflatoxin constituents of cotton seeds, which can obscure aflatoxins G_1 and G_2 in thin layer chromatograms prepared according to currently recommended analytical procedures.

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MYCOTOXIN PROBLEMS IN TOBACCO. A. I. SCHEPARTZ, D. G. BAILEY and J. H. CISLE, Eastern Utilization Research and Development Division, ARS, USDA, Philadelphia, Pennsylvania.

Although the importance of microflora on tabacco leaves has been studied for many years from the standpoint of function in the curing, fermentation and aging of tobacco as well as in crop diseases, it has only been in the past few years that attention has been drawn to the possible toxic effects of mold metabolites on the consumer of the product, the smoker. Recent findings by mycologists have shown that certain types of tobacco can, even under ordinary conditions, contain molds that are capable of producing highly toxic compounds. For example, *A. flavus* and *A. ochraceus*, both producers of known mycotoxins, have been isolated from several types of cigarette tobaccos. Thus aflatoxins and ochratoxins (as well as other mycotoxins) must be considered as possible contaminants in cigarettes. What might happen to these complex molecules in a burning cigarette? Would they be distilled over into the smoke or could they give rise to pyrolysis products that are in turn toxic? The results of recent research along these lines will be discussed.

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SIMPLE METHOD TO DETERMINE THE AFLOTOXIN-

PRODUCING POTENTIAL OF FUNGI. JERRY W. KIRKSEY, CHARLES E. HOLADAY and PHILLIP G. VINCENT, Agricultural Research Service, USDA, Albany, Georgia.

An improved method to test molds for aflatoxin-production potential appears to have advantages over other techniques described in the literature. The liquid broth medium contains soytone, dextrose, yeast extract and malt extract. Advantages of this medium include uniform composition and soluble nutrients. A liquid medium also facilitates extraction of metabolites produced by the mold. A 50 ml flask containing the broth is inoculated with a test mold and incubated for five days at 27 C on a shaking incubator. Chloroform is then added to the test vessel and shaken; the chloroform layer is spotted directly on a thin layer chromatographic plate together with an aflatoxin standard. The plate is developed in a chloroform-acetone mixture (7:1) and aflatoxins B_1, B_2, G_1 and G_2 can be identified and quantified, to some degree, by comparison with the standard. Thus far all our laboratory-tested mold which have the ability to produce aflatoxin on other substrates have also produced it on our assay broth.

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THE EFFECTS OF AFLATOXINS ON GERMINATING SEEDS OF FIELD CRESS. F. R. ROEGNER, Food and Drug Administration, Washington, D.C.

The aflatoxin inhibition of seed germination and pigment production in field cress (*Lepidium sativum*) was reported by Schoental and White in 1965. The absence of similar effects when they tested many other mold products suggested a high degree of specificity for aflatoxin B_1 and a possible bioassay procedure. Tests were performed in this laboratory by placing 50 seeds on filter paper in a Petri dish and then adding 5 ml of an aqueous solution of pure aflatoxin B_1 or G_1 at different levels. After four days in an illuminated germinator the seeds were examined to determine the degree of germination and pigmentation of the seedlings. Differences in pigment production were detected visually and quantitative determinations were made by spectrophotometric examination of acetone extracts of seedlings at 435 mm. The use of a thermal gradient plate permitted testing over a 15 to 35 C temperature range at different light intensities. Low and high temperature extremes reduced germination and pigment production in control and treated groups to about the same extent. With chlorotic effects light intensity was more critical; effects were more pronounced at a low light intensity (about 25 fc). Higher light intensities reduced the chlorotic effects. The effect with aflatoxin G_1 was more pronounced than with B_1 at a level of 1 μg/ml. The inhibitory effects were not permanent; many treated seeds germinated later and the seedlings tended to regain normal pigmentation, especially by the time the first true leaves appeared. These studies may be useful in devising an assay method because a 30% to 40% inhibition of pigment production occurs at aflatoxin levels which permit a high percentage of seed germination.

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A NEW METABOLITE FOR *ASPERGILLUS PARASITICUS*. R. D. STUBBLEFIELD, ODETTE L. SHOTWELL AND GAIL M. SHANNON, Northern Regional Research Laboratory, ARS, USDA, Peoria, Illinois.

