

# Effects of medium chain fatty acids (MCFA), myristic acid, and oleic acid on serum lipoproteins in healthy subjects

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**Abstract** In this study we investigated the effects on lipoproteins of medium chain fatty acids (MCFA) and myristic acid relative to those of oleic acid. Thirty-seven women and 23 men consumed a 3-wk run-in diet enriched in oleic acid followed by a 6-wk test diet rich in MCFA ( $n = 21$ ), myristic ( $n = 20$ ), or oleic acid ( $n = 19$ ). Experimental fats were incorporated into solid foods. Total fat intake was 40 En% fat. The dietary compositions were the same except for 10 En%, which was provided by MCFA, myristic, or oleic acids, respectively. With the myristic acid diet, low density lipoprotein (LDL) cholesterol was 0.37 mmol/L higher compared with the oleic acid diet ( $P = 0.0064$  for difference in changes). The MCFA diet increased LDL cholesterol, though not significantly, with 0.23 mmol/L relative to the oleic acid diet ( $P = 0.0752$ ). Compared with the oleic acid diet, HDL cholesterol concentrations increased with the myristic acid diet by 0.10 mmol/L ( $P = 0.0273$ ) but not with the MCFA diet. The MCFA diet slightly elevated triacylglycerol concentrations, but responses did not significantly differ between the diets. The MCFA diet significantly decreased the apoA-I to apoB ratio compared with both other diets ( $P < 0.02$ ). We conclude that MCFA raise LDL cholesterol concentrations slightly and affect the apoA-I to apoB ratio unfavorably compared with oleic acid. Myristic acid is hypercholesterolemic, although less than predicted earlier, and raises both LDL and HDL cholesterol concentrations compared with oleic acid.—**Temme, E. H. M., R. P. Mensink, and G. Hornstra.** Effects of medium chain fatty acids (MCFA), myristic acid, and oleic acid on serum lipoproteins in healthy subjects. *J. Lipid Res.* 1997. **38**: 1746–1754.

**Supplementary key words** saturated fatty acids • MCFA • myristic acid • oleic acid • lipoproteins • cholesterol • apolipoproteins • healthy humans

Medium chain fatty acids (MCFA) that have between six and ten carbon atoms are not only used in diets for patients with malabsorption disorders, but also in structured lipids like caprenin (50% caprylic acid (C8:0) plus capric acid (C10:0), and 45% behenic acid (C22:0)). After absorption, the transport of MCFA through the blood differs from longer chain fatty acids. MCFA

are not incorporated into triacylglycerols packaged with chylomicrons as longer chain fatty acids are, but are transported primarily through the portal vein to the liver and provide the body with a rapid source of energy (1). It has been suggested that effects of MCFA on serum cholesterol are similar to those of carbohydrates (2), but whether the effects of MCFA on different lipoproteins are indeed comparable remains to be determined. Studies carried out to investigate effects of MCFA on lipoproteins found total cholesterol levels similar to concentrations on western diets (3–5), increased triacylglycerol concentrations (3, 4), and decreased HDL concentrations (3, 5). These studies, however, were of short duration (3) and were carried out with a limited number of subjects (4, 6). In addition, the amount of MCFA was not the only variable among the experimental diets (3–7).

Certain edible fats like coconut oil, palm kernel oil, and butterfat do not only contain considerable amounts of MCFA, but are also rich in myristic acid. Myristic acid might be the most potent cholesterol-raising fatty acid (8, 9). Recent studies in healthy volunteers consuming diets enriched in myristic acid, however, yielded contradictory results (10, 11). Compared to palmitic acid, myristic acid raised total, LDL and HDL cholesterol in one study (10) but only increased HDL cholesterol concentrations in another (11). In both studies, however, myristic acid was less cholesterolemic than predicted by the formula developed by Hegsted et al. (8). We, therefore, decided to compare in healthy volunteers the effects on serum lipids and lipoproteins of an MCFA- and a myris-

Abbreviations: apo, apolipoprotein; CETP, cholesteryl ester transfer protein; CI, confidence interval; En%, percent of energy; HDL, high density lipoprotein; LDL, low density lipoprotein; Lp[a], lipoprotein[a]; MCFA, medium chain fatty acids; MCT, medium chain triglycerides.

tic acid-enriched diet with those of a diet enriched in oleic acid.

