TABLE II

**Mean Fatty Acid Percentage of A6 and FA8077 Based on Analysis of Three 10-Seed Samples Harvested at Various Periods of Seed Development**

| | Palmitic | | Stearic | | Oleic | | Linoleic | | Linolenic | |
|---|---|---|---|---|---|---|---|---|---|---|
| DAF[a] | FA8077 | A6 | FA8077 | A6 | FA8077 | A6 | FA8077 | A6 | FA8077 | A6 |
| 15 | 31.7 | 25.6 | 22.6 | 17.1 | 14.4 | 18.7 | 21.9 | 26.4 | 12.9 | 12.1 |
| 17 | 16.7 | 23.7 | 14.9 | 18.2 | 18.2 | 16.1 | 33.4 | 27.7 | 17.2 | 14.2 |
| 19 | 21.5 | 16.7 | 6.5 | 19.6 | 21.3 | 15.6 | 31.8 | 32.6 | 15.4 | 12.2 |
| 21 | 14.1 | 10.6 | 4.9 | 31.0 | 42.5 | 15.9 | 24.7 | 28.1 | 13.7 | 14.3 |
| 23 | 12.3 | 9.2 | 5.1 | 41.1 | 44.1 | 15.9 | 26.1 | 22.9 | 11.4 | 10.1 |
| 25 | 12.0 | 8.9 | 4.1 | 45.4 | 53.7 | 16.8 | 21.7 | 21.4 | 8.3 | 7.5 |
| 27 | 9.8 | 7.8 | 4.1 | 41.0 | 52.2 | 20.8 | 25.9 | 22.8 | 8.0 | 6.8 |
| 29 | 9.5 | 7.6 | 3.8 | 41.5 | 53.9 | 20.0 | 25.3 | 23.6 | 7.5 | 6.5 |
| 31 | 9.4 | 7.6 | 3.7 | 41.7 | 51.5 | 19.2 | 28.4 | 24.8 | 7.0 | 5.9 |
| 33 | 9.1 | 8.1 | 3.7 | 39.6 | 50.7 | 17.8 | 29.9 | 28.6 | 6.4 | 5.9 |
| 35 | 9.4 | 8.2 | 3.3 | 35.7 | 49.5 | 19.6 | 31.2 | 30.5 | 6.6 | 6.0 |
| 37 | 9.1 | 8.2 | 3.6 | 38.6 | 50.9 | 20.0 | 29.8 | 27.5 | 6.6 | 5.6 |
| 39 | 8.8 | 8.5 | 3.8 | 35.7 | 48.5 | 20.6 | 32.7 | 29.4 | 6.3 | 5.8 |
| 43 | 8.9 | 8.4 | 3.8 | 32.1 | 42.6 | 19.0 | 38.2 | 34.2 | 6.4 | 6.2 |
| 47 | 8.7 | 8.2 | 4.3 | 32.7 | 41.9 | 19.5 | 36.8 | 33.6 | 6.3 | 6.1 |
| 51 | 8.6 | 8.3 | 3.9 | 32.6 | 45.0 | 17.8 | 36.7 | 35.2 | 5.8 | 6.1 |
| LSD (0.05) | 7.2 | | 3.8 | | 3.0 | | 4.6 | | 3.0 | |

[a]Days after flowering.

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# Lipids in Margarines and Margarine-like Foods

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## ABSTRACT

The lipid composition of margarines from stores in selected locations in the U.S. is reported. The lipids determined include the fatty acids, tocopherols and major plant sterols. Data are included for isomeric octadecenoic fatty acids (14 isomers or groups of isomers) and four isomeric octadecadienoic fatty acids common in partially hydrogenated vegetable fats, insofar as these are separable by capillary gas chromatography. Amounts of individual lipids found in vegetable oil margarines, spreads, imitation and diet margarines were: palmitate, 8.49 to 13.17% (normalized weight percent, calculated as triglyceride); stearate, 4.78 to 9.53%; linoleate, 6.06 to 46.39%; linolenate, 0.18 to 3.57%; α-tocopherol, 0.3 to 24.3 mg/100g; γ-tocopherol, 3.0 to 55.0 mg/100g; δ-tocopherol, 0.5 to 18.9 mg/100g; campesterol, 10.6 to 106.3 mg/100g; stigmasterol, 13.1 to 60.9 mg/100g; sitosterol, 42.3 to 412.9 mg/100g. Amounts of *trans*-unsaturated octadecenoic fatty acids in these margarines varied from 10.74 to 30.06%. Small amounts of the *trans,trans, trans,cis* and *cis,trans* isomers of linoleate also are reported.

## INTRODUCTION

The USDA Nutrient Composition Laboratory has carried out a study of the lipids in consumer margarines to provide detailed information on fatty acids, tocopherols and sterols. Food fats may be described as either hidden, i.e. an integral part of the food and therefore not directly recognizable as fat, or visible, clearly recognizable as fat. Margarine is one of the major visible fats, although not the major contributor of fat to the U.S. diet. It was estimated in 1980 (1) that the per capita consumption of margarine in the U.S. was stabilized at 11 to 13 pounds per year, or about 11 to 13 grams of margarine fat per day. Margarine consumption at this level would supply 99 to 117 calories per day, or 5-6% of a 2000 calorie diet.

The vegetable oils from which most margarines are made are major sources for essential fatty acids and the vitamin E-active tocopherols, and variable amounts of these survive in the processed margarines. In addition, margarines contain plant sterols, principally sitosterol, campesterol and stigmasterol, that may be of significance in human diets; the few margarines made with animal fats will contain cholesterol. Processing of margarines includes partial hydrogenation, one of whose effects is the production of numerous isomeric unsaturated fatty acids differing in the position and geometric configuration of their double bonds. Data on the amounts of these lipids in foods, including margarine, are needed by those investigating the relationship of diet to health, selecting and recommending diets for the general public, or providing diets designed for specific therapeutic purposes. This work was undertaken to provide the maximum amount of such information that may be obtained using current analytical methods.

## MATERIALS AND METHODS

### Sample Collection

All samples were consumer packages bought in grocery stores. The packages were immediately placed individually in Ziploc bags and kept on dry ice until received in the laboratory. Three sample collections are represented in these data:

—I. A local collection of individual units from grocery stores in the Washington, D.C.-Baltimore, MD area; each unit was analyzed separately.

—II. A nationwide collection from stores in four selected U.S. cities, also of individual units that were analyzed separately.

—III. A second nationwide collection from selected stores in five cities.

A one-lb. package of each of the brands to be analyzed was to be bought in each of two stores in the five cities, yielding 10 packages of each brand. These were to be composited and subsampled for analysis. Because of differences in brand distribution, both within and between cities, it was not always possible to follow this plan, and the number of units composited for the samples in Collection III varied from 2 to 10.

The margarines were selected to represent the brands that supply the major part of the margarine eaten in the U.S. as well as the less widely distributed types that are of interest because of their unique composition or source oils. The brands collected and analyzed are listed in Table I; the names are given exactly as they appeared on the package labels.

### Subsampling for Analysis

Samples were stored at 3-5 C for up to one week; for longer periods they were stored at -10 C. They were warmed to the dew point while still in the Ziploc bags to avoid condensation of atmospheric moisture. A subsample was taken by one of two methods: softening or coring. Softening was done as described in AOAC Methods 16.204 (2). A 1-g sample was weighed into a tared 100-ml volumetric flask and diluted to volume with 2:1 chloroform:methanol. Softening was not used to prepare composite samples. Coring was used to subsample some of the single unit samples and all of the composite samples. Cores of approximately 0.25 gram each were taken with a plastic tuberculin syringe whose tip was cut off to leave a 2.8 x 0.18 in. cylinder. With the plunger removed the syringe was pushed into the sample (through a stick of margarine or an equivalent distance into a tub of margarine), and removed: the plunger was inserted and used to discharge the core into the receiving vessel. Samples consisting of only one unit (usually a pound) were subsampled by taking two cores from each of two sticks or from opposite quarters of a tub. Units to be composited were cored to yield a total of five grams, which was dissolved in 100 ml of 2:1 chloroform:methanol, washed in a separatory funnel with 0.58% aqueous NaCl, and then diluted to 200 ml with 2:1 chloroform:methanol.

Aliquots for fatty acid and tocopherol/sterol analysis were dispensed from a Hamilton Diluter/Dispenser (Part No. 10004, Hamilton Company, Reno, Nevada).

### Fatty Acids

Fatty acids were determined by gas chromatography. A glass capillary column 100m x 0.25mm coated with OV275 (Quadrex Corporation, New Haven, Connecticut) was used in a Hewlett Packard Model 5840 gas chromatograph equipped with an all-glass splitter system, an automatic liquid sampler and a flame ionization detector. Other details, including esterification, were as described earlier (3).

The octadecenoate isomers were identified a) from earlier work in this laboratory (4); b) by comparison of their equivalent chain lengths with those of known isomers, where available, and c) from the equivalent chain length data of Scholfield (5). Pure known standards were used to identify the four 9,12-octadecadienoic isomers, *trans,trans, cis,trans, trans,cis* and *cis,cis.*

### Tocopherols and Sterols

Tocopherols and sterols were determined by capillary gas chromatography essentially as described earlier (6), except that a 12m fused silica column coated with SP-2100 (Hewlett Packard Co., Avondale, Pennsylvania) was used at a column temperature of 230 C, with hydrogen as the carrier gas. Aliquots of the total lipid extract were freed of solvent and prepared for chromatography by saponification and derivatization to trimethylsilyl ethers.

### Validation of Methods

*Subsampling.* A series of 11 packages of stick margarines, each of a different brand, were subsampled both by softening and by coring and the results from the two subsamples compared. A t-test of the differences between subsamples found no statistical differences except for stigmasterol, for which results from coring were consistently lower (4%) than from softening.

*Quality Control.* Quality control samples were prepared to monitor analytical performance. Single sticks of margarine from the same production lot of a single margarine were wrapped in aluminum foil, placed in plastic Ziploc bags and stored at -40 to 60 C. One of these quality control margarine sticks was included with each group of samples analyzed as a check on the consistency and reliability of the procedures used. The lipid composition of this quality control sample has been stable over a period of three years (7). The mean and RSD (relative standard deviation) of the results from all analyses of this quality control sample are given in Table II.

Reference standards and internal standards were used for all fatty acid, tocopherol and sterol analyses to assure adequate analytical accuracy and precision (3,6).

## RESULTS AND DISCUSSION

Margarines must conform to a legal definition which requires that they contain at least 80% fat. Most of the items analyzed in this work conformed to this definition, but some were formulated with less than 80% fat, and were termed spreads, imitation margarines or diet margarines (Table I). Only one margarine was made with animal fat; all others contained only vegetable fat. To simplify the discussion the term margarine will be used to refer to all products made with vegetable fat, including imitation margarines, diet margarines and spreads, unless otherwise qualified.

The margarine samples are distinguished here by the source oils used in their preparation, the location where purchased, the amount of fat declared on the label and the type, either stick, or tub (one 1-lb block is included). All this information is available to the consumer. The data are presented in this way to maximize their usefulness, and not to imply that all margarines of similar description purchased in the same location will have similar compositions. The initial sampling plan was designed to permit a statistical evaluation of U.S. margarines, but since the difficulties described above prevented its completion, no such evaluation has been made.