A new metabolite related structurally to aflatoxins has been isolated and purified by chromatography on three columns (silicic acid, silica gel and neutral alumina) and recrystallized from chloroform-hexane. The compound is elaborated by *Aspergillus parasiticus* NRRL 2999 on rice or by *A. parasiticus* NRRL 3145 on wheat. Both these fungi are aflatoxin producers. The metabolite can be differentiated from aflatoxins B_1, B_2, G_1, G_2 and M and aspertoxin by thin layer chromatography and its ultraviolet absorption spectrum. Mobility of the new compound on thin layer plates lies between that of aflatoxin G_2 and M. Infrared and nuclear magnetic resonance indicate the same difurocourmarin ring system that is common to the aflatoxins.

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METABOLIC CONVERSION OF AFLATOXIN B_1 to M_1 IN VITRO. M. S. MASRI, J. R. PAGE and V. C. GARCIA, Western Utilization Research & Development Division, ARS, USDA, Albany, California.

Incubation of the 30,000 $\times$ *g* supernatant fraction of rat liver with crystalline aflatoxin B_1 resulted in the production of aflatoxin M_1. The extent of conversion ranged in different experiments from 1–5% of the aflatoxin B_1 added as substrate. In a preparative experiment the 30,000 $\times$ *g* supernatant fraction representing livers from five adult rats and prepared in 0.1 M phosphate buffer pH 7.4 was incubated with 75 mg crystalline aflatoxin B_1 in the presence of TPNH-regenerating system; about 850 μg of aflatoxin M_1 was isolated from the reaction mixture in crystalline form. The identification of aflatoxin M_1 was based on thin layer chromatography, ultraviolet spectrum, acetyl derivative formation and crystallographic measurements. In these experiments another prominent metabolite was encountered and has been isolated and partially characterized.

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REVIEW OF THE BIOLOGICAL EFFECTS OF AFLATOXINS ON SWINE, CATTLE AND POULTRY. A. N. BOOTH, Western Regional Research Laboratory, ARS, USDA, Albany, California.

The effects of feeding graded levels of aflatoxin to swine, cattle and poultry under simulated practical conditions were investigated. The parameters used to evaluate the effects were growth, mortality, feed intake, hematology, blood and liver biochemical studies, organ weights, gross and microscopic pathology and the transmission of aflatoxin into meat, milk and eggs. Swine were fed rations containing 0 to 800 ppb aflatoxin from weanling to 200 pounds body weight. No effects were observed at a level of 233 ppb aflatoxin in the ration and there was no evidence of aflatoxin residues in the meat, blood or liver. Beef steers were fed rations containing 0 to 1,000 ppb aflatoxin beginning at six to eight months of age and continuing for five months. The no effect level of aflatoxin was approximately 300 ppb. At slaughter no evidence of aflatoxin residues in the tissues was found but at the 1,000 ppb level the blood contained traces of aflatoxin. There was no evidence, however, of aflatoxin residues in the blood of cattle from which the feed was withheld for 24 hr prior to slaughter. Aflatoxin M was found in the milk of dairy cows fed aflatoxin. A rapid disappearance of aflatoxin M from the milk was observed following withdrawal of the aflatoxin. Broiler chicks were fed rations containing graded levels of aflatoxin from day-old to eight weeks of age. The findings with respect to aflatoxin intake will be discussed.

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MYCOTOXICITY OF *ASPERGILLUS OCHRACEUS* TO CHICKS. BEN DOUPNIK, JR. and JOHN C. PECKHAM, University of Georgia Coastal Plain Experiment Station, Tifton, Georgia.

Five strains of *Aspergillus ochraceus* Wilh., isolated from peanuts, were grown separately on sterile, moist corn for 14 days and fed to 1 day old Babcock B-300 cockerels to evaluate their toxic effects. Two strains were highly toxic, causing deaths of all birds during the first week of the experiment. Two strains were moderately toxic, causing severe growth suppression with some deaths occurring throughout the 3 week test period. One strain had no apparent effect. When the two most toxic strains (diets) were diluted, survival time increased but severe growth suppression was evident. Postmortem examinations revealed a few small hemorrhages in the proventriculi of birds which died between the second and fifth days. Emaciation, dehydration and dry, firm gizzard linings were observed throughout the experiment. Extensive hepatic injury consisting of either fatty changes or necrotic foci was the principal microscopic finding. Suppression of bone marrow activity and depletion of lymphoid elements in the spleen and Bursa of Fabricius were also found. The severity of the histopathologic changes was directly related to the concentration of ochratoxin A in the diet.

