a large extent in the industry—which circumstance would seem to be a reason to consider that the grade scale corresponding to the use of the proposed earth would classify refined oils more reliably for commercial utilization than does the current official (English) earth.

In considering the variations in the equivalent dosage found on different samples of refined oil, it must be emphasized that different kinds of bleaching earths act differently in bleaching vegetable oils. The pigment systems of the oils are not simple, and experience has shown that fuller's earths remove various pigments selectively to some extent; also the selectivity of one earth may be quite different from that of another earth of a different type. We therefore have to expect some appreciable variation in the percentage of one earth necessary to bleach different oils, even of the same kind, to the same color as another earth of a different type—especially one which is far more active, or far less active. An outstanding piece of evidence of this is the circumstance that a very much smaller equivalent dosage for the proposed earth was found applying to soybean oil than was found for cottonseed oil.

Accordingly, in reaching the decision to change the type of its official bleaching earth from that of English earth having relatively low activity, to a domestic earth comparable in a degree to domestic earths being used in commercial bleaching, it should have been understood that even a fair degree of uniformity of equivalent dosages between the two earths, when compared on different oils, could not necessarily be expected; and had such uniformity been set up as a prerequisite for making the change to a domestic natural earth, we believe that such a requirement should have been sufficient to rule out the change in the first place.

Although somewhat disappointed at first, after studying the matter and taking into consideration the differential selective action, the committee considered that the oil-to-oil uniformity of the proposed earth in relation to the official earth was as good as could be expected when comparing two entirely different types of earth—two which differ in activity as much as do these. Once the change is made, if it is made, there seems adequate reason to believe that the new earth will perform in the desired function as well as the English earth. About the same percentage of the refined cottonseed oil made should still be graded as bleachable as before, although a few oils here and there, a small percentage of the whole, will undoubtedly grade bleachable with the new earth, when it would have failed to do so with the old English earth—and *vice versa*.

In such cases, if one asks which earth gives the "correct" indication, the committee believes that there is just as much reason to favor the indication of the proposed, more active earth than there is to let the indication of the old (English) official earth determine the grade. In fact, since the proposed earth is far more similar to the earths used by many refiners in commercial practice, this should perhaps make it the preferred one in cases of conflicts of grade indication. But it is significant to recall that with only one earth to use, there could be no such thing as conflict between the two.

In this last connection we might also consider that if the industry had been using an official earth similar to the proposed earth during the past 25-odd years and for some reason it were considered desirable or necessary to change over to a "new" official earth similar to the currently-official earth, exactly the same discrepancies would be encountered, and one believes that such objections as might be voiced to making the change would probably be much louder than we are likely to hear in the present instance, if this change is approved—especially as it would represent a considerable departure from commercial practice.

EGBERT FREYER.

February 12, 1954

Fatty Acid Contents of Certain Processed Foods[1]

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IN recent years the role of fat in the diet has been found to be much more significant than was previously realized. In particular, such unsaturated constituents of fat as linoleic and arachidonic acids have been found to be essential to the health of laboratory animals (5, 6, 11). While there is less evidence concerning the role of these fatty acids in human nutrition, a deficiency of these acids in humans may be associated with certain forms of eczema and with other symptoms comparable to the first clinical manifestations of essential fatty acid deficiency in animals (4, 10).

Because these acids are highly unsaturated and therefore subject to possible alteration or destruction by the oxygen of the air, chemicals, or heat, the effects of ordinary processing and cooking of fat-containing foods are of significance in human nutrition. Most fat analyses reported in the literature have been performed on unprocessed, raw samples (12). Recently however studies of the effect of cooking on the essential fatty acids of meats and poultry (8), and of baked and fried foods (7) have been made. In the present work the fatty acid contents of certain foods which had undergone various types of processing are reported. The foods studied include meats cured by several processes, American and Swiss type of process cheeses, and vegetable oils altered by hydrogenation.

Samples

Fresh, representative samples of bacon, ham, luncheon meat, and frankfurters were obtained directly from a processing plant. The luncheon meat and frankfurters contained both pork and beef.

Samples of Swiss- and American-type processed cheese, together with corresponding samples of "cheese raw material," taken just before addition of the emulsifiers to the cheese products, were obtained from a manufacturer.

Oleomargarine was purchased over the counter, and a sample of bakers' shortening was obtained from a distributor.

General Procedure and Results

Two sets of data were obtained for each sample analyzed:

1. A small portion of fat was converted to fatty acids (see following section), and these were analyzed spectrophotometrically for polyunsaturated components.