## SUBJECTS AND METHODS

### Subjects

The volunteers were recruited via advertisements in local newspapers and university newsletters, via posters in university buildings and other public buildings, and announcements on local radio and television. People responding to the advertisements were informed about the study purposes and requirements; 120 volunteers underwent the selection procedure which consisted of two fasting blood samples taken 1 week apart to determine serum lipids and lipoproteins, measurement of blood pressure, collection of a urine sample for determination of glucose and protein, and a medical questionnaire. Seventy volunteers met our selection criteria. All had serum total cholesterol concentrations below 6.7 mmol/L, blood pressures below 140/80 mm Hg, no glucosuria, no proteinuria, and did not use any medication known to affect blood lipids, coagulation, fibrinolysis, or platelet aggregation. All selected volunteers gave their written informed consent.

During the study period seven subjects withdrew. One subject because of job commitments, two subjects because of illness, and four subjects because of reasons specifically related to the strict study protocol. Results of three subjects were excluded from analyses. Two subjects, after being enrolled, started to use medications known to affect blood lipids. The third subject was excluded because her results were highly deviant for most variables possibly due to stressful personal circumstances: LDL cholesterol concentrations were 3.86 mmol/L after the oleic acid run-in period and 4.91 mmol/L after the oleic acid test period.

Thus, analyses included 37 women and 23 men. Women were aged 22–60 y (mean 40 y), weighed 48–101 kg (mean 67 kg), were between 155 and 183 cm in height (mean 167 cm), while body mass indexes ranged between 20 and 30 kg/m<sup>2</sup> (mean 24 kg/m<sup>2</sup>). The men were aged 23–59 y (mean 43 y), weighed 56–88 kg (mean 78 kg), were between 163 and 190 cm in height (mean 178 cm), and had body mass indexes from 17 to 29 kg/m<sup>2</sup> (mean 25 kg/m<sup>2</sup>). Nine women were post menopausal and 10 women used oral contraceptives. Six women and five men smoked. In women, fasting concentrations of serum lipids ranged from 3.64 to 6.38 mmol/L (mean 5.28 mmol/L) for total cholesterol, 1.13 to 2.98 mmol/L (mean 1.65 mmol/L) for HDL cholesterol, and 0.17 to 1.94 mmol/L (mean 0.74

mmol/L) for triacylglycerols. In men, fasting concentrations of serum lipids ranged from 4.02 to 6.60 mmol/L (mean 5.43 mmol/L) for total cholesterol, 0.77 to 1.65 mmol/L (mean 1.23 mmol/L) for HDL cholesterol, and 0.07 to 2.72 mmol/L (mean 1.05 mmol/L) for triacylglycerols.

### Design and statistical analyses

The trial, which was approved by the medical ethics committee of the University, had a parallel design. During the run-in period all volunteers consumed a diet high in oleic acid for 3 weeks. They were then divided into three groups. For the next 6 weeks (the test period), one group consumed the MCFA diet, a second group the myristic acid diet, and a third group continued on the oleic acid diet. The groups were stratified for initial serum cholesterol concentrations and sex. The response to the experimental diet was calculated per subject as the change from the end of the run-in period to the end of the MCFA, myristic acid, or oleic acid test period. Differences in response of lipid and lipoprotein concentrations were examined with diet, sex, and diet-and-sex interaction as independent variables. The data were analyzed with the General Linear Models (GLM) procedure of the SAS Program (12). When the analyses indicated a significant effect of diet ( $P < 0.05$ ), the diets were compared pair-wise. In addition, 95% confidence intervals were calculated for the differences among the diets. The results were corrected for 3-group comparisons by the Bonferroni correction. Because changes in lipoprotein[a] (Lp[a]) concentrations were not normally distributed, the untransformed individual changes of Lp[a] were analyzed with the non-parametric Kruskal-Wallis test (13). The a priori power to detect a true difference in total cholesterol concentrations of 8% between two diets was 80%.

### Diets

Before the study started, the subjects recorded their habitual food intake for 2 working days and 1 weekend day. From these food records, each subjects' actual energy intake was calculated by using the Dutch food-composition table (14). The study diets were formulated at 13 levels of energy ranging from 5 MJ to 22 MJ, so that each subject received a diet that met his or her energy needs. Diets used in the study consisted of products in which the normal fat was replaced by the experimental fats. The products included margarines and bakery products (bread, cookies, pies, and cakes). According to the subjects' energy level, lists were computed that stated the amount of experimental products the subject had to eat each day (margarine, bread, and cookies) and each week (cake and pie). These solid

foods supplied 63% (25 En%) of the total fat energy. The remaining 37% (15 En%) of the total fat intake had to be chosen from a list of "free-choice" fat-containing products. These products were given points according to their fat contents. One point was equal to 1 gram of fat. Each subject was required to eat a certain number of points daily, again corresponding with his or her energy intake, and to list the products chosen daily on a special form. The fat composition of the calculated diets was similar except for approximately 10% of total energy intake, which was provided by MCFA, myristic acid, or oleic acid, respectively.

