The New England Journal of Medicine

VOLUME 340 JUNE 24, 1999 NUMBER 25

EFFECTS OF DIFFERENT FORMS OF DIETARY HYDROGENATED FATS ON SERUM LIPOPROTEIN CHOLESTEROL LEVELS

ALICE H. LICHTENSTEIN, D.SC., LYNNE M. AUSMAN, D.SC., SUSAN M. JALBERT, M.L.T., AND ERNST J. SCHAEFER, M.D.

ABSTRACT

Background Metabolic studies suggest that fatty acids containing at least one double bond in the trans configuration, which are found in hydrogenated fat, have a detrimental effect on serum lipoprotein cholesterol levels as compared with unsaturated fatty acids containing double bonds only in the cis configuration. We compared the effects of diets with a broad range of trans fatty acids on serum lipoprotein cholesterol levels.

Methods Eighteen women and 18 men consumed each of six diets in random order for 35-day periods. The foods were identical in each diet, and each diet provided 30 percent of calories as fat, with two thirds of the fat contributed as soybean oil (<0.5 g of trans fatty acid per 100 g of fat), semiliquid margarine (<0.5 g per 100 g), soft margarine (7.4 g per 100 g), shortening (9.9 g per 100 g), or stick margarine (20.1 g per 100 g). The effects of those diets on serum lipoprotein cholesterol, triglyceride, and apolipoprotein levels were compared with those of a diet enriched with butter, which has a high content of saturated fat.

Results The mean (±SD) serum low-density lipoprotein (LDL) cholesterol level was 177±32 mg per deciliter (4.58±0.85 mmol per liter) and the mean high-density lipoprotein (HDL) cholesterol level was 45±10 mg per deciliter (1.2±0.26 mmol per liter) after subjects consumed the butter-enriched diet. The LDL cholesterol level was reduced on average by 12 percent, 11 percent, 9 percent, 7 percent, and 5 percent, respectively, after subjects consumed the diets enriched with soybean oil, semiliquid margarine, soft margarine, shortening, and stick margarine; the HDL cholesterol level was reduced by 3 percent, 4 percent, 4 percent, 4 percent, and 6 percent, respectively. Ratios of total cholesterol to HDL cholesterol were lowest after the consumption of the soybean-oil diet and semiliquid-margarine diet and highest after the stick-margarine diet.

Conclusions Our findings indicate that the consumption of products that are low in trans fatty acids and saturated fat has beneficial effects on serum lipoprotein cholesterol levels. (N Engl J Med 1999;340: 1933-40.)

THE results of controlled metabolic studies have suggested that dietary fatty acids containing at least one double bond in the trans configuration (trans fatty acids) have a detrimental effect on serum lipid levels relative to fatty acids containing only double bonds in the cis configuration or, in some cases, saturated fatty acids.[1,2] Adverse effects of dietary trans fatty acids on the risk of the development of cardiovascular disease have also been reported in some studies of large cohorts.[3,4] Trans fatty acids are naturally present at low levels in meat and dairy products as a result of bacterial fermentation in ruminant animals. They are also formed in varying amounts during the hydrogenation of oil, a process used to transform oil from a liquid to a semisolid or solid state. In addition to forming trans double bonds, hydrogenation also results in the saturation of some double bonds and the migration of others along the acyl chain. Hydrogenated fat is used in the manufacture of margarines and vegetable shortening and is therefore in foods prepared with the use of these products.

Unresolved issues relating to the physiologic effects of dietary trans fatty acids in humans include the actual magnitude of the unfavorable effect relative to saturated or other unsaturated fatty acids on individual serum lipoprotein and apolipoprotein levels. Although recent studies consistently demonstrate a positive relation between the level of intake of trans fatty acids and low-density lipoprotein (LDL) cholesterol levels, data on the effect of trans fatty acids on high-density lipoprotein (HDL) cholesterol levels have proved less conclusive.[1,2,5,6] Similarly, trans

From the Lipid Metabolism Laboratory, Jean Mayer U.S. Department of Agriculture Human Nutrition Research Center on Aging at Tufts University, Boston (A.H.L., S.M.J., E.J.S.); and the School of Nutrition Science and Policy, Tufts University, Medford, Mass. (L.M.A.). Address reprint requests to Dr. Lichtenstein at the Jean Mayer USDA Human Nutrition Research Center on Aging at Tufts University, 711 Washington St., Boston, MA 02111, or at lichtenst_li@hnrc.tufts.edu.

fatty acids have been reported to increase serum Lp(a) lipoprotein levels in some, but not all, studies.[6-9]

We report the results of a trial in which different forms of commercially available margarines and a vegetable shortening with a wide range of trans-fatty-acid levels were substituted for butter.