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MEMBRANE STRUCTURE EXPOSED IN HYDROPHOBIC FRACTURES. DANIEL BRANTON, University of California, Berkeley, California.

Regions of weak, entropic bonding appear to provide natural fracture planes in frozen biological membranes just as they do in lipid model systems. New evidence for this type of fracture is provided by freeze fracturing and deep etching experiments in which both membrane surfaces as well as inner, hydrophobic membrane faces have been studied. Whereas the former are usually smooth, small particles are associated with the latter. Their number and distribution is a unique property of a given membrane type. The distinction between these particles and others produced during ice sublimation can be demonstrated by comparison of structure in frozen membranes and oriented single ice crystals. The small, ca. 80A particles within the plane of the membrane are susceptible to proteolytic attack and may represent centers of membrane associated function.

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STEROID SPIN-LABEL OF MEMBRANE. WAYNE L. HUBBELL and HARDEN M. MCCONNELL, Stanford University, Stanford, California.

A steroid spin label has been found to undergo rapid, anisotropic Brownian motion about its long molecular axis when incorporated in membranes or phospholipid suspensions. The spin label was prepared by oxidation of the oxazolidine obtained by reaction of 2-amino-2-methyl-1-propanol and 5α-androstan-3-one-17-ol. When bound to membranes, the nitroxide group is protected against reduction by water soluble reducing agents, indicating that the nitroxide end of the molecule is buried in the fluid hydrophobic region of the membrane, whereas the 17-hydroxyl group is surely in a polar region. Other amphiphilic spin labels having the nitroxide group at the polar end of the molecule are readily reduced by water soluble reducing agents. The very rapid motion of the relatively large and rigid steroid spin label is considered to lend some plausibility to models of membrane transport involving motions of carrier molecules within the membrane structure.

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GENETIC CONTROL INVOLVED IN THE BIOGENESIS AND FUNCTION OF MITOCHONDRIA. DOW O. WOODWARD, Stanford University, Stanford, California.

Mitochondria exhibit many functions that appear to be independent of the rest of the cell; however, they depend on nuclear and cytoplasmic functions for many vital processes. Nuclear genes and cytoplasmic ribosomes are responsible for the production of many if not all mitochondrial enzymes. Cytochrome c has been shown to be specified by a nuclear gene, while there is some controversy concerning the genetic control over the rest of the cytochrome system. Linnane interprets his data to indicate that the cytochromes as well as mitochondrial ribosomes are products of mitochondrial genes. Otherwise, the only suggested function of mitochondrial DNA is the specifying of insoluble membrane protein. Some evidence suggests that the insoluble membrane protein is genetically homogenous, i.e., that it consists of a single genetic species of protein. At the same time, other data suggest that structural heterogeneity exists in the insoluble membrane protein fraction called structural protein. If more than one structural form of membrane protein exists, these forms must be only slightly different from one another such as could be explained by after synthesis enzymatic modification of the structural protein. Differential sensitivities to inhibitors have been reported between cytoplasmic and mitochondrial protein synthesizing systems. Cycloheximide has been reported to preferentially inhibit cytoplasmic synthesis while chloramphenicol at low concentrations preferentially inhibits mitochondrial protein synthesis. By allowing only one of the two systems to function, it is possible to follow the incorporation of labeled amino acids into specific protein fractions in vivo. These data must not be hastily interpreted, however, since Attardi has found that mRNA produced in mitochondria, presumably on a mitochondrial DNA template, is transferred to the cytoplasmic ribosomes. Also, some mRNA of nuclear origin may be transported to mitochondria to direct protein synthesis there although no evidence for this has appeared as yet. However, caution must be used in identifying gene location based on the site of synthesis of the corresponding protein. The approach of using maternally inherited mutants is a more direct way of determining which genes are of cytoplasmic origin. It is not always possible to easily determine the primary function of mutant cytoplasmic genes. The only cases in which this has been attempted in Neurospora and yeast suggests that membrane protein or ribosomes or both are either modified or missing in the cytoplasmic mutants.

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STRUCTURAL PROTEIN AND MEMBRANE ORGANIZATION. SIDNEY FLEISCHER and W. L. ZAHLER, Department of Molecular Biology, Vanderbilt University, Nashville, Tennessee.