2. A second portion of fat was converted to methyl esters by methanolysis except in the case of the cheese fats. By use of special techniques the fatty acids of the latter were esterified *in toto* by conventional means without the conventional prior steam distillation of volatile components. In all cases the total methyl esters were fractionated in one distillation without separation of saturated and unsaturated portions.

Individual distillation cuts were combined into fractions representing acids having the same number of carbon atoms or into transition fractions representing acids of two different carbon chain-lengths. Each fraction was then analyzed chemically and spectrophotometrically to determine both saturated

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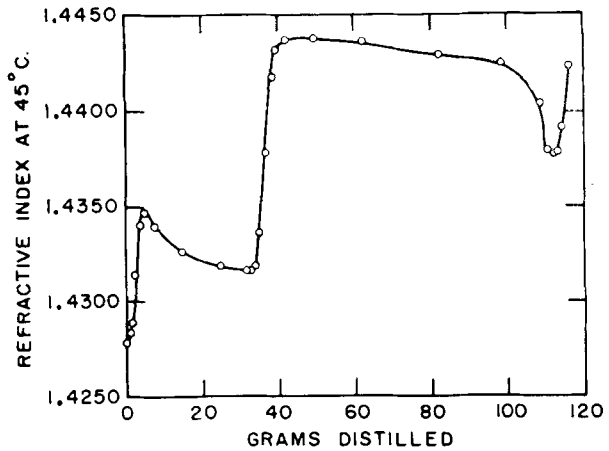

FIG. 1. Distillation of methyl esters prepared from luncheon meat fat.

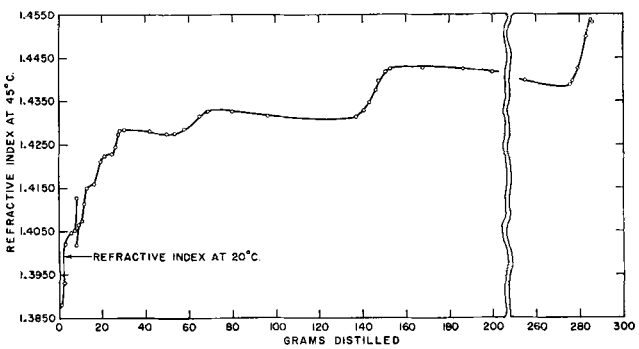

FIG. 2. Distillation of methyl esters prepared from American cheese fat.

and unsaturated fatty acid components. Typical distillation curves are illustrated in Figures 1 and 2.

Comparison of polyunsaturated components determined on the total fatty acids (Procedure 1) and in the methyl esters, calculated to the fatty acid basis, (Procedure 2) are presented in Table I. The percentages of total fatty acids of each chain-length obtained by analysis of the methyl esters (Procedure 2) are presented in Tables II, III, and IV for cheeses, cured meats, and hydrogenated oils, respectively.

The weight percentage of the fatty acid constituents in each food are presented in Table V.

Methods

Care was taken to prevent undue exposure of the samples to heat, light, and oxygen of the air. Whereever necessary, procedures were carried out in a nitrogen atmosphere or in the presence of a solvent which provided a protective blanket of vapor.

Extraction of Fat. Fat was extracted from the meat products and the "cheese raw materials" by blending 100-g. batches of roughly cut material with 200 ml. chloroform in an Oster blender for one-minute periods. The blended mixture was filtered with suction and the filtrate dried over magnesium sulfate. Oleomargarine, the only sample requiring special treatment, was heated to boiling with twice its weight of chloroform. The chloroform layer was separated while hot from the emulsified aqueous layer, which was re-extracted with two more small portions of hot chloroform. The combined chloroform layers were washed with small portions of hot water and dried over magnesium sulfate.

Fat and Moisture. Determinations were made on all samples in accordance with the appropriate A.O.A.C. methods (2). The bone and rind were removed from the ham, and the casing from the luncheon meat. The fat from the "cheese raw material" was analyzed; the results were calculated to the basis of the processed cheese, using the percentage of fat and

TABLE I

Comparison of Polyunsaturated Acids Determined on Total Fatty Acids and on the Distillation Fractions of the Methyl Esters of Food Fats (Converted to the Fatty Acid Basis)
(weight percentage acids in total fatty acids)