METHODS

Subjects

Eighteen women and 18 men who were over the age of 50 years and whose serum LDL cholesterol levels exceeded 130 mg per deciliter (3.36 mmol per liter) were recruited for the study from the greater Boston area. All 36 subjects had normal kidney, liver, thyroid, and cardiac function, as well as normal serum glucose levels after fasting. None were taking medications known to affect serum lipid levels, and none were smokers. All 18 women were postmenopausal. Subjects who were being treated for hypertension were excluded from the study only if they were taking beta-blockers; subjects using other medications were required to continue taking the same drug throughout the study period. The characteristics of the study subjects at the time of screening are shown in Table 1. This protocol was approved by the human-investigation review committee of New England Medical Center and Tufts University, and all subjects gave written informed consent.

Study Design

Study subjects were given in a double-blinded fashion six experimental diets for periods of 35 days each according to a Latin-square design. The subjects reported to our metabolic research unit four times per week, at which time they were weighed and ate one meal on site. All food and drink were provided to the subjects in containers appropriate for either microwave or conventional ovens, obviating the need to transfer the food from the containers before consumption. The subjects were required to consume all that was provided to them and not to supplement their diet with any other food or drink except water and noncaloric beverages. Initial caloric levels were estimated with use of the Harris–Benedict formula and were adjusted, when necessary, to maintain body weight. The mean (±SD) energy intake was 2114±320 kcal for the women and 2792±518 kcal for the men. Three times after day 28 of each diet, blood samples were obtained after a 14-hour fast for the measurement of serum lipids and apolipoproteins. The mean value of the three measurements is reported and was used for statistical analysis.

TABLE 1. CHARACTERISTICS OF THE SUBJECTS AT THE TIME OF SCREENING.

CHARACTERISTIC	WOMEN (N=18)	MEN (N=18)	ALL SUBJECTS (N=36)
		mean ±SD	
Age (yr)	67±4	60±5	63±6
Body-mass index*	26.6±2.4	28.1±3.4	27.4±3.0
Serum cholesterol (mg/dl)†			
Total	253±32	237±33	245±33
Very-low-density lipoprotein	31±13	28±11	29±12
Low-density lipoprotein	167±30	167±26	167±28
High-density lipoprotein	53±11	42±9	48±11
Serum triglycerides (mg/dl)‡	158±71	138±55	148±64

*The body-mass index was calculated as the weight in kilograms divided by the square of the height in meters.

†To convert values for cholesterol to millimoles per liter, divide by 38.67.

‡To convert values for triglycerides to millimoles per liter, divide by 88.54.

Diets

A single diet was designed that provided 30 percent of calories as fat. The foods included in each of the six test diets were identical, the only difference being that two thirds of the fat was provided in the form of soybean oil, soybean-oil–based margarines, soybean-oil–based shortening, or butter. The soybean-oil diet was designed to meet Step 2 criteria.[10] These criteria were achieved by first designing a diet containing 10 percent of calories as fat and then adding the soybean oil to various foods such as hot cereal, casseroles, and muffins to increase the fat content of the diet to 30 percent of calories. These foods were consumed throughout the day. The other study diets were created by substituting semiliquid margarine sold in squeeze bottles, soft margarine sold in tubs, hydrogenated soybean oil (shortening), margarine sold in sticks, or butter for the oil. All the margarines and the shortening were made from soybean oil except the semiliquid margarine, which contained some cottonseed oil. This deviation was necessary because at the time of the study no semiliquid margarine made solely with soybean oil was commercially available.

The fatty-acid profile of the test fats was determined by Best Foods Research and Engineering Center (Union, N.J.) with the use of capillary gas chromatography (Table 2). Analysis of the protein, carbohydrate, fat, and cholesterol content of the food in all diets was carried out by Covance Laboratories (Madison, Wis.), and the fatty-acid content was analyzed by capillary gas chromatography by Lipton (Baltimore) (Table 3).

Biochemical Analysis

Fasting blood samples were collected in tubes containing EDTA (final concentration, 0.15 percent). Serum was separated by centrifugation at 1100×*g* at 4°C. Very-low-density lipoprotein (VLDL) was isolated from serum by ultracentrifugation at 109,000×*g* at 4°C.[11] Serum and the infranatant (1.006 g per milliliter) were assayed for total cholesterol and triglyceride with a biochromatic analyzer (model CCX, Spectrum, Incstar, Stillwater, Minn.) with enzymatic reagents.[12] Serum HDL cholesterol was measured in the supernatant fraction after precipitation of lipoproteins containing apolipoprotein B with the use of dextran–magnesium sulfate.[13] Lipid assays were standardized through the Lipid Standardization Program of the Centers for Disease Control and Prevention (Atlanta). Serum apolipoprotein A-I and apolipoprotein B were measured by immunoturbidometric assays with a Spectrum CCX analyzer (Incstar).[14,15] Lp(a) lipoprotein was quantified as previously described (Terumo Medical, Elkton, Md.).[16]

Statistical Analysis

Before the analysis, descriptive statistics and graphs (Proc Univariate and Proc Means, SAS, Cary, N.C.) were used to summarize the overall effects of diets and distributions of the outcome measures. When we noted violations of the basic testing assumptions, we used appropriate transformations of the data. An analysis of variance (Proc GLM, SAS) with the main effect of diet and subject as the repeated measure was carried out for each outcome measure, followed by the Tukey honestly-significant-difference test for the pairwise comparisons of the six diets.