Products were not labeled in the run-in period and were coded with a yellow, red, or blue label in the test period to blind the subjects as to the nature of the diets. Products were handed out on an individual basis and were free of charge. Subjects came at least once a week to the University to receive a new supply of products, to be weighed, and to receive new forms to list the free-choice items.

Subjects recorded their food intake on one weekend and 2 weekdays in the last week of both the run-in and the test period. They were also asked to maintain the same activity level, and smoking and drinking habits throughout the study. They recorded in diaries any signs of illness, medication used, alcohol consumption, menstrual cycle, and any deviations from the study protocol.

### Experimental fats

Special experimental fats were developed (Grünau GMBH, Illertissen, Germany). The fat high in MCFA was made by interesterification of 34.0% palm stearin, 17.6% high oleic sunflower oil, 8.4% sunflower oil, and 40% medium chain fatty acid (MCT) oil. The composition of the MCT oil was 33.3% C6:0, 25.3% C8:0, and 41.5% C10:0. The myristic acid fat was made by interesterification of a blend of 34% palm stearin, 17% high oleic sunflower oil, 9% sunflower oil, and 40% trimyristin. The oleic acid fat consisted of a blend of 30% palm stearin and 70% high oleic sunflower oil. Margarines were made from the fats and consisted of 83% fat and 17% water.

*sn*-2 fatty acid composition of the margarines was determined using the Grignard reagent allyl magnesium to partially deacylate triacylglycerols leading to a representative mixture of *sn*-2 monoacylglycerols (15). In addition, the fatty acid composition of total triacylglycerols was determined (see Table 1). The percent of each fatty acid at the *sn*-2 position was calculated from the total and the *sn*-2 fatty acid compositions. For example, the percent of myristic acid present at the *sn*-2 position in the experimental margarine as a percent of total proportion myristic acid is:

$$sn\text{-}2 \text{ myristic acid} / (3 \times \text{total myristic acid}) \times 100.$$

### Blood sampling and analyses

Blood was sampled after an overnight fast and after subjects abstained from drinking alcohol the preceding day and from smoking on the morning before blood sampling. Blood was sampled at the end of the run-in period (week 2 and week 3) and in weeks 6, 8, and 9 of the test period. Blood was drawn with minimum stasis using a 1.2-mm needle (Strauss Kanule, Luer, Wächtersbach, Germany) with the volunteer in a recumbent position. First, 12 ml blood was taken for additional measurements (to be reported elsewhere), while the last 10 ml blood was drawn into a 10-ml clotting-tube for serum lipid and lipoprotein measurements. All venipunctures were performed by the same person, at the same location, and for each subject generally at the same time of the same day of the week.

Total cholesterol (CHOD-PAP method; Monotest cholesterol, Boehringer Mannheim, Mannheim, Germany), HDL cholesterol (precipitation method; Monotest cholesterol, Boehringer Mannheim, Mannheim, Germany), and triacylglycerols (GPO-Trinder; Sigma Diagnostics, St. Louis, MO) were analyzed enzymatically. The coefficient of variation within runs was 1.0% for total cholesterol, 2.6% for HDL cholesterol, and 1.5% for triacylglycerols. LDL cholesterol was calculated using the Friedewald equation (16).

Apolipoprotein (apo) A-I and apoB were measured in serum by using an immunoturbidimetric reaction (UNI-KIT apoA-I and UNI-KIT apoB, Roche, Basel, Switzerland) and antiserum raised in sheep and rabbits, respectively. The coefficients of variation within runs were 1.1% for apoA-I and 1.5% for apoB.

Lipoprotein[a] (Lp[a]) was measured in serum by an enzyme-linked immunosorbent assay (ELISA) (Tint-ElyseLp[a], Biopool, Umea, Sweden). The Lp[a] from the sample was bound to the Lp[a] antibodies in the wells. Peroxidase-conjugated Lp[a]-antibodies were then used to tag the bound Lp[a]. After washing away unbound antibodies, the peroxidase substrates were added. The extinction of the yellow color that developed was directly proportional to the amount of Lp[a] present in the sample. The coefficient of variation was 6.3% within runs.

Serum was obtained by centrifugation at 2000 g (10 min, 4°C) 1 h after venipuncture and stored at -80°C. The lipid and lipoprotein concentrations obtained for the two sampling days at the end of the run-in (week 2 and week 3) and the test period (week 8 and week 9) were averaged for data analyses. Samples of apoA-I, apoB and Lp[a] from these two sampling days, however, were pooled and analyzed as such. All samples from one subject were analyzed within one run.