Structural protein (SP) was originally isolated from mitochondria using detergents. The product shows no enzymic activities, is insoluble at neutral pH, and forms complexes with enzymes and phospholipids. These properties, and the presence

of SP in many membranes has led to the generally accepted hypothesis that SP forms the basic framework of membranes into which the functional components are inserted. Recent work has shown that SP prepared with detergents consists of several bands on disc electrophoresis. Studies in this laboratory show that SP can be extracted from mitochondria and submitochondrial vesicles with dilute acid. A constant amount of SP (approximately 40% of the protein) is associated with submitochondrial vesicles from liver, kidney and heart. On the other hand, the amount of SP in different mitochondria varies from approximately 65% in liver to 40% in heart. This variation parallels the the decrease in the amount of matrix from liver to heart mitochondria. Special fixation with glutaraldehyde allows visualization of additional material which is associated with the membrane. The material associated with the membrane can be extracted by acid or urea and has been shown to be mainly SP. Most of this material is on the matrix side of the inner membrane, in agreement with the correlation of the amount of SP and the size of the matrix. Membrane associated material has also been shown to be present in other cellular membranes. These results show that SP is not an integral part of the primary organization of the membrane, i.e., the trilaminar arrangement. It appears from these studies that SP forms a secondary level of organization which greatly extends the influence of membranes.

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EPOXIDIZED OILS AS EMULSIFIABLE PESTICIDE STABILIZERS. KEITH L. JOHNSON, Swift & Company, Oak Brook, Illinois.

The relative efficacy of epoxidized soybean oil and epoxidized linseed oil as stabilizers for emulsifiable agricultural toxicants based on Chlordane and Toxaphene are studied. The study is carried out at 0.5% and 1.0% epoxidized oil and systems having varying levels of moisture, both in the absence and presence of iron. The systems are evaluated for emulsion stability at ½, 1, 2, 4 and 24 hr, after 0, 1, 2, 4 and 8 weeks of storage at ambient temperature and at 140 F. The results demonstrate the applicability of epoxidized fatty oils to the stabilization of liquid emulsifiable pesticide concentrates based on chlorinated toxicants. The advantages of epoxidized oils in these systems are discussed. The results obtained were dependent upon the storage condition and the hardness of the water used to prepare the test emulsions. In the range tested higher oxirane oxygen levels yielded no benefit although they could be expected to give better performance for storage periods longer than those used in this study.

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A RAPID GAS CHROMATOGRAPHIC METHOD FOR THE DETERMINATION OF BHA AND BHT IN VEGETABLE OILS. KENNETH T. HARTMAN and LUCIEN C. ROSE, Frito-Lay, Inc., Irving, Texas.

A gas liquid chromatographic technique has been developed which requires about 20 min for the determination of BHA and BHT in vegetable oils. This method involves: the addition of an internal standard to a weighed portion of the oil; dilution of the mixture with carbon disulfide; and injection into the gas chromatograph. BHA and BHT are isolated from the non-volatile vegetable oil by using a short pre-column located in the sample port block of the gas chromatograph. The pre-column is cleaned at the end of each day's operation. The clean pre-column is allowed to equilibrate to oven temperature overnight for the following day's operation. Further identification of BHA and BHT can be achieved with a second GLC column which reverses the elution order of the compounds. Soybean, cottonseed, corn and peanut oil fortified with 20, 60, and 100 ppm each of BHA and BHT showed a recovery range of 97% to 104%.

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COMPOSITIONAL ANALYSIS OF MANNIDE MONOOLEATE EMULSIFYING AGENT (ARLACEL A). H. J. O'NEILL and T. N. YAMAUCHI, IIT Research Institute, Chicago, Illinois.

Analytical studies were carried out on two lots of a commercial emulsifying agent (mannide monooleate) in order to determine reproducibility among lots and overall composition. The samples were subjected to column and thin layer chromatography for major class fractionation and to gas chromatography-mass spectrometry for separation and identification of components comprising each class. The neutral fractions from the column chromatography of each lot represented 52 wt % and 36.4 wt %, respectively and consisted of at least 12 components characteristic of peracylated carbohydrate structures. The intermediate or moderately polar class represented 31.3% and 52.2%, respectively of the original lots and, of the 14 components detected by gas chromatography of their trimethylsilyl ethers, mannide monooleate represented 16.7 wt % and 28.2 wt %, respectively of the original product. The polar fraction contained predominantly carbohydrate polymer and represented 16.7 wt % and 11.2 wt %, respectively. Conjugated dienes and cyclic fatty acid components were present at levels of 1.5 to 2 wt % of the total fatty acid fraction.