Serum total, LDL, and HDL cholesterol were described through a covariance analysis with the Proc GLM program (SAS) in which levels of the design factor were defined by the response of an individual subject exposed to a set of diets; the covariates were the intake of saturated, monounsaturated, cis polyunsaturated, and trans fatty acids (expressed as the percentage of total daily energy intake) and dietary cholesterol (expressed as milligrams per 1000 kcal). The inclusion of the response of an individual subject in the equation should control for the inherent level of responsiveness of the subjects. The resulting equation, minus the subject-specific intercepts, can be used to estimate the change in total, LDL, or HDL cholesterol levels or triglyceride level resulting from a

TABLE 2. FATTY-ACID COMPOSITION OF THE FATS USED IN THE DIETS.*

FATTY-ACID SUBCLASS	SOYBEAN OIL	SEMILIQUID MARGARINE	SOFT MARGARINE	SHORTENING	STICK MARGARINE	BUTTER
			grams per 100 g of fat			
Saturated fatty acids	25.0	27.0	25.2	29.3	27.0	61.7
Monounsaturated fatty acids†	27.8	25.4	25.6	32.2	25.4	21.5
Polyunsaturated fatty acids†	41.4	42.0	35.5	25.5	20.5	7.0
Trans fatty acids	<0.5	<0.5	7.4	9.9	20.1	1.5

*Values are based on chemical analysis of the fats and oils.

†Only isomers containing cis double bonds are included.

TABLE 3. COMPOSITION OF THE SIX DIETS.*

CONSTITUENT	SOYBEAN OIL	SEMILIQUID MARGARINE	SOFT MARGARINE	SHORTENING	STICK MARGARINE	BUTTER
			percentage of total daily energy intake			
Protein	15.74	17.07	16.28	16.79	16.73	16.94
Carbohydrate	55.77	51.73	52.91	53.22	53.54	53.97
Fat	28.48	31.20	30.80	29.98	29.72	29.08
Saturated fatty acids	7.30	8.59	8.40	8.56	8.47	16.70
12:0	0.83	0.96	0.67	0.75	0.82	1.35
14:0	0.63	0.74	0.55	0.57	0.60	2.50
16:0	3.65	4.26	4.18	3.91	4.03	7.47
18:0	1.45	1.85	2.28	2.64	2.22	3.57
Monounsaturated fatty acids†	8.14	8.08	8.04	9.92	8.46	8.07
18:1	7.20	7.11	6.65	7.51	6.53	6.97
Polyunsaturated fatty acids†	12.48	13.54	11.14	8.13	6.34	2.43
18:2	10.74	12.10	9.99	7.22	5.60	2.07
18:3	1.67	1.39	1.10	0.55	0.70	0.29
Trans fatty acids	0.55	0.91	3.30	4.15	6.72	1.25
			milligrams per 1000 kcal			
Cholesterol‡	65.9	68.0	70.3	62.5	66.5	121.0

*Because of rounding, percentages may not total 100.

†Only isomers containing cis double bonds are included.

‡To convert values to milligrams per megajoule, divide by 239.

change from one diet to another, while taking into consideration the changes in the dietary fatty acid and cholesterol content. Simple and multiple regression equations, including all possible interactions, were used with the above model to examine the ability of a change in diet to predict the change in serum lipid levels.

RESULTS

Serum total and LDL cholesterol levels changed in response to the predominant fat in the diet (Table 4). Levels were lowest after subjects consumed the soybean-oil and semiliquid-margarine diets and increased progressively after subjects consumed the soft-margarine diet, shortening and stick-margarine diets, and butter diet. For both total and LDL cholesterol, the pattern of response for female and male subjects was similar.

The pattern of response of HDL serum cholesterol was distinctly different from that of total and LDL cholesterol. With the exception of the stick-margarine diet (lowest) and butter diet (highest), the differences among the diets were small and statistically indistinguishable. The differences in HDL cholesterol levels for the whole group were restricted, for the most part, to the female subjects.