Table III contains the data for the fatty acids (other than the individual isomeric unsaturated fatty acids), the tocopherols and the major sterols.

The principal saturated fatty acids were palmitate and

TABLE I

Margarines Analyzed

| Brand | Source oil[a] | Oil %[b] | Type[c] |
|---|---|---|---|
| Collection I | | | |
| Blue Bonnet Margarine | SBH/SB/CSH | 80 | S |
| Dalebrook Vegetable Margarine | SB/SBH | 80 | S |
| Dew Fresh Margarine | SB/SBH | 80 | Block |
| Empress (contains liquid corn oil) Margarine | C/CH | 80 | S |
| Empress Vegetable Oil Margarine | SBH | 80 | S |
| Farmdale Vegetable Oil Margarine | SB/SBH | 80 | S |
| Fleischmann's 100% Corn Oil Margarine | C/CH | 80 | S |
| Giant Food 100% Corn Oil Margarine | C/CH | 80 | S |
| Giant Food Vegetable Margarine | SB/SBH/CS | 80 | S |
| Golden Belle Margarine | SB/SBH | 80 | S |
| Golden Maid Margarine | SBH | 80 | S |
| Gold O'Corn Margarine | C/CH | 80 | S |
| Grand Union Corn Oil Margarine | C/CH | 80 | S |
| Grand Union Margarine | SB/SBH | 80 | S |
| Ideal Vegetable Oil Margarine | SB/SBH | 80 | S |
| Imperial Margarine | SBH/P/SN/C/CS | 80 | S |
| Land O' Lakes Corn Oil Margarine | C/CH | 80 | S |
| Little Darling Margarine | SB/SBH | 80 | S |
| Mazola Margarine | C/SBH | 80 | S |
| Mazola sweet-unsalted Margarine | C/SBH | 80 | S |
| Mrs. Filbert's 100% Corn Oil Margarine | C/CH | 80 | S |
| Mrs. Filbert's Golden Quarters Margarine | SBH | 80 | S |
| Pantry Pride Margarine | SB/SBH | 80 | S |
| Parkay Margarine | SBH | 80 | S |
| Promise Margarine | SN/SBH/CSH | 80 | S |
| Red & White Golden Quarters Margarine | SB/SBH | 80 | S |
| Richfood 100% Corn Oil Margarine | C/CH | 80 | S |
| Richfood Margarine | SB/SBH | 80 | S |
| Scotch Buy Vegetable Oil | SBH/SB | 80 | S |
| Sommer Maid Margarine | SB/SBH | 80 | S |
| Collection II | | | |
| Autumn Natural Margarine | SBH/C/SN/PH | 80 | T |
| Bluebrook Margarine | SB/SBH | 80 | S |
| Kroger Eatmore Margarine | SB/SBH | 80 | S |
| Farm Charm Corn Oil Margarine | C/CH | 80 | S |
| Farm Charm Margarine | SB/SBH | 80 | S |
| Farm Gold Light Spread | SB/SBH | 60 | S |
| Golden Soft Margarine | SBH/SB | 80 | T |
| Harvest Day Margarine | LH | 80 | S |
| Heritage House Vegetable Oil Margarine | SB/SBH | 80 | S |
| Kountry Fresh Corn Oil Margarine | C/CH | 80 | S |
| Lady Lee Corn Oil Margarine | C/CH | 80 | S |
| Lady Lee Vegetable Margarine | SBH/SB/P | 80 | S |
| Land O' Lakes Margarine | SBH | 80 | S |
| Saffola (contains pure liquid safflower oil) Margarine | SF/SB/SBH/CSH | 80 | S |
| Shedd's 52% Vegetable Oil Spread | SB/SBH | 52 | S |
| Superbrand Margarine | SB/SBH | 80 | S |
| Table Maid Spread | SBH/SB/P | 60 | S |
| Collection III | | | |
| Blue Bonnet Light Tasty Spread | SBH/CSH | 60 | T |
| Blue Bonnet Margarine | SBH/SB/CSH | 80 | S |
| soft Blue Bonnet Margarine | SBH/CSH | 80 | T |
| soft whipped Blue Bonnet Margarine | SBH/CSH | 80 | T |
| soft Chiffon Margarine | SB/SBH | 80 | T |
| soft stick Chiffon Margarine | SB/SBH/CSH | 80 | S |
| whipped Chiffon Margarine | SB/SBH | 80 | T |
| soft Diet Parkay Reduced Calorie Margarine | SBH | 40 | T |
| Fleischmann's 100% Corn Oil Margarine | C/CH | 80 | S |
| soft Fleischmann's 100% Corn Oil Margarine | C/CH | 80 | T |
| sweet unsalted Fleischmann's 100% Corn Oil Margarine | C/CH | 80 | S |
| Fleischmann's Light Corn Oil Spread | C/CH | 60 | T |
| Imperial Margarine | SBH/CSH | 80 | S |
| Imperial Margarine | SBH/CSH/SNH/CH | 80 | S |
| diet Imperial Imitation Margarine | SBH/CSH | 40 | T |
| diet Imperial Imitation Margarine | SBH/PH/P | 40 | T |
| soft Imperial Margarine | SBH/CSH | 80 | T |
| soft Imperial Margarine | SBH/CSH/SNH/CH | 80 | T |
| whipped Imperial Margarine | SBH/CSH | 80 | T |
| Land O' Lakes Margarine | SBH | 80 | S |
| Land O' Lakes soft Margarine | SBH | 80 | T |
| diet Mazola 100% Corn Oil Imitation Margarine | C/CH | 40 | T |
| Mazola Margarine | C/SBH | 80 | S |
| Mazola sweet-unsalted Margarine | C/SBH | 80 | S |
| Mrs. Filbert's 100% Corn Oil Margarine | C/CH | 80 | T |
| Mrs. Filbert's Golden Quarters Margarine | SBH | 80 | S |
| Mrs. Filbert's Soft Golden Margarine | SBH | 80 | T |
| Mrs. Filbert's Spread 25 | SBH | 60 | T |
| Mrs. Filbert's Whipped Margarine | SBH | 80 | T |
| Mrs. Filbert's Whipped Spread | SBH | 60 | S |
| Nucoa no-burn Margarine | | | |
| Nucoa Soft Margarine | SB/SBH/CSH | 80 | T |
| Parkay Light Corn Spread | C/CH | 60 | T |
| Parkay Light Spread | SBH | 60 | T |
| Parkay Margarine | SBH | 80 | S |
| soft Parkay Margarine | SBH | 80 | T |
| Promise Margarine | SN/SBH/CSH | 80 | S |
| Promise Margarine | SN/SBH/CSH | 80 | T |

[a]C, corn oil; CH, hydrogenated corn oil; CS, cottonseed oil; CSH, hydrogenated cottonseed oil; L, lard; LH, hydrogenated lard; P, palm oil; PH, hydrogenated palm oil; SB, soybean oil; SBH, hydrogenated soybean oil; SF, safflower oil; SN, sunflower oil; SNH, hydrogenated sunflower oil.
[b]Label information.
[c]S = stick, T = tub.

stearate, although saturated fatty acids with carbon chains longer and shorter than these, as well as odd chain saturated acids, were present in small amounts. Palmitate varied from 8.49 to 13.17%, and stearate from 4.78 to 9.53%. The total saturated fatty acids ranged from a low of 15.27% to a high of 23.79%. The amounts of stearate are only slightly higher than those in the unprocessed source oils (8), indicating that complete hydrogenation of unsaturates was not extensive.

The octadecenoates were 36.88 to 70.68% in all margarines. Two isomers, 18:1$\omega$9*c* and 18:1$\omega$7*c*, are present in most unprocessed vegetable oils; the other *cis* and *trans* positional isomers were produced by isomerization during partial hydrogenation. The sum of the *cis*-octadecenoates was always greater than that of the *trans*-octadecenoates: the amounts of *cis* isomers ranged from 21.61 to 40.65% and those of *trans* from 10.74 to 30.06%. Tub margarines contained lower amounts of *trans*-octadecenoates than did the stick margarines. Estimates of the amounts of individual positional and geometric mono- and di-unsaturated 18 carbon fatty acids are given in Table IV. Most available fatty acid data for margarines, as well as other foods, have been

**TABLE III**

**Margarines and Margarine-Like Foods: Fatty Acids, Tocopherols and Sterols**

| Source oils[a] | C/CH | C/CH | C/CH | C/CH | C/CH | C/CH | C/CH | C/CH | C/CH | C/CH | C/SBH | C/SBH |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Collection | I | II | II | III | III | III | III | III | III | III | I | I |
| Location[b] | W | L | A | ABCHL | C | ABCHL | AB | ABCHL | C | AH | W | W |
| No. samples | 8 | 1 | 2 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| Units/sample | 1 | 1 | 1 | 10 | 2 | 10 | 4 | 9 | 2 | 4 | 1 | 1 |
| Fat %[c] | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 60 | 60 | 40 | 80 | 80 |
| Type[d] | S | S | S | S | S | T | T | T | T | T | S | S |
| Fatty acids: (normalized wt %)[e] | | | | | | | | | | | | |
| 16:0 | 10.67 ± 0.41 | 10.67 | 10.42 ± 0.16 | 10.62 | 10.79 | 10.55 | 10.59 | 10.72 | 11.18 | 10.68 | 10.73 | 10.76 |
| 18:0 | 6.94 ± 0.46 | 7.56 | 6.31 ± 0.20 | 6.44 | 6.61 | 5.94 | 6.10 | 6.29 | 5.89 | 4.99 | 6.17 | 6.39 |
| 18:1*c* | 28.53 ± 1.08 | 28.43 | 29.66 ± 0.23 | 26.67 | 26.95 | 25.47 | 26.96 | 26.09 | 26.62 | 25.66 | 26.56 | 27.35 |
| 18:1*t* | 21.75 ± 2.04 | 22.32 | 24.89 ± 3.49 | 20.19 | 21.05 | 11.96 | 13.48 | 11.67 | 11.78 | 12.39 | 19.66 | 18.44 |
| 18:2ω6*cc* | 30.09 ± 2.61 | 28.10 | 26.85 ± 3.38 | 33.79 | 32.74 | 43.65 | 40.38 | 42.92 | 42.45 | 41.95 | 34.55 | 35.16 |
| 18:3ω3*ccc* | 0.67 ± 0.25 | 0.48 | 0.48 ± 0.11 | 0.69 | 0.56 | 0.96 | 0.76 | 0.76 | 0.79 | 0.52 | 0.45 | 0.51 |
| Other FA | 1.36 ± 0.44 | 2.43 | 1.38 ± 0.60 | 1.60 | 1.30 | 1.46 | 1.72 | 1.54 | 1.29 | 3.82 | 1.88 | 1.39 |
| Total saturated FA | 18.11 ± 0.58 | 19.00 | 17.15 ± 0.56 | 17.65 | 18.27 | 17.10 | 17.38 | 17.69 | 17.69 | 16.79 | 17.67 | 17.80 |
| Tocopherols (mg/100g): | | | | | | | | | | | | |
| α-Tocopherol | 15.8 ± 2.3 | 4.8 | 15.6 ± 6.4 | 15.2 | 0.8 | 5.1 | ND[f] | ND | ND | 1.1 | 12.8 | 14.1 |
| γ-Tocopherol | 49.8 ± 9.8 | 21.9 | 43.4 ± 9.0 | 49.7 | 12.0 | 46.0 | ND | ND | ND | 3.3 | 36.8 | 52.1 |
| δ-Tocopherol | 4.4 ± 2.0 | 1.9 | 3.6 ± 1.9 | 0.5 | 1.8 | ND | ND | ND | ND | 3.0 | 7.4 | 8.7 |
| Vitamin E (α-tocopherol equiv/100g)[g] | 20.8 ± 3.2 | 7.0 | 19.4 ± 7.3 | 20.2 | 2.0 | 9.7 | ND | ND | ND | 1.4 | 16.5 | 19.3 |
| Sterols (mg/100g): | | | | | | | | | | | | |
| Campesterol | 103.5 ±12.4 | 57.2 | 106.3 ±36.4 | 87.0 | 96.6 | 92.3 | ND | ND | ND | 62.8 | 74.2 | 78.8 |
| Stigmasterol | 47.0 ± 3.5 | 33.8 | 43.4 ± 5.6 | 43.58 | 46.1 | 45.5 | ND | ND | ND | 30.4 | 38.5 | 39.0 |
| Sitosterol | 359.8 ±28.5 | 235.4 | 357.1 ±119.3 | 363.1 | 412.9 | 393.1 | ND | ND | ND | 297.2 | 280.0 | 279.2 |

