Effect of Triacylglycerols Containing Medium- and Long-Chain Fatty Acids on Serum Triacylglycerol Levels and Body Fat in College Athletes

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(Received June 29, 2001)

Summary Triacylglycerols containing medium- and long-chain fatty acids (TML) have medium- and long-chain fatty acids in the same molecule. The effects of dietary TML on serum lipid levels and body fat were studied in six young men belonging to a university rowing club. A double-blind crossover study was performed in which for 3 wk the subjects ingested a liquid diet containing 20 g/d of soybean oil or TML in addition to their regular diets. Throughout the study, they were asked to keep their usual lifestyle, including diet and physical activity. The body composition of the subjects was measured weekly. Blood samples were taken at 0, 2, and 3 wk of each treatment period. There was no significant difference in energy intake between the soybean oil diet period and the TML diet period. The rate of variation of serum triacylglycerol concentration was significantly lower after a consumption of the TML liquid diet for 3 wk compared with the soybean oil liquid diet. The rate of variation of body fat mass was also significantly lower after a consumption of the TML liquid diet for 3 wk compared with the soybean oil liquid diet. However, the serum cholesterol concentration did not change significantly during either dietary treatment. These results suggest that TML, compared with soybean oil, may have the potential to prevent hypertriglyceridemia and obesity caused by consumption of a high-fat diet.

Key Words body fat accumulation, serum triacylglycerol, triacylglycerols containing medium- and long-chain fatty acids, high-fat diet, men

The effect of triacylglycerol on blood lipid concentration and body fat accumulation differs with the type of component fatty acids (1, 2). Medium-chain triacylglycerols (MCT), composed of 8–10 carbon-saturated fatty acids are easily degraded by β-oxidation in the liver and have been reported to decrease blood lipid concentration (3–5) and body fat accumulation (6–8). Ingested MCT is more easily degraded to fatty acids and glycerol by pancreatic lipase compared with long-chain triacylglycerols and is absorbed mainly from the small intestine. Most medium-chain fatty acids absorbed by the small intestinal mucosa are bound to albumin and transferred to the liver via the portal vein. Absorbed long-chain fatty acids form chylomicrons in the small intestine and flow into the venous system via lymphatic vessels. The long-chain fatty acids in chylomicrons are transported to peripheral tissues such as adipose tissue and muscle, showing a markedly different metabolism from that of medium-chain fatty acids (9). Medium-chain fatty acids transported to the liver are very unlikely to be used as materials for the synthesis of triacylglycerol and phospholipids in the hepatocyte cytoplasm. These fatty acids can be transported through the mitochondrial membrane without binding to carnitine and easily move into the mitochondria, where β-oxidation occurs (10).

Although the usefulness of MCT has already been shown and studied in detail, MCT are not generally used as dietary oils. One reason is the low smoking temperature of MCT, about 140°C, which is not appropriate for cooking with heat. The addition of a small amount of MCT to common dietary oils decreases the smoking temperature.

Triacylglycerols containing medium- and long-chain fatty acids (TML) have medium- and long-chain fatty acids in the same molecule and are made by transesterification of MCT with long-chain triacylglycerols. The smoking temperature of TML is higher than those of MCT and LCT mixtures, and TML is superior for cooking.

From the early 1980s, the safety of dietary TML and its effects on whole body lipid oxidation have been studied in patients. It has been shown that administration of TML via the central vein decreases plasma triacylglycerol concentration with no side effects (11) and increases the oxidation rate of body fat in postoperative patients (12). We have demonstrated that total energy expenditure was higher after TML ingestion than after LCT ingestion in healthy young women (13). Furthermore, we have observed that body fat accumulation was lower in rats fed a TML diet than in those fed

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a soybean oil diet (14). TML may have the potential for lowering the hyperlipidemia and obesity caused by the consumption of a high-fat diet in humans. The present study, therefore, was designed to determine the effect of dietary TML on serum lipid concentration and body fat accumulation in college athletes.

**MATERIALS AND METHODS**

**Subjects.** Six young men (aged 18–23 years) were recruited from the rowing club of the University of Tsukuba (Tsukuba, Japan) to participate in this study. All subjects lived in the rowing club house and kept their usual daily schedules. No subjects were taking alcoholic drinks or medication that affected body weight. The characteristics of the subjects are shown in Table 1. All procedures were approved in advance by the Ethical Committee of the Research Laboratory of Nisshin Oil Mills and were in accordance with the Helsinki Declaration of 1964, as revised in 1989. After a detailed explanation of this study, each subject gave written informed consent.