The highest serum triglyceride and VLDL cholesterol levels were observed after subjects consumed the stick-margarine diets and lowest after they con-

TABLE 4. SERUM LIPID, LIPOPROTEIN, AND APOLIPOPROTEIN LEVELS AT THE END OF EACH DIET.*

VARIABLE	SOYBEAN OIL	SEMILIQUID MARGARINE	SOFT MARGARINE	SHORTENING	STICK MARGARINE	BUTTER
Total cholesterol (mg/dl)	225±32†‡	226±30†‡	232±28†‡	235±32†	243±37§¶‖	251±36§¶‖**
Women	225±29†‡	229±28†‡	238±30†	238±32†	244±32§¶	254±31§¶‖**
Men	226±35†‡	223±36†‡	226±26†	233±33†	242±42§¶	249±41§¶‖**
VLDL cholesterol (mg/dl)	28±13	28±9‡	30±13	29±10	33±15¶	29±13
Women	25±11	28±8	27±8	28±11	30±13	27±11
Men	32±14	28±10‡	34±17	29±9‡	36±17¶**	32±14
LDL cholesterol (mg/dl)	154±28†‡**	155±27†‡	159±26†	164±28†§	168±30†§¶	177±32‡§¶‖**
Women	153±32†‡	155±25†‡	164±29†	164±29	169±29§¶	177±31§¶‖
Men	155±25†	156±30†	154±23†	164±28	167±32	177±33§¶‖
HDL cholesterol (mg/dl)	43±9	43±10	43±9	43±9	42±9†	45±10‡
Women	47±9	47±10	47±9	45±10†	45±9†	50±10‡**
Men	40±7	40±8	39±7	40±8	39±8	40±9
Triglyceride (mg/dl)††	143±64	133±54‡	149±78	147±53	156±68¶	146±57
Women	131±51	133±53	138±52	145±58	151±64	138±46
Men	155±74	134±56	159±98	148±49	161±74	154±66
Total cholesterol:HDL cholesterol	5.43±1.24†‡	5.47±1.21†‡	5.65±1.20‡	5.68±1.12	6.03±1.27§¶‖	5.85±1.40§¶
Women	4.99±1.17‡**	5.11±1.09‡	5.29±1.20	5.44±1.20§	5.62±1.12§¶	5.31±1.24
Men	5.86±1.19	5.83±1.25‡	6.01±1.12	5.92±1.01	6.44±1.30¶	6.39±1.36
Apolipoprotein A-I (mg/dl)	147±23‡	145±23†	145±20†	145±21†	141±21†§	151±25‡¶‖**
HDL cholesterol:apolipoprotein A-I	0.29±0.04	0.30±0.04	0.29±0.03	0.29±0.04	0.29±0.04	0.30±0.04
Apolipoprotein B	132±26†‡	131±25†‡**	135±23†‡	138±27¶	143±27§¶‖	144±25§¶‖
LDL cholesterol:apolipoprotein B	1.17±0.11†	1.19±0.11†	1.18±0.11†	1.18±0.10†	1.18±0.12†	1.24±0.13‡§¶‖**
Apolipoprotein B:apolipoprotein A-I	0.91±0.21†‡**	0.92±0.21†‡	0.95±0.21‡	0.97±0.22‡§	1.03±0.22§¶‖**	0.98±0.22§¶
Lp(a) lipoprotein						
Mean (mg/dl)††	23±20	23±21	24±22	24±21	24±22†	22±21‡
Median (mg/dl)	15.2	14.7	14.1	14.8	15.7	13.0
≤30 mg/dl (no. of subjects)‡‡	13±8	13±7	13±7	13±7	14±9†	12±7‡

*Plus–minus values are means ±SD. Analysis of variance with the main effect of diet and subject as the repeated measure was carried out for each outcome measure followed by the Tukey honestly-significant-difference test for the pairwise comparisons of the six diets. To convert values for cholesterol to millimoles per liter, divide by 38.67. To convert values for triglyceride to millimoles per liter, divide by 88.54. VLDL denotes very-low-density lipoprotein, LDL low-density lipoprotein, and HDL high-density lipoprotein.

†P<0.05 for the comparison with butter.

‡P<0.05 for the comparison with stick margarine.

§P<0.05 for the comparison with soybean oil.

¶P<0.05 for the comparison with semiliquid margarine.

‖P<0.05 for the comparison with soft margarine.

**P<0.05 for the comparison with shortening.

††Values were log-transformed before statistical analysis.

‡‡Values were 30 mg per deciliter or less in 28 subjects at the time of screening.

sumed the semiliquid-margarine diets. The pattern of response was not related to that of total, LDL, or HDL cholesterol levels.

Serum apolipoprotein A-I and apolipoprotein B levels mirrored, for the most part, those of HDL and LDL cholesterol, respectively. The magnitude of the difference in the apolipoprotein levels was not as great as that in the lipoprotein cholesterol levels. The patterns of response were similar for female and male subjects (data not shown). The ratios of HDL cholesterol to apolipoprotein A-I and of LDL cholesterol to apolipoprotein B showed only small variations in response to alterations in the type of dietary fat, suggesting that there was little change in the composition of the HDL and LDL particles.