| Source oils | C/SBH | C/SBH | SBH | SBH | SBH | SBH | SBH | SBH | SBH | SBH | SBH | SBH | SBH |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Collection | III | III | I | II | III | III | III | III | III | III | III | III | III |
| Location | ABHL | C | W | C | ABCHL | B | C | A | ABCHL | B | C | A | A |
| No. samples | 1 | 1 | 4 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| Units/sample | 8 | 2 | 1 | 1 | 10 | 2 | 2 | 1 | 10 | 2 | 2 | 2 | 2 |
| Fat % | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 60 | 80 | 80 | 80 | 80 | 60 |
| Type | S | S | S | S | S | S | S | S | T | T | T | T | T |
| Fatty acids (normalized wt %): | | | | | | | | | | | | | |
| 16:0 | 10.66 | 10.88 | 10.74 ± 0.36 | 9.79 | 11.04 | 10.55 | 10.48 | 10.97 | 10.77 | 10.90 | 10.46 | 10.80 | 10.80 |
| 18:0 | 6.38 | 7.08 | 7.50 ± 0.73 | 6.93 | 7.38 | 7.71 | 8.41 | 8.22 | 7.04 | 7.50 | 6.59 | 7.17 | 7.71 |
| 18:1*c* | 26.54 | 26.42 | 32.74 ± 5.82 | 40.65 | 38.51 | 32.42 | 33.58 | 35.90 | 30.44 | 31.48 | 34.12 | 31.92 | 32.48 |
| 18:1*t* | 19.14 | 18.59 | 25.64 ± 4.99 | 30.03 | 28.80 | 30.06 | 23.44 | 28.22 | 12.67 | 15.45 | 18.44 | 16.71 | 16.44 |
| 18:2ω6*cc* | 33.84 | 32.22 | 16.19 ± 9.94 | 6.06 | 7.56 | 8.98 | 17.42 | 9.15 | 30.43 | 26.78 | 23.72 | 26.83 | 25.60 |
| 18:3ω3*ccc* | 0.41 | 0.62 | 1.31 ± 1.52 | 0.30 | 0.18 | 0.25 | 2.04 | 0.36 | 2.24 | 1.70 | 1.90 | 1.58 | 1.27 |
| Other FA | 3.03 | 4.21 | 5.87 ± 2.83 | 6.24 | 6.54 | 10.04 | 4.63 | 7.18 | 6.40 | 6.19 | 4.78 | 5.00 | 5.71 |
| Total saturated FA | 18.96 | 19.81 | 19.12 ± 1.02 | 17.41 | 19.29 | 19.56 | 19.64 | 20.01 | 21.22 | 19.20 | 17.70 | 18.59 | 19.14 |

**TABLE III (Continued)**

| | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Tocopherols (mg/100g): | | | | | | | | | | | | | |
| α-Tocopherol | 9.8 | 0.3 | 5.1 ± 0.5 | 2.9 | 5.4 | 7.2 | ND | 5.2 | 5.2 | 2.2 | 3.0 | 6.7 | 4.0 |
| γ-Tocopherol | 43.6 | 17.2 | 35.1 ± 7.2 | 24.2 | 29.1 | 48.0 | ND | 39.5 | 34.1 | 11.1 | 52.1 | 10.0 | 29.8 |
| δ-Tocopherol | 7.1 | 3.3 | 13.7 ± 1.2 | 8.3 | 8.2 | 13.1 | ND | 11.4 | 10.0 | 7.1 | 21.5 | 5.9 | 9.2 |
| Vitamin E (α-tocopherol equiv/100 g) | 14.2 | 2.0 | 8.6 ± 1.1 | 5.3 | 8.4 | 12.0 | ND | 9.2 | 8.6 | 3.4 | 8.2 | 7.7 | 6.9 |
| Sterols (mg/100g): | | | | | | | | | | | | | |
| Campesterol | 76.5 | 70.0 | 35.2 ± 5.7 | 27.9 | 25.7 | 35.5 | ND | 28.5 | 30.3 | 38.6 | 48.5 | 31.5 | 24.1 |
| Stigmasterol | 37.8 | 36.4 | 32.9 ± 2.1 | 23.3 | 26.6 | 32.9 | ND | 33.4 | 30.0 | 41.5 | 41.4 | 35.5 | 27.3 |
| Sitosterol | 266.6 | 291.0 | 98.4 ± 7.2 | 81.7 | 80.8 | 93.4 | ND | 103.1 | 96.2 | 116.6 | 126.8 | 101.3 | 83.9 |

| Source oils | SBH | SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SBH/SB |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Collection | III | III | I | I | II | II | II | II | II | III | III | III | I |
| Location | C | AC | W | W | H | A | C | A | C | AL | ABCH | ACHL | W |
| No. samples | 1 | 1 | 1 | 10 | 1 | 2 | 2 | 1 | 1 | 1 | 1 | 1 | 1 |
| Units/sample | 2 | 4 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 4 | 8 | 6 | 1 |
| Fat % | 60 | 40 | 80 | 80 | 80 | 80 | 80 | 60 | 52 | 80 | 80 | 80 | 80 |
| Type | T | T | B | S | S | S | S | S | S | S | T | T | S |
| Fatty acids (normalized wt %): | | | | | | | | | | | | | |
| 16:0 | 10.56 | 11.05 | 10.42 | 10.39 ± 0.19 | 11.27 | 10.50 ± 0.29 | 10.13 ± 0.51 | 11.50 | 10.47 | 10.78 | 10.50 | 10.46 | 12.49 |
| 18:0 | 7.01 | 6.90 | 8.20 | 8.98 ± 0.61 | 8.48 | 8.32 ± 0.34 | 7.83 ± 1.09 | 7.65 | 8.11 | 8.58 | 6.56 | 6.36 | 9.53 |
| 18:1*c* | 30.44 | 31.47 | 28.23 | 28.07 ± 0.61 | 30.00 | 29.10 ± 0.30 | 30.39 ± 0.72 | 34.96 | 30.06 | 27.14 | 29.00 | 29.52 | 25.61 |
| 18:1*t* | 13.35 | 12.09 | 21.04 | 21.27 ± 1.15 | 20.44 | 24.17 ± 0.04 | 20.12 ± 0.11 | 25.23 | 21.09 | 19.19 | 10.74 | 11.04 | 24.97 |
| 18:2ω6*cc* | 30.71 | 31.22 | 27.04 | 26.30 ± 1.60 | 24.12 | 23.16 ± 1.39 | 26.31 ± 1.90 | 12.43 | 24.61 | 27.44 | 36.69 | 36.32 | 18.85 |
| 18:3ω3*ccc* | 2.40 | 1.96 | 3.30 | 2.93 ± 0.21 | 2.37 | 2.10 ± 0.03 | 3.57 ± 0.30 | 0.44 | 2.59 | 2.79 | 3.76 | 3.44 | 1.56 |
| Other FA | 5.53 | 5.32 | 1.76 | 2.06 ± 0.45 | 3.31 | 2.64 ± 1.02 | 1.66 ± 1.20 | 7.80 | 3.08 | 4.08 | 2.75 | 2.85 | 6.98 |
| Total saturated FA | 18.38 | 18.88 | 19.44 | 20.01 ± 0.76 | 20.50 | 19.32 ± 0.07 | 18.56 ± 1.85 | 20.17 | 19.36 | 20.77 | 17.72 | 17.44 | 23.79 |
| Tocopherols (mg/100g): | | | | | | | | | | | | | |
| α-Tocopherol | 1.0 | 1.2 | 4.7 | 6.7 ± 1.1 | 3.1 | 5.5 ± 0.4 | 5.3 ± 0.6 | 3.5 | 7.9 | 3.3 | 2.5 | 4.3 | 4.2 |
| γ-Tocopherol | 21.0 | 11.8 | 38.1 | 47.1 ± 3.3 | 30.9 | 38.8 ± 2.9 | 40.5 ± 8.6 | 26.9 | 44.9 | 16.9 | 53.7 | 50.4 | 25.7 |
| δ-Tocopherol | 7.2 | 3.5 | 14.1 | 16.6 ± 1.8 | 13.2 | 12.8 ± 1.3 | 14.0 ± 3.5 | 10.4 | 15.5 | 11.6 | 16.6 | 16.7 | 13.3 |
| Vitamin E (α-tocopherol equiv/100g) | 3.1 | 2.4 | 8.5 | 11.4 ± 1.3 | 6.2 | 9.4 ± 0.7 | 9.3 ± 1.4 | 6.2 | 12.4 | 5.0 | 7.9 | 9.3 | 6.8 |
| Sterols (mg/100g): | | | | | | | | | | | | | |
| Campesterol | 29.0 | 10.6 | 32.5 | 45.7 ± 5.8 | 30.9 | 38.6 ± 3.4 | 38.6 ±10.4 | 24.3 | 42.5 | 42.0 | 39.5 | 44.2 | 23.1 |
| Stigmasterol | 27.4 | 13.1 | 31.9 | 37.0 ± 2.9 | 30.0 | 33.4 ± 1.0 | 31.8 ± 6.3 | 22.6 | 32.1 | 37.8 | 39.8 | 39.9 | 22.1 |
| Sitosterol | 78.8 | 42.3 | 100.1 | 114.9 ±18.2 | 104.6 | 105.8 ± 7.7 | 105.6 ±23.0 | 76.5 | 104.6 | 120.1 | 119.9 | 116.7 | 71.0 |