	Diene	Conj. Diene	Triene	Conj. Triene	Tetraene	Conj. Tetraene
Bacon						
Total Acids	11.3	0.22	0.56	Trace	0.51	Trace
Distillation Fractions	10.0(1)	0.26	0.57	0.01	0.42	Trace
Cheese, American						
Total Acids	1.39	1.58	0.61	0.04	0.46	0.01
Distillation Fractions	1.48	1.08	0.88	0.01	0.18	0.00
Cheese, Swiss						
Total Acids	1.43	1.60	0.78	0.03	0.40	0.00
Distillation Fractions	1.07	1.30	0.76	Trace	0.13	0.00
Frankfurters						
Total Acids	8.53	0.28	0.60	Trace	0.40	Trace
Distillation Fractions	8.01	0.30	0.47	0.01	0.28	Trace
Ham						
Total Acids	10.2	0.26	0.60	0.00	0.47	0.00
Distillation Fractions	9.62	0.36	0.72	0.01	0.43	Trace
Luncheon Meat						
Total Acids	7.40	0.47	0.51	0.01	0.44	0.00
Distillation Fractions	7.55	0.43	0.19	0.01	0.42	Trace
"MFB" Shortening						
Total Acids	5.06	0.17	0.04	Trace	0.00	0.00
Distillation Fractions	4.74	0.23	0.07	0.01	Trace	0.00
Oleomargarine						
Total Acids	14.3	0.47	0.00	0.06	Trace	0.00
Distillation Fractions	13.3(8)	0.47	0.05	0.02	Trace	0.00

NOTE: Figures which are not significant have been placed in parentheses.

TABLE II

Composition of Fatty Acids of Cheeses
(weight percentage acids in total fatty acids)

	American Cheese (Processed)	Swiss Cheese (Processed)
C_4 Butyric	2.72	2.36
C_6 Caproic	1.76	2.17
C_8 Caprylic	1.18	1.11
Octenoic	0.01	Trace
C_{10} Capric	2.32	2.42
Decenoic	0.10	0.10
C_{12} Lauric	2.48	2.49
Dodecenoic	0.25	0.24
C_{14} Myristic	10.5(5)	10.8
Tetradecenoic	0.78	0.88
C_{16} Palmitic	26.2	25.6
Palmitoleic	2.21	2.11
C_{18} Stearic	11.6(4)	12.6
Oleic	30.6(8)	31.8
Diene		
Linoleic	1.48	1.06
Conjugated	0.27	1.18
Triene		
Linolenic	0.80	0.74
Conjugated	0.01	Trace
C_{20} Saturated	1.75	0.58
Monoene	1.74	1.43
Diene		
Conjugated	0.81	0.12
Triene		
Non-conjugated	0.08	0.02
Tetraene		
Arachidonic	0.18	0.13

NOTE: Figures which are not significant have been placed in parentheses.

TABLE III

Composition of Fatty Acids of Cured Meats
(weight percentage acids in total fatty acids)

Acids	Bacon	Frankfurters	Ham	Luncheon Meat
C_{12} Lauric	0.10	0.10	0.47	0.22
C_{14} Myristic	1.03	1.08	1.75	0.72
Tetradecenoic			0.08	
C_{16} Palmitic	21.7	25.0	21.7(2)	25.4
Palmitoleic	2.57	3.34	2.71	3.51
C_{18} Stearic	9.72	11.5	13.7(2)	9.3(4)
Oleic	50.4	47.4	44.4(1)	48.0
Diene				
Linoleic	9.56	7.73	9.07	7.23
Conjugated	0.10	0.18	0.16	0.12
Triene				
Linolenic	0.45	0.46	0.57	0.11
C_{20} Saturated	0.66	0.28	0.40	1.24
Monoene	2.59	2.08	3.60	2.97
Diene				
Non-conjugated	0.45	0.28	0.55	0.32
Conjugated	0.16	0.12	0.20	0.31
Triene				
Non-conjugated	0.12	0.11	0.15	0.08
Conjugated	0.01	0.01	0.01	0.01
Tetraene				
Arachidonic	0.42	0.28	0.43	0.42
Conjugated	Trace	Trace	Trace	Trace

NOTE: Figures which are not significant have been placed in parentheses.