The ratios of total cholesterol to HDL cholesterol reflected the combined and, at times, opposite effect of the various fats. The ratio was least favorable after subjects consumed the stick-margarine diet and most favorable after subjects consumed the soybean-oil diet and the semiliquid-margarine diet. The data were similar when female and male subjects were considered separately. The pattern observed for the ratios of total cholesterol to HDL cholesterol was also reflected in the ratios of apolipoprotein B to apolipoprotein A-I.

Serum total cholesterol levels were 10 percent lower after the consumption of the soybean-oil diet or semiliquid-margarine diet than after the butter diet, and LDL cholesterol levels were 11 to 12 percent lower (Fig. 1). The decreases were 3 percent and 5 per-

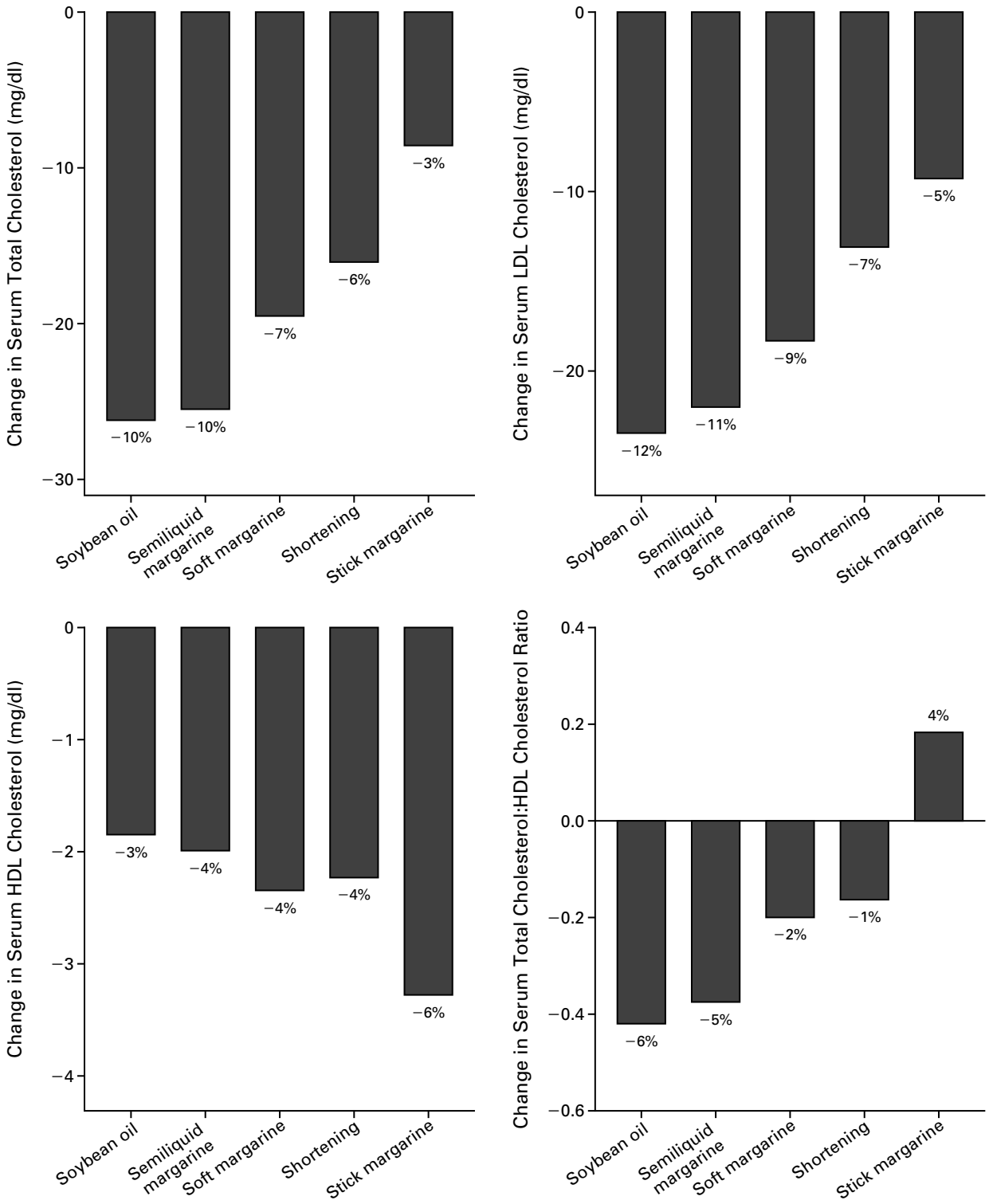

Figure 1. Change in Serum Lipid Levels after the Consumption of Diets Enriched in Soybean Oil, Semiliquid Margarine, Soft Margarine, Shortening, and Stick Margarine.