TABLE 1. Total and *sn*-2 fatty acid composition and the percentage of each fatty acid at the *sn*-2 position (%*sn*-2)<sup>a</sup> in triacylglycerols of the experimental margarines

Fatty Acid	MCFA Margarine			Myristic Acid Margarine			Oleic Acid Margarine		
	Total	<i>sn</i> -2	% <i>sn</i> -2	Total	<i>sn</i> -2	% <i>sn</i> -2	Total	<i>sn</i> -2	% <i>sn</i> -2
Saturated	76.7	65.1	28.3	64.4	67.8	35.1	23.6	8.4	11.9
MCFA	59.6	38.7	21.6	1.4	0.5	11.9	0.0	0.0	0.0
C12:0	0.2	0.4	66.7	0.5	0.7	46.7	0.1	0.1	33.3
C14:0	0.5	0.7	46.7	40.2	41.8	34.7	0.5	0.4	26.7
C16:0	14.3	22.1	51.5	18.6	20.7	37.1	17.5	7.1	13.5
C18:0	1.9	3.2	56.1	3.5	4.0	38.1	4.8	0.8	5.6
Monounsaturated	17.4	26.5	50.8	26.2	23.9	30.4	70.1	83.4	39.7
<i>cis</i> C18:1	16.9	26.6	52.5	25.6	23.9	31.1	69.9	83.4	39.8
Polyunsaturated	5.9	8.4	47.5	9.4	8.3	29.4	6.3	8.2	43.4
<i>cis,cis</i> C18:2	5.7	8.4	49.1	9.1	8.3	30.4	5.8	8.0	46.0

<sup>a</sup>Calculated as (*sn*-2 fatty acids/3 × total fatty acids) × 100. In random distributions this value would equal 33%.

Fatty acid composition of serum total lipids at the end of the run-in and at the end of the test period was determined in samples pooled according to diet and sex. Serum lipids were extracted with a modified Folch extraction (17), and transmethylated to their corresponding methyl esters (18), which were then quantified on a gas chromatograph, with a polar capillary column (CP Sil 88, Chrompack, Middelburg, The Netherlands) as described before (19).

## RESULTS

### Diets and dietary adherence

Table 1 shows the total and *sn*-2 fatty acid composition of the experimental margarines and the percentage of each fatty acid at the *sn*-2 position. In the MCFA margarine, 22% of MCFA was attached to the *sn*-2 position and the remaining 78% to either the *sn*-1 or the *sn*-3 position. Palmitic acid, oleic acid, and linoleic acid, the other major fatty acids in this fat, were mainly on the *sn*-2 position. In the myristic acid margarine, myristic acid and the other major fatty acids (palmitic, oleic, and linoleic acids) were equally distributed among each position of the glycerol molecule.

The composition of the diets, as determined by a 3 day food record, is given in Table 2. The average intake of oleic acid was 15.1 En% during the run-in period and decreased in the test period to 6.6 En% with the MCFA and to 6.3 En% with the myristic acid diet. It was exchanged for extra MCFA or myristic acid. Oleic acid intake during the oleic acid diet did not change from the run-in to the test period. Changes of other nutrients, except for En% saturated and monounsaturated fatty acids, did not significantly differ among the diets.

Dietary adherence was confirmed by determination

of serum total fatty acid composition. Serum fatty acids with the MCFA diet group showed minor increases in the proportions of total fatty acids in capric acid (from 0.1% to 0.2%), myristic acid (from 1.7% to 2.0%), and stearic acid (from 5.6% to 6.2%). With the myristic acid diet the proportion of myristic acid increased from 2.1% to 3.2% of total fatty acids. The proportion of oleic acid with the oleic acid diet was similar after the run-in and test period (23.0% and 24.0%) but decreased with both the MCFA (from 22.2% to 18.3%) and myristic acid diets (from 25.1% to 18.8%).

Records of "free choice" fat-containing items during the experiment showed minimum deviation from the prescribed grams of "free choice" fat (mean ± standard deviation: 0 ± 6%). The largest discrepancies were recorded during the first week of the run-in period, when the subjects had to familiarize themselves with the protocol.

Mean body weight (means ± standard deviation) in the MCFA group was 70 ± 10 kg after the run-in and 70 ± 10 kg after the MCFA diet, in the myristic acid group 72 ± 9 kg after the run-in and 72 ± 10 kg after the myristic acid diet, and in the oleic acid group 71 ± 12 kg after the run-in and 71 ± 12 kg after the oleic acid diet. Body weights within the diet groups did not differ from the run-in to the end of the test periods.