| Source oils | SBH/SB | SB/SBH/ CS | SB/SBH/ CSH | SB/SBH/ CSH | SBH/SB CSH | SBH/SB/ CSH | SBH/ CSH | SBH/ CSH | SBH/ CSH | SBH/ CSH | SBH/ CSH | SBH/ CSH | SBH/ CSH |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Collection | II | I | III | III | I | III | III | III | III | III | III | III | III |
| Location | L | W | CH | L | W | ABCHL | BCH | ABCL | AH | H | BC | ABCHL | L |
| No. samples | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| Units/sample | 1 | 1 | 4 | 2 | 1 | 10 | 6 | 8 | 4 | 2 | 3 | 9 | 2 |
| Fat % | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 60 | 40 |
| Type | T | S | S | T | S | S | S | T | T | T | T | T | T |
| Fatty acids (normalized wt %): | | | | | | | | | | | | | |
| 16:0 | 10.26 | 10.88 | 11.37 | 12.44 | 10.47 | 10.51 | 11.26 | 10.49 | 10.89 | 10.92 | 11.11 | 10.70 | 13.17 |
| 18:0 | 6.55 | 8.74 | 7.58 | 7.41 | 8.00 | 7.84 | 6.67 | 7.36 | 6.10 | 7.13 | 5.96 | 7.78 | 4.78 |
| 18:1*c* | 35.87 | 29.19 | 30.55 | 26.22 | 29.78 | 29.67 | 29.96 | 30.58 | 32.19 | 33.25 | 28.79 | 30.96 | 26.00 |
| 18:1*t* | 16.81 | 19.98 | 18.53 | 14.44 | 20.85 | 21.85 | 23.33 | 14.56 | 17.84 | 16.64 | 14.78 | 14.84 | 17.61 |
| 18:2ω6*cc* | 23.86 | 26.55 | 26.30 | 32.14 | 26.05 | 24.75 | 22.41 | 29.73 | 25.11 | 25.04 | 32.70 | 28.46 | 19.46 |
| 18:3ω3*ccc* | 2.85 | 3.04 | 3.02 | 2.72 | 3.01 | 2.81 | 1.87 | 2.38 | 1.76 | 1.40 | 3.17 | 2.26 | 0.87 |
| Other FA | 3.80 | 1.62 | 2.66 | 4.63 | 1.84 | 2.57 | 4.50 | 4.90 | 6.11 | 5.62 | 3.50 | 5.00 | 18.10 |
| Total saturated FA | 17.61 | 20.34 | 19.92 | 21.15 | 19.12 | 19.21 | 18.94 | 18.68 | 17.82 | 18.66 | 17.76 | 19.34 | 19.11 |

**TABLE III (Continued)**

| | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Tocopherols (mg/100g): | | | | | | | | | | | | | |
| α-Tocopherol | ND | 8.4 | 4.7 | 1.4 | 8.7 | 5.8 | 5.7 | 4.8 | 3.2 | 2.0 | 5.2 | ND | 2.0 |
| γ-Tocopherol | ND | 55.0 | 3.6 | 8.0 | 49.1 | 44.0 | 14.7 | 3.0 | 7.5 | 2.5 | 4.6 | ND | 11.5 |
| δ-Tocopherol | ND | 18.9 | 1.5 | 4.6 | 17.1 | 13.4 | 9.5 | 1.8 | 6.0 | 1.5 | ND | ND | 5.9 |
| Vitamin E (α-tocopherol equiv/100g) | ND | 13.9 | 5.1 | 2.2 | 13.6 | 10.2 | 7.2 | 5.1 | 3.9 | 2.2 | 5.6 | ND | 3.2 |
| Sterols (mg/100g): | | | | | | | | | | | | | |
| Campesterol | ND | 37.5 | 39.5 | 18.4 | 51.1 | 32.0 | 47.2 | 36.1 | 40.7 | 44.5 | 23.9 | ND | 21.6 |
| Stigmasterol | ND | 34.0 | 37.8 | 16.1 | 39.2 | 34.4 | 35.6 | 36.6 | 45.8 | 49.4 | 21.4 | ND | 19.2 |
| Sitosterol | ND | 108.2 | 124.6 | 64.6 | 142.6 | 112.0 | 125.6 | 122.5 | 144.5 | 150.1 | 91.3 | ND | 56.8 |

| Source oils | SBH/C/ SN/PH | SBH/ SB/P | SBH/ SB/P | SBH/P/ SN/C/CS | SBH/PH/ P | SBH/CSH/ SNH/CH | SBH/CSH/ SNH/CH | SF/SB/ SBH/CSH | SN/SBH CSH | SN/SBH CSH | SN/SBH CSH | LH |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Collection | II | II | II | I | III | III | III | II | I | III | III | II |
| Location | L | L | L | W | C | AL | CL | L | W | BC | BC | L |
| No. samples | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| Units/sample | 1 | 1 | 1 | 1 | 2 | 4 | 3 | 1 | 1 | 4 | 2 | 1 |
| Fat % | 80 | 80 | 60 | 80 | 40 | 80 | 80 | 80 | 80 | 80 | 80 | 80 |
| Type | T | S | S | S | T | S | T | S | S | S | T | S |
| Fatty acids (normalized wt %): | | | | | | | | | | | | |
| 16:0 | 11.20 | 9.96 | 9.85 | 11.90 | 10.80 | 11.70 | 11.89 | 10.64 | 8.85 | 9.21 | 8.49 | 24.30 |
| 18:0 | 5.32 | 8.81 | 8.81 | 6.43 | 6.29 | 6.41 | 5.58 | 6.64 | 7.34 | 6.69 | 5.91 | 15.18 |
| 18:1*c* | 25.38 | 27.84 | 28.32 | 26.97 | 28.95 | 26.60 | 26.86 | 21.61 | 22.09 | 22.44 | 22.24 | 41.67 |
| 18:1*t* | 18.24 | 20.87 | 20.40 | 23.39 | 17.02 | 25.49 | 18.27 | 17.12 | 14.82 | 22.04 | 14.64 | 3.94 |
| 18:2ω6*cc* | 21.34 | 28.16 | 28.19 | 17.74 | 26.21 | 14.37 | 20.52 | 40.35 | 43.57 | 35.49 | 46.39 | 8.94 |
| 18:3ω3*ccc* | 0.84 | 3.12 | 3.53 | 1.01 | 1.83 | 0.58 | 0.94 | 1.92 | 0.71 | 0.26 | 0.58 | 0.27 |
| Other FA | 17.68 | 1.24 | 0.90 | 12.57 | 8.90 | 14.85 | 15.92 | 1.72 | 2.63 | 3.86 | 1.76 | 5.70 |
| Total saturated FA | 17.74 | 19.24 | 19.18 | 19.66 | 17.94 | 19.34 | 18.56 | 18.06 | 17.16 | 17.44 | 15.27 | 41.78 |
| Tocopherols (mg/100g): | | | | | | | | | | | | |
| α-Tocopherol | ND | 6.5 | 4.9 | 4.4 | 0.8 | 4.3 | 4.5 | 5.2 | 24.3 | 5.6 | 5.2 | ND |
| γ-Tocopherol | ND | 49.7 | 37.9 | 27.5 | 15.2 | 25.0 | 4.1 | 39.1 | 13.8 | 9.8 | 5.7 | 1.7 |
| δ-Tocopherol | ND | 17.1 | 14.2 | 12.6 | 6.7 | 14.1 | 2.5 | 14.6 | 5.9 | 2.5 | 5.2 | ND |
| Vitamin E (α-tocopherol equiv/100 g) | ND | 11.5 | 8.7 | 7.2 | 2.3 | 6.8 | 4.9 | 9.1 | 25.7 | 6.6 | 5.8 | 0.2 |
| Sterols (mg/100g): | | | | | | | | | | | | |
| Campesterol | ND | 42.5 | 33.9 | 35.0 | 18.8 | 48.4 | 61.6 | 33.8 | 28.1 | 33.0 | 25.7 | 2.0 |
| Stigmasterol | ND | 39.3 | 28.2 | 30.1 | 17.1 | 41.5 | 60.9 | 30.2 | 26.6 | 23.0 | 20.0 | 1.6 |
| Sitosterol | ND | 123.8 | 79.5 | 92.1 | 56.0 | 154.1 | 219.9 | 84.1 | 141.3 | 137.7 | 133.8 | 5.4 |

[a]C = corn oil, CH = hydrogenated corn oil, CS = cottonseed oil, CSH = hydrogenated cottonseed oil, L = lard, LH = hydrogenated lard, P = palm oil, PH = hydrogenated palm oil, SB = soybean oil, SBH = hydrogenated soybean oil, SF = safflower oil, SN = sunflower oil and SNH = hydrogenated sunflower oil.

[b]A = Atlanta, B = Boston, C = Chicago, H = Houston, L = Los Angeles and W = Washington-Baltimore.

[c]Label information.

[d]S = stick, T = tub and B = block.

[e]Means for two samples are given as the mean and the range. Means for more than two samples are given as the mean ± standard deviation.

[f]ND = not detected.

[g]α-tocopherol equiv = mg α-tocopherol + (0.1)mg γ-tocopherol.

obtained in the past using packed columns. On packed columns, as well as many capillary columns, these isomers do not separate, and are reported only as 18:1, or octadecenoate, or sometimes as oleate, the ω9*c* isomer. Although ω9*c* was the major isomer, it was often less than 50% of the total 18:1. For example, the first item in Tables III and IV had a total monoene content of 50.58% and only 18.72% oleate. Because of these differences, comparisons with existing data should be made with caution.

All isomers were not separated, either because no separation was possible or because the separation, though possible, did not occur for that particular sample. Column overload, or the presence of a large adjacent peak, affected separation. No complete separation of all octadecenoic isomers has ever been achieved, by GLC or any other method. As a result some of the data in Table IV and Table V are listed as the sums of two or more isomers that could not be separated. Most of these isomers have been reported previously in partially hydrogenated vegetable fats (9).

There were seven chromatographic peaks that were wholly or principally *cis*, and seven that were wholly or principally *trans*. The groups of *cis*- and *trans*-octadecenoate peaks were not completely separated, but overlapped. This overlap is characteristic of octadecenoates chromatographed on the polar columns that are preferred for isomer separation. The column temperature used for these analyses was chosen because it gave the best overall separation of the individual peaks and of the groups of *cis* and *trans* peaks. Even so, ω5*t* was not separable from ω12*c* and ω11*c*, nor was ω13*c* separable from ω9*t* and ω11*t*. The peak that would include the ω5*t* was included in the *cis* summation, and the peak that would include ω13*c* in the *trans* summation, because the other accompanying isomers had the double bond nearer the center of the carbon chain. It has been found that isomers with the double bonds near the center of the chain predominate in partially hydrogenated fats. It is assumed that the amounts of ω5*t* and ω13*c* were low.