TABLE IV

Composition of Fatty Acids of Hydrogenated Oils
(weight percentage acids in total fatty acids)

	"MFB" Shortening	Oleomargarine
C_{14} Myristic	1.04	0.54
C_{16} Palmitic	13.8	22.0
Palmitoleic	0.21	2.09
C_{18} Stearic	6.99	2.77
Oleic	40.2	34.1
Iso-oleic	20.9	17.5
Diene		
Linoleic	4.7	13.36
Iso-linoleic	5.41	3.66
Conjugated	0.02	0.22
Triene		
Linolenic	0.04	0.01
Conjugated	Trace	Trace
C_{20} Saturated	3.25	1.87
Monoene	1.84	0.94
Iso-monoene	1.08	0.44
Diene		
Non-conjugated	0.04	0.08
Iso-diene	0.28	0.11
Conjugated	0.21	0.25
Triene		
Non-conjugated	0.03	0.04
Conjugated	0.01	0.02

moisture obtained on the finished product. This procedure was followed because separation of fat from finished processed cheese, containing emulsifiers, requires chemical treatment which may alter the fat. In contrast, fat may be obtained from "cheese raw material," to which emulsifiers have not yet been added, by simple extraction. The procedure of going from cheese raw material to finished product, involving 5 or 6 minutes' blending at approximately 65°C., was considered not to have any effect on the fat.

Preparation of Fatty Acids. A portion of each fat solution was stripped of solvent under nitrogen with the use of reduced pressure. Fatty acids free from non-saponifiable materials were prepared from the fat by the method of Lundberg (15). Special care was taken to insure quantitative recovery of low molecular weight constituents in the case of cheese fats.

Preparation of Methyl Esters of Meat Fats and Hydrogenated Oils. The methanolysis procedure of Kurz (13) was used except that the fat was in chloroform rather than ethereal solution. The percentage of fat in an aliquot of the chloroform extract obtained from a sample was determined to insure correct proportions of the reactants; free fatty acids in the fat solution were determined (they were always negligible) to ascertain the essential neutrality of the solutions. Finally the reaction mixtures obtained were, after being worked up, subjected to a second methanolysis to convert traces of unreacted glycerides to esters.

Preparation of Methyl Esters of Cheese Fats. The fat, freed from solvent, was converted to fatty acids in 50-g. batches by Lundberg's method (15) except that sulfuric rather than hydrochloric acid was used to liberate the acids after saponification and removal of the alcohol. The acids were extracted with ether, dried over magnesium sulfate, and the ether removed. The acids, *in toto*, were then esterified with purified methanol, using sulfuric acid as the catalyst, and worked up by essentially conventional procedures: The catalyst was nearly neutralized, most of the methanol distilled off, and the esters, taken up in ether, were washed with water, sodium carbonate, and water again. The ethereal solution was dried over magnesium sulfate and stripped of solvent. Unesterified acids were recovered from the alkaline wash, esterified, and added to the main product.

Special care however was taken in this procedure to prevent loss of soluble and volatile low-molecular weight components. All aqueous washes were exhaustively extracted with ether, and the extracts added to the main solutions; solvents (ether and excess methanol) were stripped using an efficient column. Finally the aqueous washings were steam-distilled, the distillate titrated with standard base, and the results calculated as butyric acid. Better than 96% yield of esters could thus be obtained. No separation of "saturated" and "unsaturated" portions of the esters was made in any instance.

TABLE V

Weight Percentage Fatty Acids in Foods

	Percentage Moisture in Food	Percentage Fat in Food [4]	Saturated						Unsaturated				
			C_{12}	C_{14}	C_{16}	C_{18}	C_{20} and Above	Total Saturated	Monoene	Diene	Triene	Tetraene	Total Unsaturated
Bacon	23.75	59.45	0.06	0.59	12.3	5.5	0.37	18.8(2)	31.63	5.82	0.32	0.24	38.0(1)
Cheese, American [1]	40.39	30.75	0.72	3.1	7.6	3.4	0.51	17.6(4)	10.37	0.75	0.25	0.05	11.4(2)
Cheese, Swiss [2]	42.39	26.14	0.61	2.7	6.3	3.1	0.14	14.8(3)	9.08	0.58	0.18	0.03	9.8(7)
Frankfurters	48.10	33.05	0.03	0.34	7.9	3.6	0.09	11.9(6)	16.72	2.64	0.19	0.09	19.6(4)
Ham	56.25	25.11	0.11	0.42	5.2	3.3	0.09	9.1(2)	12.23	2.40	0.16	0.10	14.8(9)
Luncheon Meat	57.50	20.37	0.04	0.14	4.95	1.82	0.24	7.1(9)	10.62	1.55	0.03	0.08	12.2(8)
"MFB" Shortening	0.07 [3]	99.93	.....	0.99	13.2	6.67	3.11	23.9(7)	61.29	10.19	0.08	.....	71.5(6)
Oleomargarine	14.04	80.64	.....	0.42	17.0	2.14	1.44	21.0(0)	42.47	13.57	0.05	.....	66.0(9)

[1] In addition to acids tabulated, contains 0.79% C_4, 0.51% C_6, 0.34% C_8, and 0.67% C_{10} saturated acids.
[2] In addition to acids tabulated, contains 0.58% C_4, 0.53% C_6, 0.27% C_8, and 0.60% C_{10} saturated acids.
[3] Assumed.
[4] In converting fatty acids in fats to fatty acids in foods, the non-saponifiable matter in the fat was ignored for purposes of calculation.
NOTE: Figures which are not significant have been placed in parentheses.