### Serum lipid and lipoproteins

Mean changes of serum total, LDL, and HDL cholesterol concentrations are given in Table 3. Serum total cholesterol concentration increased by 0.55 mmol/L when the subjects switched to the myristic acid diet. This change was significantly different from the change with the oleic acid diet (difference in changes of 0.40 mmol/L; *P* = 0.0057; 95% confidence interval (CI) 0.10 to 0.69 mmol/L). The difference in changes between the MCFA and the oleic acid diet group was 0.23



TABLE 2. Mean daily intake of nutrients as calculated from the recorded food intakes during 2 weekdays and 1 weekend day in the run-in and the test period with diets enriched in MCFA (n = 21), myristic acid (n = 20), or oleic acid (n = 19)

	MCFA Diet	Myristic Acid Diet	Oleic Acid Diet
<b>Energy (MJ/day)</b>			
Run-in	9.2 ± 3.0	11.0 ± 3.6	9.8 ± 2.2
Test period	9.1 ± 2.8	10.9 ± 2.9	9.5 ± 2.0
Change	-0.1 ± 1.1	-0.2 ± 1.6	-0.4 ± 1.4
<b>Fat (En%)</b>			
Run-in	38.7 ± 6.5	38.4 ± 4.6	39.7 ± 3.2
Test period	40.4 ± 3.7	39.7 ± 4.5	40.0 ± 3.9
Change	1.7 ± 5.1	1.3 ± 4.3	0.3 ± 4.1
<b>Saturates</b>			
Run-in	11.3 ± 2.0	11.3 ± 1.4	11.7 ± 1.4
Test period	21.3 ± 2.4	21.0 ± 2.2	11.6 ± 1.9
Change	10.0 ± 2.2	9.6 ± 2.1	-0.1 ± 2.0
<b>MCFA<sup>a</sup></b>			
Run-in	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
Test period	9.9 ± 1.9	0.0 ± 0.0	0.0 ± 0.1
Change	9.9 ± 1.9	0.0 ± 0.0	0.0 ± 0.1
<b>Myristic acid<sup>a</sup></b>			
Run-in	0.1 ± 0.2	0.1 ± 0.1	0.0 ± 0.0
Test period	0.0 ± 0.0	9.7 ± 1.6	0.1 ± 0.0
Change	-0.1 ± 0.1	9.6 ± 1.6	0.0 ± 0.0
<b>Oleic acid<sup>a</sup></b>			
Run-in	15.0 ± 4.2	14.8 ± 2.3	15.7 ± 1.6
Test period	6.6 ± 1.3	6.3 ± 1.0	16.7 ± 2.8
Change	-8.5 ± 4.0	-8.5 ± 2.1	0.9 ± 2.3
<b>Linoleic acid</b>			
Run-in	4.0 ± 0.8	4.0 ± 0.9	3.7 ± 0.5
Test period	4.0 ± 0.6	4.0 ± 0.8	4.0 ± 0.5
Change	0.0 ± 0.7	0.0 ± 0.8	0.3 ± 0.8
<b>Carbohydrates</b>			
Run-in	47.4 ± 5.5	46.4 ± 5.2	46.3 ± 4.0
Test period	44.0 ± 5.5	45.4 ± 5.6	44.6 ± 4.9
Change	-3.4 ± 4.9	-1.0 ± 5.5	-1.7 ± 4.3
<b>Protein</b>			
Run-in	11.8 ± 2.8	11.9 ± 1.3	12.4 ± 1.9
Test period	13.3 ± 3.8	12.2 ± 2.4	12.5 ± 2.6
Change	1.5 ± 2.2	0.3 ± 1.3	0.2 ± 2.1
<b>Alcohol (mg/mj)</b>			
Run-in	2.0 ± 2.2	3.1 ± 3.6	1.6 ± 2.6
Test period	2.2 ± 3.7	2.6 ± 4.0	2.8 ± 5.5
Change	0.2 ± 2.5	-0.5 ± 2.5	1.2 ± 3.9
<b>Cholesterol (mg/Mj)</b>			
Run-in	16.3 ± 4.1	14.4 ± 3.1	15.8 ± 5.0
Test period	17.0 ± 4.7	14.8 ± 5.8	14.3 ± 4.7
Change	0.7 ± 5.5	0.4 ± 5.3	-1.5 ± 4.4
<b>Fiber (g/Mj)</b>			
Run-in	2.4 ± 0.3	2.2 ± 0.4	2.4 ± 0.4
Test period	2.3 ± 0.3	2.5 ± 0.5	2.4 ± 0.5
Change	-0.1 ± 0.3	0.3 ± 0.5	-0.1 ± 0.7

Values are means ± standard deviations.