The ω8*c* isomer was sometimes, but not always, separated from ω9*c*+10*c*: frequently it appeared only as a nonquantifiable shoulder on the rear slope of the ω9*c*+10*c* peak. If this occurred, the data given in Table IV are for the com-

TABLE II

**Validation: Variability of Quality Control (Sticks) Analyzed Over a 3-Year Period**

| Identity[a] | N | Mean | RSD |
|---|---|---|---|
| Fatty acids (normalized wt %): | | | |
| 16:0 | 26 | 10.43 | 1.91 |
| 18:0 | 26 | 7.09 | 1.35 |
| 18:1ω13*t* | 17 | 0.08 | 11.1 |
| 18:1ω12*t*+10*t* | 28 | 3.62 | 3.13 |
| 18:1ω9*t*+11*t*+13*c* | 28 | 4.29 | 1.71 |
| 18:1ω8*t* | 28 | 5.03 | 1.59 |
| 18:1ω7*t* | 28 | 3.87 | 2.90 |
| 18:1ω6*t* | 28 | 2.82 | 1.14 |
| 18:1ω?*t* | 26 | 0.74 | 23.8 |
| 18:1ω12*c*+11*c*+5*t* | 28 | 3.33 | 2.02 |
| 18:1ω9*c*+10*c* | 28 | 18.52 | 1.10 |
| 18:1ω8*c* | 28 | 1.70 | 5.52 |
| 18:1ω7*c* | 28 | 1.70 | 2.02 |
| 18:1ω6*c* | 28 | 1.69 | 1.98 |
| 18:1ω5*c* | 28 | 0.53 | 15.9 |
| 18:1ω?*c* | 3 | 0.36 | 8.42 |
| 18:2ω6*cc* | 26 | 33.31 | 1.50 |
| 18:2ω6*ct* | 18 | 0.15 | 10.4 |
| 18:2ω6*tc* | 10 | 0.04 | 40.9 |
| Unsaponifiables (mg/100g): | | | |
| α-Tocopherol | 16 | 12.8 | 9.1 |
| γ-Tocopherol | 16 | 34.3 | 5.7 |
| δ-Tocopherol | 16 | 2.3 | 11.6 |
| Campesterol | 16 | 94.7 | 6.7 |
| Stigmasterol | 16 | 49.5 | 4.4 |
| Sitosterol | 16 | 353.9 | 2.3 |

[a]The number before the colon gives the number of carbon atoms in the chain; the first number after the colon gives the number of double bonds. The number following the ω gives the number of carbons following the double bond farthest from the carboxyl. *Cis* and *trans* are designated *c* and *t*.

Uncertainty regarding the position of the double bond is indicated by "?".

bination of the three isomers, although the principle isomer was undoubtedly ω9*c*. In a few instances the ω12*t*+10*t* peak was not separated from ω9*t*+10*t*+13*c*, nor ω9*t*+11*t*+13*c* separated from ω8*t*, and data are given for these combinations.

Ratios of the amounts of *trans* and *cis* at the same position can be estimated for only the ω6, ω7 and ω8 octadecenoates, since these were the only ones adequately separated. The unsaturation at the ω6 position was 53.8±11.6% *trans*, that at the ω7 position 66.8±3.87% *trans* and that at the ω8 position 75.3±2.54% *trans*. The differences in the percentages of *trans* may have been due to the fact that not all of the double bonds of the six isomers participated in the isomerization. Some of the ω7*c* could have been present originally in the oils, and some of the ω6*c* could have been the result of the saturation of the Δ9 double bond of linoleate. Only the ω8, both *cis* and *trans*, was wholly formed during partial hydrogenation. Catalytic isomerization has been reported to yield an equilibrium mixture that is 75% *trans* (10), a figure identical to that found for ω8, which suggests that *cis* and *trans* isomers at a chain position not originally present may be equilibrium mixtures of *cis* and *trans*. The observation also suggests that the data are internally consistent, and provides a degree of validation.

Linoleate, 18:2ω6*cc*, was the principal polyunsaturated fatty acid, as well as the fatty acid with the greatest variation, ranging from 6.06 to 46.39%, over all margarines. Up to 3.57% of linolenate, 18:3ω3*ccc*, was found: this acid averaged 1.62%. The percentage of *cis,cis*-methylene-interrupted dienoic fatty acid can be found by summing the amounts of 18:2ω6*cc* and 18:3ω3*ccc*.

Partial hydrogenation produces positional and geometric isomers of the polyunsaturated fatty acids as well as of the monounsaturated acids, but we have data for only the 9,12-octadecadienoates. Other small peaks adjacent to them may have been positional isomers, but these were not identified; they have been included in the "Other" classification in Table III. All vegetable oil margarines contained at least trace amounts of all four 9,12 isomers. By this method, the *trans,trans* isomer exceeded 1% in only four of the 63 items listed. It usually was accompanied by a closely following larger peak which would not be separated on shorter or less efficient columns. The *trans,cis* and *cis,trans* isomers always were present, in amounts seldom exceeding 2%.

The sums of the minor fatty acids are given in Table III as "Other"; these fatty acids included 10:0, 12:0, 14:0, 17:0, 19:0, 20:0, 22:0, 16:1ω7*c*, 20:1ω9*c*, and a number of small unidentified peaks. The unidentified peaks had retentions that coincided with those of unknown fatty acids (3), but could include nonfatty acids.

The unsaponifiables in vegetable oils, and therefore margarines made with these oils, contain a great many compounds, including sterols, triterpene alcohols, tocopherols and hydrocarbons. Complete analyses, either qualitative or quantitative, were not done. Sterols other than campesterol, stigmasterol and sitosterol are present in many of the margarines, but with few exceptions these three were the major ones. α-, γ- and δ-Tocopherols are sometimes accompanied by small amounts of β-tocopherol and the tocotrienols, but these were either absent or present at very low levels.

Amounts of tocopherols and sterols were highly variable. This may result both from the natural variability of the crude oils and from the effects of processing and storage. Tocopherols may be lost through oxidation and may be removed by deodorization. Sterols are more resistant to oxidation, but also may be lost during deodorization. The effects of processing on sterols have been reviewed extensively by Kochhar (11), who reported that more than 50% of the initial sterol content in a vegetable oil may be removed during processing.

The tocopherols determined were α-, γ- and δ-tocopherol. Very small amounts of β-tocopherol were sometimes present, but this is an exceedingly minor form in these oils. Tocotrienols, if present, probably would be at least partially hydrogenated. α-Tocopherol varied from a high of 24.3 to a low of 0.3 mg/100g. γ-Tocopherol, the most abundant form, with a maximum of 55 mg/100g, has 10% of the activity of α-tocopherol (12). δ-Tocopherol has no vitamin E activity, but may function as an antioxidant in the margarine. Only α- and γ-tocopherols contributed to the vitamin E content, calculated as α-tocopherol equivalents, which ranged from 1.4 to 25.7 mg/100g in the vegetable oil margarines.

Sitosterol was the major sterol in all margarines. In margarines made solely from soybean oil, campesterol and stigmasterol were present in approximately equal amounts; in margarines made solely with corn oil the amounts of campesterol were about twice the amounts of stigmasterol. The margarine made with hydrogenated lard also contained small amounts of plant sterols, in addition to cholesterol.

The collections of single units (Collections I and II) and the nationwide collection of multiple units that were composited (Collection III) had nine identical brands in common. Comparison of the lipid composition of these samples, shown in Table V, may be helpful in assessing the validity of analyses based on single units.

Differences between amounts of most fatty acids were less than 1%, and in only three instances did the difference exceed 3%. The linoleate values for margarines 5, 7 and 9

TABLE IV

**Margarines and Margarine-Like Foods: Isomeric Fatty Acids**

| Source oils[a] | C/CH | C/CH | C/CH | C/CH | C/CH | C/CH | C/CH | C/CH | C/CH | C/CH | C/SBH | C/SBH |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Collection | I | II | II | III | III | III | III | III | III | III | I | I |
| Location[b] | W | L | A | ABCHL | C | ABCHL | AB | ABCHL | C | AH | W | W |
| No. samples | 8 | 1 | 2 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| Units/sample | 1 | 1 | 1 | 10 | 2 | 10 | 4 | 9 | 2 | 4 | 1 | 1 |
| Fat %[c] | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 60 | 60 | 40 | 80 | 80 |
| Type[d] | S | S | S | S | S | T | T | T | T | T | S | S |
| Octadecenoates (normalized wt %, as triglycerides)[e] | | | | | | | | | | | | |
| ω13*t* | 0.10 ±0.05 | trace | ~0.1 | 0.09 | 0.06 | 0.04 | 0.04 | 0.04 | trace | trace | ~0.1 | 0.08 |
| ω12*t* + 10*t* | 3.94 ±0.55 | 2.80 | 4.10 0.22 | 3.54 | 3.32 | 2.16 | 2.14 | 2.07 | 1.83 | 5.11 | 2.98 | 2.87 |
| ω9*t* + 11*t* + 13*c* | 4.45 ±0.70 | 5.41 | 5.09 0.88 | 4.47 | 4.73 | 2.62 | 3.04 | 2.57 | 2.58 | | 4.46 | 3.78 |
| ω8*t* | 5.43 ±0.72 | 5.69 | 6.32 1.04 | 4.74 | 5.17 | 2.76 | 3.31 | 2.74 | 3.03 | 3.02 | 4.78 | 4.64 |
| ω7*t* | 4.15 ±0.49 | 4.71 | 4.96 0.83 | 3.67 | 4.01 | 2.18 | 2.60 | 2.14 | 2.25 | 2.30 | 4.03 | 3.84 |
| ω6*t* | 3.02 ±0.32 | 3.28 | 3.38 0.55 | 2.78 | 2.90 | 1.64 | 1.84 | 1.58 | 1.60 | 1.74 | 2.85 | 2.66 |
| ω?*t* | 0.68 ±0.18 | 0.43 | 1.01 0.48 | 0.89 | 0.86 | 0.57 | 0.52 | 0.53 | 0.49 | 0.23 | 0.56 | 0.57 |
| ω12*c* + 11*c* + 5*t* | 3.57 ±0.35 | 2.98 | 4.13 0.27 | 3.16 | 3.06 | 1.69 | 1.85 | 1.75 | 1.53 | 1.32 | 3.21 | 2.91 |
| ω9*c* + 10*c* | 18.72 ±0.76 | 18.72 | 18.61 1.98 | 18.43 | 18.61 | 21.45 | 22.28 | 21.97 | 22.58 | 21.83 | 18.15 | 18.71 |
| ω8*c* | 1.77 ±0.13 | 1.77 | 2.09 0.36 | 1.58 | 1.63 | | | | | | 1.74 | 1.57 |
| ω7*c* | 1.81 ±0.12 | 1.80 | 2.01 0.27 | 1.66 | 1.69 | 1.24 | 1.40 | 1.25 | 1.28 | 1.27 | 1.69 | 1.65 |
| ω6*c* | 1.85 ±0.51 | 2.33 | 2.24 0.73 | 1.35 | 1.50 | 0.79 | 1.11 | 0.84 | 0.97 | 0.72 | 1.25 | 1.39 |
| ω5*c* | 0.55 ±0.04 | 0.43 | 0.60 0.07 | 0.49 | 0.48 | 0.30 | 0.32 | 0.28 | 0.26 | 0.26 | 0.52 | 0.48 |
| ω?*c* | 0.51 ±0.23 | 0.40 | ND[f] | ND | ND | ND | ND | ND | ND | 0.25 | ND | 0.64 |
| Δ9,12-Octadecadienoates (normalized wt %, as triglycerides) | | | | | | | | | | | | |
| *cc* | 30.09 ±2.61 | 28.10 | 26.85 3.38 | 33.79 | 32.74 | 43.65 | 40.38 | 42.92 | 42.45 | 41.95 | 34.55 | 35.16 |
| *tc* | 0.14 ±0.12 | 0.44 | 0.18 0.02 | 0.14 | 0.08 | 0.20 | 0.28 | 0.23 | 0.10 | 0.83 | 0.35 | 0.13 |
| *ct* | 0.27 ±0.14 | 0.52 | 0.32 0.25 | 0.34 | 0.22 | 0.29 | 0.34 | 0.28 | 0.19 | 0.84 | 0.44 | 0.20 |
| *tt* | trace | 0.25 | 0.12 | 0.10 | 0.06 | trace | trace | 0.06 | 0.06 | 0.18 | 0.04 | trace |