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DEVELOPMENT OF IMPROVED BAKER'S SHORTENING THROUGH STATISTICAL UNDERSTANDING OF EMULSIFIER EFFECTIVENESS. D. T. RUSCH and H. M. TRUAX, Atlas Chemical Industries, Inc., Wilmington, Delaware.

High performance, low use level emulsifier blends were developed from information gained from a two phase statistically-oriented experimental plan. Separate blends were developed for specialty cake and specialty icing shortenings, as well as general purpose shortenings. The initial phase of the work tested the relative effects of various surfactants on cake and icing quality attributes. It also determined the replication necessary to overcome the inherent variation in measurement of these attributes and indicated reasonable surfactant use levels. Results suggested that the surfactants highly functional in cakes were inversely effective in icing. The second phase considered more precisely the effects of selected surfactants of Phase 1. Statistical analysis of the results of the Phase 2 work indicated sorbitan monostearate highly functional in improving cake volume and tenderness. The data confirmed Phase 1 indications concerning the inverse functionality of sorbitan monostearate in icings. Polysorbate 60 and plastic type mono- and diglycerides were especially functional in baker's icing shortening.

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DETERMINATION OF POLYSORBATE 60 IN FOODS. CHARLES F. SMULLIN, FRANK P. WETTERAU and VIRGINIA L. OLSANSKI, Atlas Chemical Industries, Inc., Wilmington, Delaware.

To obtain government approval to use emulsifiers in food, it is necessary to develop a valid analytical procedure. Procedures for extracting and isolating polysorbate 60 from foods such as bread, yeast-raised doughnuts, chocolate, chocolate type and sugar type confectionery coatings, cake, cake mixes, shortening, foam-mat dried foods and salad dressings will be presented. A simple gravimetric procedure using the heteropoly barium phosphomolybdate complex as the precipitant will also be described. Where possible, the composition of the food will be presented, along with the applicable validation and recovery data.

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CONJUGATION OF POLYUNSATURATED FATTY ACIDS: METHYL LINOLENATE. T. L. MOUNTS, D. GLOVER and H. J. DUTTON, Northern Regional Research Laboratory, Peoria, Illinois.

The conjugation-isomerization reaction of methyl linolenate with potassium *t*-butoxide has been examined. Reactions were performed at 60 C, 20 hr; 95 C, 4 hr; and 140 C, 2 hr. Products were separated by silver-resin chromatography and characterized according to advanced techniques of analysis. Experiments were performed with tritium-labeled, deuterium-labeled and unlabeled reagents. The isotopic experiments assisted in confirming the theoretical carbanion mechanism of conjugation. The following conclusions were supported by the experimental evidence: the 12 double bond of linolenate (9,12,15) is most susceptible to rearrangement; formation of conjugated triene is a stepwise reaction through conjugated dienetriene; an activated methylene group is normally a prerequisite for the formation of the carbonion; only when a high activation energy is applied to the reaction, for example at 140 C, will conjugated triene be formed from conjugated diene-triene having no activated methylene group; and during high-activation energy reactions (140 C) multiple deprotonations and subsequent protonations occur readily.

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CONJUGATION OF POLYUNSATURATED FATTY ACIDS: METHYL LINOLEATE. T. L. MOUNTS, D. GLOVER and H. J. DUTTON, Northern Regional Research Laboratory, Peoria, Illinois.

The conjugation-isomerization reaction of methyl linoleate with potassium *t*-butoxide has been investigated. Advanced techniques of analysis have been applied to the characterization of the products. Isotopic tracers (^3H, ^2H) aided the determination of the mechanism of conjugation. The following conclusions were supported by the experimental evidence: there is no selectivity as to which bond is shifted; reaction proceeds through the theoretical carbanion mechanism which is subsequently protonated by the reagent; isomerization of the *cis,trans* conjugated isomers to the *trans,trans* conjugated isomer does not proceed through a carbanion but rather some thermal or other mechanism; and the 9-*cis*,11-*trans* conjugated isomer is most susceptible to this secondary reaction.