<sup>a</sup>As provided by the experimental fats only.

mmol/L ( $P = 0.0913$ ; 95% CI -0.06 to 0.52 mmol/L). LDL cholesterol concentrations were 0.37 mmol/L higher with the myristic acid diet compared with the oleic acid diet ( $P = 0.0064$ ; 95% CI 0.09 to 0.64 mmol/L). The change of LDL cholesterol with the MCFA diet did not significantly differ from the change with the oleic acid diet (difference in changes of 0.23 mmol/L;  $P = 0.0752$ ; 95% CI -0.04 to 0.50 mmol/L) and the change with the myristic acid diet (difference in changes of -0.14 mmol/L;  $P = 0.284$ ; 95% CI -0.41 to 0.13 mmol/L). The HDL cholesterol concentration was higher with the myristic acid diet compared with the MCFA diet (difference in changes of 0.11 mmol/L;  $P = 0.0086$ ; 95% CI 0.02 to 0.21 mmol/L), and compared with the oleic acid diet (difference in changes of 0.10 mmol/L;  $P = 0.0273$ ; 95% CI 0.00 to 0.19). The MCFA diet did not raise HDL cholesterol concentrations compared with the oleic acid diet. Although the change in triacylglycerol concentrations with the MCFA diet was greater compared with the myristic acid diet, effects were not significantly different (difference in changes of 0.19 mmol/L;  $P = 0.0873$ ; 95% CI -0.05 to 0.44 mmol/L). The total cholesterol to HDL ratio increased with the MCFA diet. However, this increase was not significantly different from the change on the other diets.

ApoA-I concentrations increased with the myristic acid diet compared with the MCFA diet (difference in changes of 119 mg/L;  $P = 0.0093$ ; 95% CI 22 to 216 mg/L) (Table 4). Changes in apoB concentrations were not significantly different among the three diets. ApoA-I to apoB ratios significantly decreased with the MCFA compared with the myristic (difference in changes of -0.15;  $P = 0.0127$ ; 95% CI -0.27 to -0.02) and the oleic acid diet (difference in changes of -0.14;  $P = 0.0197$ ; 95% CI -0.27 to -0.01).

Lp[a] concentrations decreased with the myristic acid diet compared with the oleic acid diet ( $P = 0.0083$  for difference in changes) (Table 5). Changes in Lp[a] concentrations with the MCFA diet did not differ from those with the myristic acid and the oleic acid diets. Changes in Lp[a] concentrations were more pronounced in subjects with higher Lp[a] values with the oleic acid run-in period.

For all variables, responses to the diets were similar in women and men.

## DISCUSSION

In this project we studied the effects of MCFA, myristic, and oleic acids on serum lipids and lipoproteins. Results of the food records, serum total fatty acid com-

TABLE 3. Serum lipids and lipoprotein concentrations on diets enriched in MCFA, myristic, or oleic acids

	MCFA Diet	Myristic Acid Diet	Oleic Acid Diet
Total cholesterol (mmol/L)			
Run-in	5.26 ± 0.58	5.15 ± 0.70	5.03 ± 0.88
Test period	5.65 ± 0.42	5.77 ± 0.88	5.19 ± 0.87
Change	0.39 ± 0.39 <sup>a,b</sup>	0.55 ± 0.51 <sup>a</sup>	0.16 ± 0.39 <sup>b</sup>
HDL cholesterol (mmol/L)			
Run-in	1.49 ± 0.32	1.52 ± 0.54	1.52 ± 0.32
Test period	1.51 ± 0.32	1.65 ± 0.60	1.55 ± 0.32
Change	0.02 ± 0.11 <sup>a</sup>	0.13 ± 0.16 <sup>b</sup>	0.04 ± 0.14 <sup>a,b</sup>
LDL cholesterol (mmol/L) <sup>c</sup>			
Run-in	3.35 ± 0.67	3.13 ± 0.76	3.07 ± 0.81
Test period	3.67 ± 0.63	3.59 ± 0.88	3.16 ± 0.83
Change	0.32 ± 0.36 <sup>a,b</sup>	0.46 ± 0.51 <sup>a</sup>	0.09 ± 0.36 <sup>b</sup>
Triacylglycerols (mmol/L)			
Run-in	0.92 ± 0.42	1.15 ± 0.81	0.98 ± 0.43
Test period	1.02 ± 0.56	1.11 ± 0.66	1.04 ± 0.49
Change	0.11 ± 0.29	-0.09 ± 0.47	0.06 ± 0.26
Total cholesterol:HDL cholesterol ratio			
Run-in	3.73 ± 1.09	3.84 ± 1.50	3.49 ± 1.14
Test period	3.95 ± 1.08	3.92 ± 1.63	3.48 ± 0.97
Change	0.21 ± 0.34	0.08 ± 0.48	0.00 ± 0.44

Values are means ± standard deviations.