| Source oils | C/SBH | C/SBH | SBH | SBH | SBH | SBH | SBH | SBH | SBH | SBH | SBH | SBH | SBH |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Collection | III | III | I | II | III | III | III | III | III | III | III | III | III |
| Location | ABHL | C | W | C | ABCHL | B | C | A | ABCHL | B | C | A | A |
| No. samples | 1 | 1 | 4 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| Units/sample | 8 | 2 | 1 | 1 | 10 | 2 | 2 | 1 | 10 | 2 | 2 | 2 | 2 |
| Fat % | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 60 | 80 | 80 | 80 | 80 | 60 |
| Type | S | S | S | S | S | S | S | S | T | T | T | T | T |
| Octadecenoates (normalized wt %, as triglycerides) | | | | | | | | | | | | | |
| ω13*t* | 0.06 | 0.04 | 0.10 ±0.02 | 0.08 | 0.08 | 0.06 | 0.10 | ~0.1 | ~0.1 | ~0.1 | 0.06 | ~0.1 | ~0.1 |
| ω12*t* + 10*t* | 2.74 | 2.60 | 3.50 ±0.62 | 3.96 | 3.53 | 9.80 | 3.46 | 3.22 | 1.81 | 2.01 | 2.72 | 2.01 | 1.95 |
| ω9*t* + 11*t* + 13*c* | 4.64 | 4.66 | 4.57 ±1.22 | 13.65 | 13.49 | | 3.61 | 5.14 | 1.94 | 2.79 | 2.47 | 3.25 | 2.94 |
| ω8*t* | 4.72 | 4.63 | 6.73 ±1.98 | | | 8.48 | 7.13 | 8.48 | 3.97 | 4.50 | 5.73 | 4.84 | 4.91 |
| ω7*t* | 3.75 | 3.59 | 5.80 ±1.54 | 7.60 | 7.13 | 6.89 | 5.43 | 6.87 | 2.98 | 3.72 | 4.41 | 3.92 | 4.04 |
| ω6*t* | 2.71 | 2.61 | 3.48 ±0.61 | 3.83 | 3.78 | 4.06 | 3.04 | 3.80 | 1.62 | 2.02 | 2.43 | 2.25 | 2.16 |

TABLE IV (Continued)

| | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| ω?t | 0.53 | 0.46 | 0.93 ±0.08 | 0.91 | 0.79 | 0.77 | 0.67 | 0.70 | 0.35 | 0.41 | 0.62 | 0.44 | 0.45 |
| ω12c + 11c +5t | 2.87 | 2.72 | 4.01 ±0.75 | 4.64 | 4.31 | 3.85 | 3.65 | 3.96 | 1.88 | 2.20 | 3.13 | 2.43 | 2.43 |
| ω9c + 10c | 18.34 | 18.49 | 19.16 ±2.55 | 22.82 | 22.29 | 18.39 | 20.31 | 21.06 | 23.30 | 23.34 | 23.61 | 23.26 | 23.44 |
| ω8c | 1.55 | 1.48 | 2.04 ±0.34 | 2.26 | 2.00 | 2.31 | 1.77 | 2.18 | | | | | |
| ω7c | 1.65 | 1.64 | 2.48 ±0.27 | 2.87 | 2.65 | 2.63 | 2.37 | 2.68 | 1.91 | 2.13 | 2.29 | 2.21 | 2.20 |
| ω6c | 1.27 | 1.34 | 3.83 ±2.10 | 6.53 | 5.90 | 3.90 | 4.23 | 4.86 | 2.68 | 3.03 | 4.02 | 3.05 | 3.46 |
| ω5c | 0.46 | 0.42 | 0.66 ±0.14 | 0.75 | 0.64 | 0.63 | 0.58 | 0.62 | 0.33 | 0.40 | 0.52 | 0.42 | 0.42 |
| ω?c | 0.39 | 0.33 | 0.75 | 0.78 | 0.72 | 0.70 | 0.68 | 0.55 | 0.34 | 0.37 | 0.55 | 0.54 | 0.53 |
| Δ9,12-Octadecadienoates (normalized wt %, as triglycerides) | | | | | | | | | | | | | |
| cc | 33.84 | 32.22 | 16.20 ±9.96 | 6.06 | 7.56 | 8.98 | 17.42 | 9.15 | 30.43 | 26.78 | 23.72 | 26.83 | 25.60 |
| tc | 0.29 | 0.66 | 1.40 ±0.86 | 0.90 | 1.71 | 2.26 | 1.14 | 1.94 | 1.53 | 1.32 | 1.10 | 1.00 | 1.38 |
| ct | 0.50 | 0.83 | 1.18 ±0.63 | 1.20 | 1.25 | 1.92 | 0.89 | 1.58 | 1.27 | 1.38 | 0.95 | 1.14 | 1.24 |
| tt | 0.14 | 0.15 | 0.27 ±0.23 | 0.15 | 0.23 | 0.60 | 0.16 | 0.35 | 0.13 | 0.16 | 0.09 | 0.18 | 0.18 |

| Source oils | SBH | SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH | SB/SBH |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Collection | III | III | I | I | II | II | II | II | II | III | III | III | III |
| Location | C | AC | W | W | H | A | C | A | C | AL | ABCH | ACHL | |
| No. samples | 1 | 1 | 1 | 10 | 1 | 2 | 2 | 1 | 1 | 1 | 1 | 1 | |
| Units/sample | 2 | 4 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 4 | 8 | 6 | |
| Fat % | 60 | 40 | 80 | 80 | 80 | 80 | 80 | 60 | 52 | 80 | 80 | 80 | |
| Type | T | T | B | S | S | S | S | S | S | S | T | T | |
| Octadecenoates (normalized wt %, as triglycerides) | | | | | | | | | | | | | |
| ω13t | ~0.1 | ~0.1 | 0.11 | 0.09 ±0.02 | 0.07 | ~0.1 | ~0.1 | ~0.1 | 0.11 | 0.07 | ~0.1 | 0.05 | |
| ω12t + 10t | 1.88 | 1.68 | 4.05 | 3.52 ±0.48 | 3.30 | 3.29 0.08 | 3.26 0.23 | 2.37 | 3.60 | 2.93 | 1.93 | 1.96 | |
| ω9t + 11t + 13c | 1.97 | 1.92 | 3.65 | 3.96 ±0.34 | 3.13 | 4.79 0.32 | 3.54 0.04 | 3.71 | 3.52 | 4.25 | 1.67 | 1.68 | |
| ω8t | 4.14 | 3.66 | 5.44 | 5.56 ±0.45 | 5.79 | 6.35 0.04 | 5.44 0.08 | 7.93 | 5.86 | 4.92 | 2.94 | 3.12 | |
| ω7t | 3.31 | 2.89 | 4.05 | 4.41 ±0.48 | 4.68 | 5.56 0.18 | 4.32 0.21 | 7.18 | 4.40 | 3.79 | 2.35 | 2.36 | |
| ω6t | 1.68 | 1.58 | 3.01 | 3.03 ±0.18 | 2.80 | 3.43 0.03 | 2.77 0.13 | 3.40 | 2.86 | 2.69 | 1.45 | 1.47 | |
| ω?t | 0.36 | 0.35 | 0.73 | 0.73 ±0.18 | 0.67 | 0.75 0.19 | 0.69 0.07 | 0.64 | 0.74 | 0.55 | 0.39 | 0.41 | |
| ω12c + 11c + 5t | 1.99 | 1.86 | 4.02 | 3.70 ±0.31 | 3.85 | 3.90 0.02 | 3.58 0.20 | 3.64 | 3.73 | 3.06 | 2.00 | 2.08 | |
| ω9c + 10c | 23.13 | 24.04 | 16.72 | 17.18 ±0.42 | 19.67 | 17.10 0.48 | 19.23 0.58 | 22.48 | 18.90 | 17.68 | 22.89 | 23.10 | |
| ω8c | | | 1.76 | 1.82 ±0.08 | | 2.06 0.03 | 1.62 0.11 | | 1.73 | 1.50 | | | |
| ω7c | 1.96 | 1.94 | 2.11 | 2.20 ±0.09 | 2.11 | 2.44 0.02 | 2.20 0.11 | 2.37 | 2.24 | 2.01 | 1.82 | 1.86 | |
| ω6c | 2.74 | 2.85 | 2.16 | 2.25 ±0.36 | 3.26 | 2.74 0.16 | 2.44 0.07 | 5.42 | 2.44 | 1.80 | 1.69 | 1.86 | |
| ω5c | 0.34 | 0.33 | 0.62 | 0.57 ±0.03 | 0.53 | 0.58 0.02 | 0.62 0.05 | 0.50 | 0.62 | 0.49 | 0.33 | 0.35 | |
| ω?c | 0.28 | 0.44 | 0.51 | 0.50 ±0.08 | 0.59 | 0.59 | 0.71 0.17 | 0.54 | 0.40 | 0.62 | 0.27 | 0.26 | |
| Δ9,12-Octadecadienoates (normalized wt %, as triglycerides) | | | | | | | | | | | | | |
| cc | 30.71 | 31.22 | 27.04 | 26.30 ±1.60 | 24.12 | 23.16 1.39 | 26.31 1.90 | 12.43 | 24.61 | 27.44 | 36.69 | 36.32 | |
| tc | 1.12 | 1.17 | 0.25 | 0.22 ±0.09 | 0.72 | 0.35 0.11 | 0.15 0.24 | 2.22 | 0.41 | 0.56 | 0.47 | 0.50 | |
| ct | 1.21 | 1.04 | 0.36 | 0.34 ±0.08 | 0.68 | 0.53 0.27 | 0.26 0.16 | 1.70 | 0.50 | 0.62 | 0.48 | 0.52 | |
| tt | 0.13 | 0.11 | trace | 0.12 ±0.06 | trace | 0.24 | trace | 0.30 | trace | 0.09 | 0.04 | 0.05 | |