Distillation of Methyl Esters. Distillations were made through a jacketed, electrically-heated column with a total condensation, variable take-off head. The column, constructed of 14-mm. Pyrex tubing, was packed with single-turn 1/8-inch glass helices for a length of 32 inches. The 500-ml. distilling flask was heated by an oil bath. Bumping was prevented by use of a magnetic stirrer.

In most cases 120-g. samples were taken. Distillations were carried out at 5.0 mm. until the end of the C_{18} plateau was reached, when the pressure was lowered to 1 or 2 mm. and distillation continued until the C_{20} range was attained. About 25 cuts were taken in a continuous distillation which usually took some 24 hours to complete.

In the case of the cheese fat methyl esters 300-g. samples were taken to insure sufficiently large amounts of each component for satisfactory analysis. Distillation was started at atmospheric pressure, and the pressure was gradually diminished, being adjusted to 5.0 mm. by the time the C_{14} range was reached. Sixty-five to 70 cuts were taken in distillations lasting 55 to 60 hours. The refractive index was determined on each cut after weighing and plotted against total grams of esters distilled.

Treatment of Distillation Fractions. From consideration of the distillation curves, individual cuts were combined quantitatively into 5 or 6 fractions for each sample or, in the case of the cheese fat esters, into 12 or 14 fractions, representing acids of the same carbon chain length or transition fractions containing acids of two chain lengths.

The final fraction in each case contained the distillation residue and was assumed to include all the non-saponifiable material originally present in the ester sample. This fraction was therefore converted to non-saponifiable-free fatty acids by Lundberg's procedure (15). Non-saponifiable matter was determined quantitatively on a sample of the original methyl esters; the amount in the total charge was calculated and subtracted from the weight of the residue.

Saponification equivalents (or neutral equivalents, in the case of fatty acids) were run on all fractions. Iodine values (Wijs) were determined on all fractions except the C_4 and C_6 cuts of the cheese fat methyl esters. Polyunsaturated components were determined spectrophotometrically on all cuts containing C_{18} or higher components by the original method of Brice *et al.* (3) except that nitrogen blanketing was used during isomerization. "Liquid and solid fatty acids" were determined on the hydrogenated fats, and from these results and the spectrophotometric data already obtained, iso-diene acids were calculated by the method of Lemon (14).

All determinations were made at least in duplicate. Official methods of the American Oil Chemists' Society (1) were used wherever applicable.

Calculations

Calculations of fatty acid components from the ester-fractionation data were made essentially as described by Hilditch (12) for fractions up to and including the C_{16} fractions. In the fractions containing C_{18} or higher components, total unsaturated material obtained by spectrophotometric analysis and iodine value determinations was calculated, and the saturated portion obtained by difference.

Acids in the final fraction, both saturated and unsaturated, were calculated as C_{20} acids though small proportions of higher molecular weight acids were no doubt present in some instances. All acids over C_{18} are tabulated simply as "C_{20}" acids. In the case of hydrogenated acids it was assumed that "higher" unsaturates would isomerize in the same manner as C_{18} material and that the ratio of "higher" iso-monoene acid to "higher" iso-diene acid would be the same as that of iso-oleic to iso-linoleic acid.

Finally the standard factor 0.956 was used to convert the percentages of individual fatty acids to percentages fatty acids in fat except in the case of those from cheese, where the factor 0.945 was used.

Discussion

The various procedures employed in the manufacture of the samples studied seem to have little effect on the essential fatty acid contents except, of course, in the case of oils which have been hydrogenated. These samples still contained significant proportions of linoleic acid, plus iso-acids which may or may not function as essential acids (9).

Also it may be noted that even the relatively severe conditions of distillation seem to have little effect on the polyunsaturated components in the distilled compared with the non-distilled material. In no instance was there any significant increase in conjugated material in the distilled esters. This is in agreement with a previous study on the effect of heat on fatty esters (16) in which it was shown that the changes effected even in the course of prolonged distillation were less significant than the effect of chemical reagents prior to distillation.