Values are expressed as the percent change from the values for butter; absolute values are given in Table 4. To convert values for cholesterol to millimoles per liter, divide by 38.67. LDL denotes low-density lipoprotein, and HDL high-density lipoprotein.

cent, respectively, after the consumption of the stick-margarine diet. The values for the soft-margarine and shortening diets were intermediate. In contrast, HDL cholesterol levels were 3 percent lower after the consumption of the soybean-oil diet and 6 percent lower after the consumption of the stick-margarine diet. These changes resulted in a 4 percent increase in the ratio of total cholesterol to HDL cholesterol in the case of the stick-margarine diet and a 5 to 6 percent decrease in the case of the soybean-oil and semiliquid-margarine diets.

Considerable attention has been focused on the effect of the intake of trans fatty acids and hydrogenated fat on Lp(a) lipoprotein levels.[6-9] There was a small but significant effect of the type of dietary fat on Lp(a) lipoprotein levels that was largely restricted to subjects with Lp(a) lipoprotein levels of 30 mg per deciliter or less at the time of screening (Table 4). Lp(a) lipoprotein levels were lowest after subjects consumed the butter diet and highest after they consumed the stick-margarine diet.

Analysis of covariance indicated that the changes in the levels of serum total cholesterol, LDL and HDL cholesterol, and triglyceride were best reflected by the following relations: the change in total cholesterol = 2.77 ($\pm$0.34) $\times$ the change in saturated fatty acids + 2.54 ($\pm$0.49) $\times$ the change in trans fatty acids, with 215 df; the change in LDL cholesterol = 2.46 ($\pm$0.30) $\times$ the change in saturated fatty acids + 2.04 ($\pm$0.44) $\times$ the change in trans fatty acids, with 215 df; the change in HDL cholesterol = 0.22 ($\pm$0.08) $\times$ the change in saturated fatty acids $-$ 0.23 ($\pm$0.11) $\times$ the change in trans fatty acids, with 215 df; and the change in triglyceride = 2.53 ($\pm$0.93) $\times$ the change in trans fatty acids, with 215 df. Values in parentheses are standard errors of the regression coefficient. Because the intakes of energy, carbohydrate, and protein were controlled by the study design, the coefficients for saturated and trans fatty acids are adjusted for each other and represent the effects of exchanging these fats for equivalent amounts of cis unsaturated fatty acids (polyunsaturated and monounsaturated fats combined).[17] In models used to predict the triglyceride level, the model with trans fatty acids alone had a significant positive coefficient, suggesting that trans fatty acids elevate triglyceride levels. When other fatty acids were added to this model, none of the coefficients were significant.

DISCUSSION

A number of public health organizations and advisory committees have issued both general recommendations (e.g., choose a diet low in fat, saturated fat, and cholesterol)[18] and more specific guidelines (e.g., consume a diet containing $\leq$30 percent of calories from fat, or follow a diet containing $<$10 percent of calories from saturated fat [Step 1 diet] or $<$7 percent of calories from saturated fat [Step 2 diet] and $<$300 mg of cholesterol per day [Step 1 diet] or $<$200 mg of cholesterol per day [Step 2 diet])[10,19] for dietary fat intake. No specific recommendations for the intake of hydrogenated fat or trans fatty acids have been made, although an advisory statement from the American Heart Association has suggested substituting "unhydrogenated oil for hydrogenated or saturated fat in processed foods . . . [and substituting] softer for harder margarines and cooking fats."[20] The latter recommendation was based on conclusions from a limited number of studies. Our data on the effect of different sources of commercially available hydrogenated fat representing a wide range of trans-fatty-acid levels on lipoprotein and apolipoprotein levels[21] were derived from subjects with moderately elevated LDL cholesterol levels — hence, candidates for dietary intervention — and within the context of reduced-fat diets, as currently recommended for these people.[10,19]

Our results suggest that the use of soybean oil or semiliquid margarine results in the most favorable total and LDL cholesterol levels and ratios of total cholesterol to HDL cholesterol, whereas the use of stick margarine or butter results in the opposite effect. Soybean oil and semiliquid margarine have the lowest levels of trans fatty acids of the various fats that we studied, and they are also low in saturated fat. The fats with intermediate levels of trans and saturated fatty acids resulted in intermediate values. These findings corroborate and extend the work of previous investigations.[1,2,5-8,22-24] In addition, from a practical perspective, they provide a sound basis on which to make a strong recommendation to the general public and food manufacturers to emphasize the use of vegetable oils in their natural state and after minimal hydrogenation.

Considerable attention has been focused on the effect of trans fatty acids on HDL cholesterol levels after initial reports suggesting not only an LDL cholesterol–raising effect but also an HDL cholesterol–lowering effect.[1,22] We also identified an HDL cholesterol–lowering effect of trans fatty acids, similar in magnitude to the HDL cholesterol–raising effect of saturated fatty acids. The HDL cholesterol–lowering effect was most marked with the consumption of stick margarine, which has the highest content of trans fatty acids.