<sup>a,b</sup>Values with different letter superscripts are significantly different, *P* < 0.02.

<sup>c</sup>Calculated with Friedewald equation.

TABLE 4. Apolipoprotein A-I and apolipoprotein B concentrations on diets enriched in MCFA, myristic, or oleic acids

	MCFA Diet	Myristic Acid Diet	Oleic Acid Diet
ApoA-I (mg/L)			
Run-in	1508 ± 265	1556 ± 354	1523 ± 226
Test period	1489 ± 217	1656 ± 396	1564 ± 239
Change	-19 ± 209 <sup>a</sup>	100 ± 115 <sup>b</sup>	41 ± 92 <sup>a,b</sup>
ApoB (mg/L)			
Run-in	898 ± 153	922 ± 165	859 ± 215
Test period	980 ± 134	998 ± 201	892 ± 217
Change	82 ± 106	77 ± 124	32 ± 70
ApoA-I:apoB ratio			
Run-in	1.74 ± 0.47	1.76 ± 0.59	1.89 ± 0.60
Test period	1.57 ± 0.42	1.74 ± 0.58	1.86 ± 0.56
Change	-0.17 ± 0.24 <sup>a</sup>	-0.02 ± 0.17 <sup>b</sup>	-0.03 ± 0.16 <sup>b</sup>

Values are means ± standard deviations.

<sup>a,b</sup>Values with different letter superscripts are significantly different, *P* < 0.02.

position, and the daily records of the “free choice” products used indicated good compliance to the strict protocol. The energy percentage of the studied fatty acids was planned to be the only difference among the diets. To achieve this, MCFA and myristic acid were given to the subjects with synthetic fats. As in dairy fat, MCFA from the MCFA fat were mainly attached to the *sn*-1 or *sn*-3 positions of the glycerol molecule. In the myristic acid fat, myristic acid and other saturates were equally distributed among the *sn*-2 and *sn*-1,3 positions. Myristic acid-rich fat sources as palm kernel or coconut fat have myristic acid mainly at the *sn*-1,3 (20), whereas dairy fat carries half of the total proportion of myristic acid at the *sn*-2 position (21). So far, dietary experiments comparing fats rich in palmitic acid (22, 23) and stearic acid (24) suggested that the positional distribution of these saturates did not affect lipid and lipoprotein concentrations. One small study reported that randomized butter oil was less hypercholesterolemic than a non-randomized butter oil diet (25). Although this

TABLE 5. Lipoprotein[a] concentrations on diets enriched in MCFA, myristic, or oleic acids

	MCFA Diet	Myristic Acid Diet	Oleic Acid Diet
Lipoprotein[a] (mg/L)			
Run-in	43 (0 to 677)	89 (1 to 691)	105 (0 to 767)
Test period	95 (3 to 597)	54 (2 to 689)	118 (0 to 782)
Change	0 (-297 to 95) <sup>a,b</sup>	-4 (-164 to 202) <sup>a</sup>	13 (-34 to 190) <sup>b</sup>

Values are given as median levels (ranges).

<sup>a,b</sup>Values with different letter superscripts are significantly different, *P* < 0.02.

may suggest that our results underestimate the cholesterol-raising effects of natural fats rich in MCFA and myristic acid, the results of that study (25) are difficult to interpret as only eight subjects were involved. Larger studies are necessary to examine whether the positional distribution of MCFA and myristic acid influences their cholesterolemic effects.

### Effects of MCFA

The exchange of oleic acid by MCFA increased serum total and LDL cholesterol concentrations. The responses, however, did not differ significantly from the responses in the oleic acid and myristic acid diet groups. Other studies have reported similar serum total or LDL cholesterol concentrations with MCFA and western experimental or western habitual diets (3–5). These latter diets were high in saturated fatty acids, which cause higher plasma cholesterol levels than high oleic acid diets (26). Therefore, these earlier studies can also be interpreted as demonstrating a moderate cholesterol-raising effect of MCFA compared with oleic acid. In agreement, a recent study (27) demonstrated plasma total and LDL cholesterol-raising effects of both diets rich in MCFA or palmitic acid compared with a diet rich in a high oleic acid sunflower oil.