TABLE IV (Continued)

| Source oils | SBH/SB | SBH/SB | SB/SBH/CS | SB/SBH/CSH | SB/SBH/CSH | SBH/SB/CSH | SBH/SB CSH | SBH/CSH | SBH/CSH | SBH/CSH | SBH/CSH | SBH/CSH | SBH/CSH |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Collection | I | II | I | III | III | I | III | III | III | III | III | III | III |
| Location | W | L | W | CH | L | W | ABCHL | BCH | ABCL | AH | H | BC | ABCHL |
| No. samples | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| Units/sample | 1 | 1 | 1 | 4 | 2 | 1 | 10 | 6 | 8 | 4 | 2 | 3 | 9 |
| Fat % | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 80 | 60 |
| Type | S | T | S | S | T | S | S | S | T | T | T | T | T |
| Octadecenoates (normalized wt %, as triglycerides) | | | | | | | | | | | | | |
| ω13*t* | 0.24 | ~0.1 | 0.08 | 0.08 | ~0.1 | 0.06 | 0.09 | 0.09 | 0.05 | 0.04 | ~0.1 | 0.09 | 0.05 |
| ω12*t* + 10*t* | 3.25 | 2.30 | 3.54 | 3.13 | 1.93 | 3.60 | 3.58 | 4.00 | 2.15 | 2.56 | 2.04 | 2.88 | 2.21 |
| ω9*t* + 11*t* + 13*c* | 6.40 | 2.23 | 3.32 | 3.13 | 2.86 | 3.59 | 4.17 | 4.44 | 2.26 | 3.04 | 2.86 | 2.50 | 2.53 |
| ω8*t* | 6.08 | 5.46 | 5.29 | 5.30 | 3.74 | 5.81 | 5.90 | 6.06 | 4.41 | 5.27 | 4.99 | 3.72 | 4.30 |
| ω7*t* | 4.76 | 4.18 | 4.23 | 3.76 | 3.08 | 4.43 | 4.48 | 4.82 | 3.28 | 4.14 | 4.20 | 3.12 | 3.35 |
| ω6*t* | 3.23 | 2.11 | 2.83 | 2.55 | 1.93 | 2.83 | 3.02 | 3.24 | 1.94 | 2.32 | 2.13 | 1.99 | 1.96 |
| ω?*t* | 1.02 | 0.53 | 0.68 | 0.59 | 0.90 | 0.53 | 0.62 | 0.70 | 0.47 | 0.47 | 0.42 | 0.49 | 0.44 |
| ω12*c* + 11*c* + 5*t* | 3.42 | 2.82 | 3.77 | 3.07 | 2.09 | 3.49 | 3.57 | 3.99 | 2.18 | 2.69 | 2.42 | 2.62 | 2.28 |
| ω9*c* + 10*c* | 15.34 | 24.80 | 18.23 | 21.81 | 20.31 | 18.58 | 18.47 | 17.73 | 22.63 | 23.08 | 24.71 | 20.88 | 22.78 |
| ω8*c* | 2.01 | | 1.68 | | | 1.67 | 1.68 | 1.93 | | | | | |
| ω7*c* | 2.03 | 2.15 | 2.09 | 2.17 | 1.82 | 2.08 | 2.21 | 2.30 | 2.00 | 2.21 | 2.14 | 1.86 | 1.98 |
| ω6*c* | 1.38 | 5.17 | 2.47 | 2.52 | 1.66 | 2.91 | 2.51 | 2.78 | 2.83 | 3.19 | 3.16 | 2.17 | 2.98 |
| ω5*c* | 0.54 | 0.44 | 0.58 | 0.50 | 0.34 | 0.53 | 0.55 | 0.61 | 0.41 | 0.43 | 0.39 | 0.41 | 0.40 |
| ω?*c* | 0.90 | 0.48 | 0.37 | 0.49 | ND | 0.51 | 0.68 | 0.63 | 0.52 | 0.59 | 0.43 | 0.86 | 0.54 |
| Δ9,12-Octadecadienoates (normalized wt %, as triglycerides) | | | | | | | | | | | | | |
| *cc* | 18.85 | 23.86 | 26.55 | 26.30 | 32.14 | 26.05 | 24.75 | 22.41 | 29.73 | 25.11 | 25.04 | 32.70 | 28.46 |
| *tc* | 0.73 | 0.64 | 0.15 | 0.23 | 0.84 | 0.12 | 0.18 | 0.89 | 1.04 | 1.60 | 1.20 | 0.59 | 1.09 |
| *ct* | 1.2 | 0.52 | 0.31 | 0.38 | 0.86 | 0.25 | 0.41 | 0.88 | 0.94 | 1.45 | 1.27 | 0.82 | 0.95 |
| *tt* | 0.76 | 0.11 | ND | 0.07 | trace | 0.09 | 0.09 | 0.18 | 0.10 | 0.19 | 0.19 | 0.10 | 0.18 |

| Source oil | SBH/CSH | SBH/C/SN/PH | SBH/SB/P | SBH/SB/P | SBH/P SN/C/CS | SBH/PH/P | SBH/CSH/SNH/CH | SBH/CSH/SNH/CH | SF/SB/SBH/CSH | SN/SBH/CSH | SN/SBH/CSH | SN/SBH/CSH | LH |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Collection | III | II | II | II | I | III | III | III | II | I | III | III | II |
| Location | L | L | L | L | W | C | AL | CL | L | W | BC | BC | L |
| No. samples | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| Units/sample | 2 | 1 | 1 | 1 | 1 | 2 | 4 | 3 | 1 | 1 | 4 | 2 | 1 |
| Fat % | 40 | 80 | 80 | 60 | 80 | 40 | 80 | 80 | 80 | 80 | 80 | 80 | 80 |
| Type | T | T | S | S | S | T | S | T | S | S | S | T | S |
| Octadecenoates (normalized wt %, as triglycerides) | | | | | | | | | | | | | |
| ω13*t* | 0.05 | 0.13 | 0.14 | 0.08 | 0.26 | 0.12 | 0.43 | 0.26 | 0.07 | 0.38 | 0.10 | 0.12 | ND |
| ω12*t* + 10*t* | 6.43 | 2.84 | 4.16 | 4.07 | 4.65 | 3.08 | 4.74 | 2.93 | 3.20 | 4.22 | 4.33 | 3.39 | 0.46 |
| ω9*t* + 11*t* + 13*c* | | 4.39 | 3.73 | 3.63 | 4.61 | 3.28 | 5.41 | 4.10 | 3.45 | 2.28 | 4.74 | 2.61 | 1.54 |
| ω8*t* | 4.75 | 4.34 | 4.95 | 5.14 | 5.20 | 4.20 | 5.57 | 4.24 | 3.98 | 2.55 | 4.91 | 3.28 | 1.64 |
| ω7*t* | 3.96 | 3.75 | 4.15 | 3.95 | 4.58 | 3.47 | 4.93 | 3.74 | 3.41 | 2.35 | 4.16 | 2.64 | ND |
| ω6*t* | 1.93 | 2.20 | 2.99 | 2.79 | 3.13 | 2.27 | 3.22 | 2.16 | 2.51 | 2.08 | 3.14 | 2.08 | 0.30 |
| ω?*t* | 0.50 | 0.58 | 0.74 | 0.73 | 0.94 | 0.60 | 1.20 | 0.85 | 0.50 | 0.95 | 0.67 | 0.53 | ND |
| ω12*c* + 11*c* + 5*t* | 1.72 | 2.64 | 4.38 | 4.22 | 4.01 | 2.92 | 4.50 | 2.87 | 3.28 | 3.62 | 3.87 | 2.94 | 0.32 |
| ω9*c* + 10*c* | 20.80 | 17.57 | 16.45 | 17.11 | 15.63 | 20.50 | 14.94 | 19.49 | 13.47 | 13.98 | 12.40 | 14.47 | 37.76 |
| ω8*c* | | 1.25 | 1.81 | 1.81 | 1.86 | | 2.02 | | 1.53 | 1.32 | 1.87 | 1.22 | ND |
| ω7*c* | 1.69 | 1.67 | 2.04 | 2.09 | 1.98 | 1.95 | 1.97 | 1.73 | 1.74 | 1.20 | 1.75 | 1.41 | 2.92 |
| ω6*c* | 1.03 | 1.34 | 1.92 | 1.92 | 2.02 | 2.44 | 1.70 | 1.74 | 1.13 | 0.70 | 1.22 | 1.00 | 0.38 |
| ω5*c* | 0.23 | 0.37 | 0.64 | 0.60 | 0.60 | 0.46 | 0.60 | 0.40 | 0.46 | 0.49 | 0.59 | 0.45 | 0.29 |
| ω?*c* | 0.53 | 0.56 | 0.61 | 0.58 | 0.86 | 0.69 | 0.88 | 0.64 | ND | 0.78 | 0.74 | 0.76 | ND |
| Δ9,12-Octadecadienoates (normalized wt %, as triglycerides) | | | | | | | | | | | | | |
| *cc* | 19.46 | 21.34 | 28.16 | 28.19 | 17.74 | 26.21 | 14.37 | 20.52 | 40.35 | 43.57 | 35.49 | 46.39 | 8.94 |
| *tc* | 4.44 | 4.78 | 0.20 | ND | 3.49 | 2.44 | 3.36 | 3.96 | 0.20 | 0.50 | 0.69 | 0.26 | ND |
| *ct* | 4.64 | 5.31 | 0.34 | 0.17 | 3.46 | 2.31 | 3.76 | 4.27 | 0.39 | 0.80 | 0.82 | 0.42 | ND |
| *tt* | 1.23 | 1.47 | trace | trace | 0.94 | 0.46 | 1.28 | 1.33 | trace | trace | 0.28 | trace | ND |

[a]C = corn oil, CH = hydrogenated corn oil, CS = cottonseed oil, CSH = hydrogenated cottonseed oil, L = lard, LH = hydrogenated lard, P = palm oil, PH = hydrogenated palm oil, SB = soybean oil, SBH = hydrogenated soybean oil, SF = safflower oil, SN = sunflower oil and SNH = hydrogenated sunflower oil.

[b]A = Atlanta, B = Boston, C = Chicago, H = Houston, L = Los Angeles and W = Washington-Baltimore.

[c]Label information.

[d]S = stick, T = tub and B = block.

[e]Means for two samples are followed by the range. Means for more than two samples are followed by the standard deviation.

[f]ND = not detected.