The differential effects of hydrogenated fat on the individual lipoprotein subclasses resulted in the lowest ratios of total cholesterol to HDL cholesterol after the consumption of the soybean oil or lightly hydrogenated margarine (semiliquid), intermediate ratios after the consumption of moderately hydrogenated fats (soft margarine, shortening, and butter), and highest ratios after the consumption of the heavily hydrogenated margarine (stick). These data are consistent with those reported for other metabolic studies[1,2,6,22-24] and suggest that the current recommendation to re-

strict the intake of both saturated fat and trans fatty acids is appropriate.[10,18,19]

Elevated Lp(a) lipoprotein levels are associated with an increased risk of the development of cardiovascular disease.[25] Circulating levels are determined in part by one's phenotype and to a limited extent by other factors, including diet.[26] Trans fatty acids have been reported to increase Lp(a) lipoprotein levels, although this finding has not been universal and in some cases has been limited to persons with relatively high Lp(a) lipoprotein levels.[5-8,22,23] A confounding factor in some studies may be the independent Lp(a) lipoprotein–lowering effect of saturated fatty acids. Hence, data from studies designed to assess the effect of trans fatty acids alone need to be interpreted with caution relative to data from studies designed to assess the effect of hydrogenated fat on Lp(a) lipoprotein levels. Our results confirm those of previous studies and suggest an Lp(a) lipoprotein–lowering effect of saturated fat and an Lp(a) lipoprotein–raising effect of trans fatty acids. In contrast to previous reports, this effect was not restricted to persons with Lp(a) lipoprotein levels above 30 mg per deciliter.[8]

A limitation of our study is that relatively high levels of a single fat were used, in contrast to the more likely situation in real life in which multiple sources and forms of fat are freely exchanged in the diet. This approach allowed us to assess different forms of commercially available fats, and our conclusions can be extrapolated to mixed products. There is a wide range of absolute intakes of fat and considerable variability in patterns of food intake among and within subgroups of the population in the United States. Precise data on the sources and quantity of the intakes of trans fatty acids are lacking. However, it appears that the range of intakes of trans fatty acids included in our study spans that currently consumed in the United States.[21]

Another limitation of this work is that each dietary fat assessed included a mixture of fatty-acid isomers containing at least one trans double bond. Although this approach does not allow an assessment of the relative effect of the individual isomers, it does allow the assessment of the hydrogenated vegetable fats currently available. Only fats derived from soybean oil were studied, to minimize dietary variables other than hydrogenation. Soybean oil accounts for approximately three quarters of the vegetable oil consumed in the United States, a little more than half of which is hydrogenated to some extent. Hydrogenated fats made from other vegetable oils would be expected to have different fatty-acid profiles; however, previous work has suggested that consumption of hydrogenated fats made from a wide range of oils has similar effects.[27]

Our results suggest that both the general public and patients with hypercholesterolemia should be encouraged to use vegetable oil in its natural state or after minimal hydrogenation and to use products made from this type of oil. Vegetable shortening and stick margarine have advantages over butter with respect to LDL cholesterol levels, yet they result in higher LDL cholesterol levels than does soybean oil and similar or less favorable ratios of total cholesterol to HDL cholesterol, and they are therefore less preferable.

Supported by a grant (HL 54727) from the National Institutes of Health and by a contract (53-3KO6-5-10) with the Department of Agriculture.

The contents of this article do not necessarily reflect the views or policies of the Department of Agriculture, nor does mention of trade names, commercial products, or organizations imply endorsement by the U.S. government.

We are indebted to the staff of the Metabolic Research Unit, including Helen Rasmussen, M.S., R.D., for their expert care of the study subjects; to John Griffith, Ph.D., for advice regarding the statistical analysis of the data; and to the study subjects, without whom this investigation would not have been possible.

REFERENCES

1. Mensink RP, Katan MB. Effect of dietary trans fatty acids on high-density and low-density lipoprotein cholesterol levels in healthy subjects. N Engl J Med 1990;323:439-45.

2. Judd JT, Clevidence BA, Meusing RA, Wittes J, Sunkin ME, Podczasy JJ. Dietary trans fatty acids: effects on plasma lipids and lipoproteins of healthy men and women. Am J Clin Nutr 1994;59:861-8.

3. Hu FB, Stampfer MJ, Manson JE, et al. Dietary fat intake and the risk of coronary heart disease in women. N Engl J Med 1997;337:1491-9.

4. Kromhout D, Menotti A, Bloemberg B, et al. Dietary saturated and trans fatty acids and cholesterol and 25-year mortality from coronary heart disease: the Seven Countries Study. Prev Med 1995;24:308-15.

5. Zock PL, Mensing RP. Dietary trans-fatty acids and serum lipoproteins in humans. Curr Opin Lipidol 1996;7:34-7.

6. Lichtenstein AH, Ausman LM, Carrasco W, Jenner JL, Ordovas JM, Schaefer EJ. Hydrogenation impairs the hypolipidemic effect of corn oil in humans: hydrogenation, trans fatty acids, and plasma lipids. Arterioscler Thromb 1993;13:154-61.