Although the MCFA diet slightly increased triacylglycerol concentrations compared with myristic acid, triacylglycerol responses were not significantly different among the three diets. Other authors (3, 4) reported considerably elevated serum triacylglycerol concentrations with MCFA diets. In these studies (3, 4), however, diets provided 32 En% as MCFA, whereas in the present study only 10 En% MCFA was given. MCFA probably raise triacylglycerols in a dose-dependent manner as McGandy, Hegsted, and Myers (4) demonstrated that a diet with 18 En% MCFA caused a smaller increase of serum triacylglycerol concentrations than a diet with 32 En% MCFA. In agreement with our results, diets with 6 En% MCFA did not significantly increase serum triacylglycerol concentrations (5). However, in studies that applied diets with high energy percentages of MCFA (3, 27) increased triglyceride concentrations did not affect the composition of the VLDL particles (3) or the ratio of triglyceride to VLDL cholesterol concentrations (27).

Compared with the oleic acid diet, the MCFA diet did not change HDL cholesterol nor did it change apoA-I concentrations. Also Cater, Heller, and Denke (27) demonstrated similar HDL cholesterol concentrations on diets rich in MCFA or oleic acid. Diets enriched in carbohydrates, however, showed significantly decreased HDL cholesterol and apoA-I concentrations and similar LDL cholesterol concentrations compared with oleic acid (28). Thus, effects of MCFA on HDL cho-

lesterol and apoA-I concentrations are likely to differ from those of carbohydrates, but this hypothesis needs to be confirmed in studies that compare the effects of MCFA and carbohydrates side-by-side.

In addition to slightly raised LDL cholesterol concentrations after the MCFA diet, other potent lipid predictors of coronary heart disease (29, 30), the ratio of apoA-I to apoB and the total to HDL cholesterol ratio, were unfavorably changed by this diet, although the latter ratio was not significantly different among the three diets.

### Effects of myristic acid

Serum total cholesterol concentrations with the myristic acid diet increased with 0.55 mmol/L when 10 En% from oleic acid was replaced by myristic acid. This change was the result of an increase in both LDL and HDL cholesterol concentrations. Zock, de Vries, and Katan (10) found an increase in total cholesterol of 0.66 mmol/L when the intake of myristic acid increased by 10 En% at the expense of oleic acid. Changes are smaller than predicted from the regression equations of Hegsted et al. (8). This equation estimates that total cholesterol concentrations would increase with 2.19 mmol/L when 10 En% from oleic acid is replaced by myristic acid.

The HDL cholesterol-raising effect of myristic acid was also found by other investigators (10, 11). In our earlier study (31), a comparable increase of 10% was found when 8 En% of lauric acid was exchanged for oleic acid. Raised HDL cholesterol concentrations were also reported in studies on coconut versus palm oil (32) and in studies that specifically investigated mixtures of lauric and myristic acids versus palmitic or oleic acids (33, 34). In addition, higher HDL cholesterol concentrations on western diets compared with MCFA diets (3, 5) might also be due to increased intakes of lauric or myristic acids with the western diets. It is possible that a lower plasma cholesteryl ester transfer protein (CETP) activity may have contributed to the higher HDL cholesterol concentration on the myristic acid diet. An *in vitro* study indeed indicated a lower rate of CETP-mediated transfer of lauric- or myristic acid-rich cholesteryl esters from synthetic HDL compared with longer chain saturated fatty acids (35). The inverse relation between CETP activity and HDL cholesterol concentrations was also demonstrated in human studies comparing diets rich in *trans* fatty acids with those rich in linoleic or oleic acids (36, 37). However, as compared with a safflower oil diet, two myristic acid-rich diets, one high in butter fat and the other high in coconut fat, induced similar HDL cholesterol concentrations, but the cholesteryl ester transfer activity was increased on the butter and not on the coconut fat diet (38). Clearly,

more studies are needed to clarify the regulatory mechanism behind changes in HDL cholesterol concentrations.

Myristic acid decreased Lp[a] compared with oleic acid which emphasized that Lp[a] concentrations were regulated independently from LDL cholesterol concentrations. From another study (39) it was concluded that oleic acid raised Lp[a] levels relative to a mixture of saturated fatty acids, but not to stearic acid. In our previous study we did not observe decreased Lp[a] concentrations when comparing lauric or palmitic acids with oleic acid (31). The suggestion, however, that myristic acid in particular decreases Lp[a] concentrations is not supported by a recent study reporting similar Lp[a] concentrations after diets rich in myristic or palmitic acids (40).

To summarize, the MCFA diet increased total and LDL cholesterol slightly, though not significantly, but not HDL cholesterol concentrations compared with the oleic acid diet. The MCFA diet unfavorably affected the apoA-I to apoB ratio compared with both other diets. Myristic acid is a hypercholesterolemic saturated fatty acid and although responses on serum total cholesterol concentrations are smaller than has been suggested earlier (8). Part of the cholesterolemic effect of myristic acid is due to increased HDL cholesterol concentrations. ■■

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