TABLE V

**Lipids in Margarines: Comparison of Single and Composite Samples**

| | Margarine 1 | | Margarine 2 | | Margarine 3 | | Margarine 4 | | Margarine 5 | | Margarine 6 | | Margarine 7 | | Margarine 8 | | Margarine 9 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Single | Comp. | Single | Comp. | Single | Comp. | Single | Comp. | Single | Comp. | Single | Comp. | Single | Comp. | Single | Comp. | Single | Comp. |
| Fatty acids (normalized wt %): | | | | | | | | | | | | | | | | | | |
| 16:0 | 10.46 | 10.62 | 10.73 | 10.66 | 10.76 | 10.88 | 10.98 | 11.04 | 10.20 | 10.55 | 8.85 | 9.21 | 10.30 | 10.59 | 10.47 | 10.51 | 9.79 | 10.48 |
| 18:0 | 7.10 | 6.44 | 6.17 | 6.38 | 6.39 | 7.08 | 6.95 | 7.38 | 7.03 | 7.71 | 7.34 | 6.69 | 6.94 | 6.10 | 8.00 | 7.84 | 6.93 | 8.41 |
| 16:1ω7*c* | 0.04 | 0.06 | ND[a] | 0.06 | 0.05 | 0.05 | 0.04 | 0.04 | 0.03 | 0.03 | ND | ND | 0.04 | 0.07 | 0.03 | 0.04 | 0.04 | ND |
| 18:1ω13*t* | 0.09 | 0.09 | ND | 0.06 | 0.08 | 0.04 | 0.08 | 0.08 | 0.12 | 0.06 | 0.38 | 0.10 | 0.09 | 0.04 | 0.06 | 0.09 | 0.08 | 0.10 |
| 18:1ω12*t*+10*t* | 3.61 | 3.54 | 2.98 | 2.74 | 2.87 | 2.60 | 3.42 | 3.53 | 4.19 | – | 4.22 | 4.33 | 4.26 | 2.14 | 3.60 | 3.58 | 3.96 | 3.46 |
| 18:1ω9*t*+11*t*+13*c* | 4.28 | 4.47 | 4.46 | 4.64 | 3.78 | 4.66 | – | – | 3.87 | – | 2.28 | 4.47 | 4.61 | 3.04 | 3.59 | 4.17 | – | 3.61 |
| 18:1ω12*t*+10*t*+9*t*+11*t*+13*c* | – | – | – | – | – | – | – | – | – | 9.80 | – | – | – | – | – | – | – | – |
| 18:1ω9*t*+11*t*+13*c*+8*t* | – | – | – | – | – | – | 13.52 | 13.49 | – | – | – | – | – | – | – | – | 13.65 | – |
| 18:1ω8*t* | 5.01 | 4.74 | 4.78 | 4.72 | 4.64 | 4.63 | – | – | 8.06 | 8.48 | 2.55 | 4.91 | 6.43 | 3.31 | 5.81 | 5.90 | – | 7.13 |
| 18:1ω7*t* | 3.69 | 3.67 | 4.03 | 3.75 | 3.84 | 3.59 | 7.26 | 7.13 | 6.18 | 6.89 | 2.35 | 4.16 | 4.67 | 2.60 | 4.43 | 4.48 | 7.60 | 5.43 |
| 18:1ω6*t* | 2.78 | 2.78 | 2.85 | 2.71 | 2.66 | 2.61 | 3.87 | 3.78 | 3.69 | 4.06 | 2.08 | 3.14 | 3.29 | 1.84 | 2.83 | 3.02 | 3.83 | 3.04 |
| 18:1ω?*t* | 0.47 | 0.89 | 0.56 | 0.53 | 0.57 | 0.46 | 0.86 | 0.79 | 0.88 | 0.77 | 0.95 | 0.67 | 0.61 | 0.52 | 0.53 | 0.62 | 0.91 | 0.67 |
| 18:1ω12*c*+11*c*+5*t* | 3.20 | 3.16 | 3.21 | 2.87 | 2.91 | 2.72 | 4.35 | 4.31 | 4.64 | 3.85 | 3.62 | 3.87 | 3.93 | 1.85 | 3.49 | 3.57 | 4.64 | 3.65 |
| 18:1ω9*c*+10*c* | 18:45 | 18.43 | 18.15 | 18.34 | 18.71 | 18.49 | 22.47 | 22.29 | 19.90 | 18.39 | 13.98 | 12.40 | 18.62 | – | 18.58 | 18.47 | 22.82 | 20.31 |
| 18:1ω9*c*+10*c*+8*c* | – | – | – | – | – | – | – | – | – | – | – | – | – | 22.28 | – | – | – | – |
| 18:1ω8*c* | 1.61 | 1.58 | 1.74 | 1.55 | 1.57 | 1.48 | 2.06 | 2.00 | 2.16 | 2.31 | 1.32 | 1.87 | 1.95 | – | 1.67 | 1.68 | 2.26 | 1.77 |
| 18:1ω7*c* | 1.64 | 1.66 | 1.69 | 1.65 | 1.65 | 1.64 | 2.68 | 2.65 | 2.56 | 2.63 | 1.20 | 1.75 | 2.00 | 1.40 | 2.08 | 2.21 | 2.87 | 2.37 |
| 18:1ω6*c* | 1.65 | 1.35 | 1.25 | 1.27 | 1.39 | 1.34 | 6.17 | 5.90 | 4.98 | 3.90 | 0.70 | 1.22 | 2.27 | 1.11 | 2.91 | 2.51 | 6.53 | 4.23 |
| 18:1ω5*c* | 0.51 | 0.49 | 0.52 | 0.46 | 0.48 | 0.42 | 0.67 | 0.64 | 0.71 | 0.63 | 0.49 | 0.59 | 0.61 | 0.32 | 0.53 | 0.55 | 0.75 | 0.58 |
| 18:1ω?*c* | 0.33 | ND | ND | 0.39 | 0.64 | 0.33 | 0.64 | 0.72 | 0.74 | 0.70 | 0.78 | 0.74 | 0.48 | ND | 0.51 | 0.68 | 0.78 | 0.68 |
| 18:2ω6*cc* | 33.49 | 33.79 | 34.55 | 33.84 | 35.16 | 32.22 | 7.58 | 7.56 | 13.89 | 8.98 | 43.57 | 35.49 | 26.59 | 40.38 | 26.05 | 24.75 | 6.06 | 17.42 |
| 18:2ω6*ct* | 0.14 | 0.34 | 0.44 | 0.50 | 0.20 | 0.83 | 1.15 | 1.25 | 1.24 | 1.92 | 0.80 | 0.82 | 0.32 | 0.34 | 0.25 | 0.41 | 1.20 | 0.89 |
| 18:2ω6*tc* | 0.07 | 0.14 | 0.35 | 0.29 | 0.13 | 0.66 | 1.67 | 1.71 | 1.31 | 2.26 | 0.50 | 0.69 | 0.22 | 0.28 | 0.12 | 0.18 | 0.90 | 1.14 |
| 18:2ω6*tt* | ND | 0.10 | 0.04 | 0.14 | ND | 0.15 | 0.21 | 0.23 | 0.16 | 0.60 | ND | 0.28 | ND | ND | 0.09 | 0.09 | 0.15 | 0.16 |
| 18:3ω3*ccc* | 0.50 | 0.69 | 0.45 | 0.41 | 0.51 | 0.62 | 0.16 | 0.18 | 0.74 | 0.25 | 0.71 | 0.26 | 0.78 | 0.76 | 3.01 | 2.81 | 0.30 | 2.04 |
| Other fatty acids | 0.87 | 0.93 | 1.03 | 1.96 | 1.00 | 2.50 | 3.15 | 3.28 | 2.72 | 5.21 | 1.19 | 2.05 | 0.96 | 1.00 | 1.94 | 1.81 | 3.93 | 2.43 |
| Tocopherols (mg/100g): | | | | | | | | | | | | | | | | | | |
| α-Tocopherol | 13.5 | 15.2 | 12.8 | 9.8 | 14.1 | 0.3 | 4.6 | 5.4 | 5.7 | 7.2 | 24.3 | 5.6 | [b] | | 8.7 | 5.8 | | |
| γ-Tocopherol | 36.2 | 49.7 | 36.8 | 43.6 | 52.1 | 17.2 | 38.4 | 29.1 | 41.1 | 48.0 | 13.8 | 9.8 | | | 49.1 | 44.0 | | |
| δ-Tocopherol | 2.4 | 0.5 | 7.4 | 7.1 | 8.7 | 3.3 | 12.7 | 8.2 | 14.7 | 13.1 | 5.9 | 2.5 | | | 17.1 | 13.4 | | |
| Sterols (mg/100g): | | | | | | | | | | | | | | | | | | |
| Campesterol | 100.0 | 87.0 | 74.2 | 76.5 | 78.8 | 70.0 | 29.3 | 25.7 | 39.0 | 35.5 | 28.1 | 33.0 | | | 51.1 | 32.0 | | |
| Stigmasterol | 46.6 | 43.6 | 38.5 | 37.8 | 39.0 | 36.4 | 30.0 | 26.6 | 33.3 | 32.9 | 26.6 | 23.0 | | | 39.3 | 34.4 | | |
| Sitosterol | 347.7 | 363.1 | 280.0 | 266.6 | 279.2 | 291.0 | 92.2 | 80.8 | 107.3 | 93.4 | 141.3 | 137.7 | | | 142.6 | 112.0 | | |

[a]ND = not detected.

[b]Blanks indicate insufficient data for comparison.

differed by 4.91, 13.79 and 11.36%, respectively. Tocopherol contents were much more variable, especially margarines 3 and 6. Sterols were more consistent, and varied less than 18% (relative). From these limited data, it appears that the compositions of single samples of margarine are representative in general of the lipids in specific brands, and that linoleate and the tocopherols will be most variable.

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# Analysis of Lipids by High Performance Liquid Chromatography—Chemical Ionization Mass Spectrometry

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## ABSTRACT

The apparatus and techniques for the analysis of lipids by high performance liquid chromatography in conjunction with computer controlled chemical ionization-mass spectrometry are presented. The interfaces between the liquid chromatograph and the mass spectrometer and between the computer and the mass spectrometer are described. The identification of lipid classes separated by adsorption chromatography and molecular species by reverse phase chromatography also is presented. The techniques are applied to reference compounds and to examples of plant and animal lipids.

## INTRODUCTION

The inherent difficulties of combining high performance liquid chromatography (HPLC) with conventional mass spectrometry have been elaborated by McFadden and coworkers (1,2). Present methods generally are limited to the narrow band of compounds that are too unstable for analysis by gas liquid chromatography (GLC), but are sufficiently stable or volatile to be introduced directly into the mass spectrometer. Kuksis and his colleagues (3-5) have successfully applied HPLC with chemical ionization mass spectrometry (CIMS) to the molecular species of triglycerides by a Direct Liquid Inlet (DLI) technique in which the species are simultaneously separated and analyzed.

Jungalwala et al. (6) reported the use of the Finnegan moving belt system for the analysis of spingolipid bases after hydrolysis and derivatization. More recently (7), these investigators applied this system to the analysis of phospholipids which they found could be separated by HPLC by the use of a solvent system containing ammonium hydroxide. The use of ammonium hydroxide in solvent systems for the separation of phospholipids was first used by Rouser et al. (8) in the gravity flow chromatography of lipid classes. We reported the use of $NH_4OH$-methanol in a gradient solvent system in the HPLC of plant and animal glycolipids and phospholipids about 1973 (9,10). More recently, we reported similar systems for the separation of lipofusion substances, neutral and polar lipid classes (11) and in our method for the quantitative analysis of the lipid classes (12).

Patton et al. (13) and Smith and Jungalwala (14) developed methods for the separation of phospholipids by HPLC using conventional UV detectors, but methods based on the use of photometric detectors are very difficult to employ for quantitative analysis as illustrated in the work of Perrin and Naudet et al. (15).

In previous work (16-18), we developed a simple interface for the purpose of combining HPLC with CIMS for the analysis of lipids based on their continuous conversion to volatile products by reduction with hydrogen prior to their introduction into the mass spectrometer. The operation of the interface was demonstrated on the lipid classes and related compounds by CIMS with methane as the reagent gas (16,17). Hydrogen was introduced into the reactor in this system to convert the acyl groups of the lipids to hydrocarbons. However, some of the double bonds in the acyl groups were hydrogenated, voiding identification of the fatty acid constituents. Hence we turned to the use of an inert gas in place of hydrogen and developed a rapid method (ca. 30 seconds) for the quantitative analysis of the 19 most common fatty acids found in plant and animal tissues directly by CIMS (19). With an inert carrier gas in our system, the acyl groups of the lipid classes are split from the backbone structures and identified as the $[RCOOH + 1]^+$ and $[RCO]^+$ ions. Since the products of the backbone structures of the lipid classes are produced by thermal degradation, their formation does not appear to be affected by the change to an inert gas. The application of this technique to the lipid classes and molecular species of triglycerides is described here.

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