7. Mensink RP, Zock PL, Katan MB, Hornstra G. Effect of dietary cis and trans fatty acids on serum lipoprotein[a] levels in humans. J Lipid Res 1992;33:1493-501.

8. Clevidence BA, Judd JT, Schaefer EJ, et al. Plasma lipoprotein (a) levels in men and women consuming diets enriched in saturated, cis-, or trans-monounsaturated fatty acids. Arterioscler Thromb Vasc Biol 1997;17:1657-61.

9. Nestel P, Noakes M, Belling B, et al. Plasma lipoprotein lipid and Lp[a] changes with substitution of elaidic acid for oleic acid in the diet. J Lipid Res 1992;33:1029-36.

10. The Expert Panel. Report of the National Cholesterol Education Program Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults. Arch Intern Med 1988;148:36-69.

11. Havel RJ, Eder HA, Bragdon JH. Distribution and chemical composition of ultracentrifugally separated lipoproteins in human serum. J Clin Invest 1955;34:1345-53.

12. McNamara JR, Schaefer EJ. Automated enzymatic standardized lipid analyses for plasma and lipoprotein fractions. Clin Chim Acta 1987;166:1-8.

13. Warnick GR, Benderson J, Albers JJ. Dextran sulfate-Mg2+ precipitation procedure for quantitation of high-density-lipoprotein cholesterol. Clin Chem 1982;28:1379-88.

14. Contois JH, McNamara JR, Lammi-Keefe CJ, Wilson PWF, Massov T, Schaefer EJ. Reference intervals for plasma apolipoprotein A-I determined with a standardized commercial immunoturbidimetric assay: results from the Framingham Offspring Study. Clin Chem 1996;42:507-14.

15. *Idem.* Reference intervals for plasma apolipoprotein B determined with a standardized commercial immunoturbidimetric assay: results from the Framingham Offspring Study. Clin Chem 1996;42:515-23.

16. Jenner JL, Ordovas JM, Lamon-Fava S, et al. Effects of age, sex, and menopausal status on plasma lipoprotein(a) levels: the Framingham Offspring Study. Circulation 1993;87:1135-41.

17. Hegsted DM, Ausman LM, Johnson JA, Dallal GE. Dietary fat and serum lipids: an evaluation of the experimental data. Am J Clin Nutr 1993;57:875-83. [Erratum, Am J Clin Nutr 1993;58:245.]

18. Kennedy E, Meyers L, Layden W. The 1995 dietary guidelines for Americans: an overview. J Am Diet Assoc 1996;96:234-7.
19. Krauss RM, Deckelbaum RJ, Ernst N, et al. Dietary guidelines for healthy American adults: a statement for health professionals from the Nutrition Committee, American Heart Association. Circulation 1996;94: 1795-800.
20. Lichtenstein AH. Trans fatty acids, plasma lipid levels, and risk of developing cardiovascular disease: a statement for healthcare professionals from the American Heart Association. Circulation 1997;95:2588-90.
21. Emken EA. Physicochemical properties, intake, and metabolism. Am J Clin Nutr 1995;62:659S-669S.
22. Zock PL, Katan MB. Hydrogenation alternatives: effects of trans fatty acids and stearic acid versus linoleic acid on serum lipids and lipoproteins in humans. J Lipid Res 1992;33:399-410.
23. Almendingen K, Jordal O, Kierulf P, Sandstad B, Pedersen JI. Effects of partially hydrogenated fish oil, partially hydrogenated soybean oil, and butter on serum lipoproteins and Lp[a] in men. J Lipid Res 1995;36:1370-84.
24. Aro A, Jauhiainen M, Partanen R, Salminen I, Mutanen M. Stearic acid, trans fatty acids, and dairy fat: effects on serum and lipoprotein lipids, apolipoproteins, lipoprotein(a), and lipid transfer proteins in healthy subjects. Am J Clin Nutr 1997;65:1419-26.
25. Wild SH, Fortmann SP, Marcovina SM. A prospective case-control study of lipoprotein(a) levels and apo(a) size and risk of coronary heart disease in Stanford Five-City Project participants. Arterioscler Thromb Vasc Biol 1997;17:239-45. [Erratum, Arterioscler Thromb Vasc Biol 1997;17:1010.]
26. Brown SA, Morrisett JD, Boerwinkle E, Hutchinson R, Patsch W. The relation of lipoprotein[a] concentrations and apolipoprotein[a] phenotypes with asymptomatic atherosclerosis in subjects of the Atherosclerosis Risk in Communities (ARIC) Study. Arterioscler Thromb 1993;13:1558-66.
27. Denke MA. Serum lipid concentrations in humans. Am J Clin Nutr 1995;62:693S-700S.