Milk fat has always posed a problem to the analyst, not only because of its complexity but because of the volatility and water solubility of its lower molecular weight components when converted to free acids or methyl esters for analysis. The ether extraction of the acids and removal of the ether prior to esterification of the whole sample, as was done in the present work, involves less handling of the fatty material in its most sensitive form than the usual 4- or 5-hour steam-distillation of the fatty acid mixture. Furthermore fatty acids separated by steam contain, besides the normally steam-volatile components, traces of oleic and presumably of other acids. When such a mixture is fractionated and analyzed, calculations are necessarily somewhat uncertain for a small proportion of the acids. Although methanolysis, as used on the other samples, is undoubtedly the mildest method possible for the preparation of methyl esters, this process did not seem advisable in the case of the cheese fats because of the solubility of the lower molecular weight esters in the aqueous washes required. Therefore methyl esters of the total cheese fatty acids were prepared by conventional means. It was felt that fractionation of the entire sample in a single distillation should provide more reliable results than can be obtained by usual techniques.

Summary

Fatty acid constituents of two cheeses, four cured meats, and two hydrogenated oils were determined; two sets of data were obtained for each sample. Unsaturated acids were determined spectrophotometrically in the total fatty acids prepared from each fat, and saturated and unsaturated components were de-

termined chemically and spectrophotometrically on each fraction obtained from distilling the methyl esters prepared from the fats. Close agreement was obtained between the percentages of unsaturated components found in distilled and non-distilled samples. Methyl esters of cheese fats were prepared without prior distillation of steam-volatile acids; no separation of saturated and unsaturated components was made on any sample. Fatty acid components were also converted to percent of each fatty acid in each food.

Acknowledgments

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Fatty Acid Contents of Several Food Products[1]

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IN connection with a program for determining the fatty acid contents of certain processed foods (1), the fats of several foods which had undergone only relatively mild processing were also analyzed. These slightly processed foods were rolled oats, whole white corn meal, roasted peanuts, peanut butter, and walnuts. Also included in the program were Crisco shortening and raw peanuts.

The amount of each fatty acid constituent in each food is reported in Table I as the percentage by weight in the food.

The analytical methods used were essentially those previously described (1). The extracted fats were converted to their methyl esters by methanolysis, the methyl esters fractionally distilled, and the fractions analyzed chemically and spectrophotometrically.

The handling of these samples differed from that of the samples already reported principally in details of the distillation procedure. Bumping was prevented by means of a fine nitrogen capillary, except in the case of the corn oil and walnut oil esters. The distillations, other than these same two samples, were carried out intermittently nearly to the end of the C_{18} plateau. The remaining high-boiling components were then (except in the case of corn oil esters) distilled through a small vapor take-off column. The exceptions noted were treated by the refined procedures reported in the previous paper: a magnetic stirrer was utilized to prevent bumping, and fractionation was completed in a single continuous distillation.

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TABLE I

Fatty Acids in Foods
(weight percentage)

	Saturated					Unsaturated				
	C_{14}	C_{16}	C_{18}	C_{20} and Above	Total Saturated	Monoene	Diene	Triene	Tetraene	Total Unsaturated
Cornmeal (3.77% Fat)	.....	0.33	0.05	0.07	0.4(5)	1.39	1.74	0.02	.....	3.1(5)
Crisco (100.0% Fat)	0.56	14.1	6.3	1.62	22.5(8)	61.18	11.64	0.20	.....	73.0(2)
Peanut Butter (51.65% Fat)	0.54	5.84	2.9	4.0	13.2(8)	18.8	17.3	.....	.....	36.1(0)
Raw Peanuts (50.25% Fat)	0.5	5.9	3.3	5.0	14.7(0)	17.4	15.94	.....	.....	33.3(4)
Roasted Peanuts (51.3% Fat)	0.4	5.9	2.3	2.8	11.4(0)	21.8	15.84	.....	.....	37.6(4)
Rolled Oats (5.92% Fat)	0.02	0.76	0.24	0.26	1.2(8)	1.88	2.42	0.07	0.01[1]	4.3(8)
Walnuts (66.8% Fat)	0.29	3.49	1.86	.....	5.6(4)	10.63	38.90	8.66	0.03[1]	58.1(6)

NOTE: Figures in parentheses are not significant.

[1] Probably due to autoxidation products of linolenic acid (2).