**Test oils.** Soybean oil, MCT, and rapeseed oil were purchased commercially (Nisshin Oil Mills, Tokyo). After 800 g of rapeseed oil and 200 g of medium-chain triacylglycerols were mixed, TML was prepared by transesterification, with sodium methoxide as a catalyst. The TML was bleached by adding activated clay and deodorized by steam distillation. The compositions of triacylglycerols and fatty acids of the test oils are given in Table 2.

**Experimental design.** A double-blind crossover trial was used to investigate the effect of dietary TML on serum triacylglycerol concentration and body fat accumulation. Body weight, height, body-fat percentage, energy intake, and serum lipid level were investigated before the beginning of the study. The subjects randomly interchanged two 3-wk treatment periods. The first was carried out from November 15 to December 6 in 1999. After a 9-wk washout period, the second treatment was carried out February 7 to 28, 2000. During the treatment period of 3 wk, the subjects were given a liquid diet containing 20 g of soybean oil or TML in addition to their regular diet. One package (200 g) of liquid diet containing 10 g of test oil was given with breakfast and dinner, respectively. Both liquid diets contained the following ingredients in grams per kilogram: sucrose, 12.5; dextrin, 125; sodium caseinate, 66; fat (soybean oil or TML), 50; flavor and emulsifier, 8; and water, 738.5. The energy of two packages of liquid diet was 506 kcal. The subjects were asked to keep their usual daily schedules and maintain their usual lifestyle, including diet and physical activity, throughout the study. They got up at 5:00 a.m. and had rowing practice in the morning (5:30–7:00) and in the afternoon (17:30–19:00) from Tuesday to Sunday. The subjects were asked to record the contents of daily meals, snacks, and beverages in the diet diary for the entire test period.

The body composition of subjects was measured once a week from 15:00 p.m. to 17:00 p.m. on Monday when the subjects had no rowing practice. The subjects' height and weight, from which body mass index was calculated, were measured by conventional methods. Body fat mass was determined by a bioelectrical impedance analyzer (Business Model TBF-102; Tanita, Tokyo) in duplicate. Blood samples were taken on the Monday morning (8:00–9:00) at 0, 2, and 3 wk of each treatment period after a 12 h overnight fast. The concentrations of serum triacylglycerol, serum cholesterol, plasma glucose, serum insulin, serum acetocetatic acid, serum 3-hydroxybutyric acid, serum total ketone bodies, serum nonesterified fatty acids, and serum glycerol were determined by Scripps Reference Laboratory (SRL, Tokyo). Daily food intake was determined from questionnaires completed by the subjects. Food consumption was calculated by the Statistical Package for

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**Table 1. Characteristics of subjects before the experiment.**

| Characteristic | 6 |
|----------------|---|
| Number         | Male |
| Sex            | 20±1 |
| Age (y)        | 174±2 |
| Height (cm)    | 67.1±3.7 |
| Body weight (kg) | 22.0±0.6 |
| Body-fat percentage (%) | 20.7±1.7 |
| Body-fat mass (kg) | 14.2±1.9 |
| Energy intake (kcal/d) | 3,107±283 |
| Serum triacylglycerol concentration (mmol/L) | 0.95±0.18 |
| Serum cholesterol concentration (mmol/L) | 3.89±0.20 |

1 Mean±SE.

**Table 2. Triacylglycerol and fatty acid compositions of the test oils.**

| Test oil | Soybean oil | TML |
|----------|-------------|-----|
| Triacylglycerol composition (g/100 g) | | |
| L, L, L | 100 | 38.4 |
| L, L, M | — | 44.2 |
| L, M, M | — | 15.9 |
| M, M, M | — | 1.5 |

Fatty acid composition (g/100 g)

| 8:0 | 14.4 |
| 10:0 | 4.8 |
| 16:0 | 3.2 |
| 18:0 | 1.6 |
| 18:1 | 49.2 |
| 18:2 | 17.9 |
| 18:3 | 8.9 |
| Others | 0.3 | — |

1 Not detected.
Table 3. Energy and lipid intakes during consumption of soybean oil or triacylglycerols containing medium- and long-chain fatty acids (TML).

|                        | 1 wk        | 2 wk        | 3 wk        | Average     |
|------------------------|-------------|-------------|-------------|-------------|
| Energy intake (kcal/d) |             |             |             |             |
| Soybean oil diet       | 3,853±215   | 3,398±145   | 3,543±244   | 3,598±182   |
| TML diet               | 3,679±83    | 3,435±231   | 3,574±196   | 3,563±135   |
| Lipid intake (g/d)     |             |             |             |             |
| Soybean oil diet       | 148±10      | 126±10      | 136±13      | 137±9       |
| TML diet               | 133±3       | 114±8       | 133±10      | 127±5       |

Values are means±SE.

Table 4. Body weight and body-mass index before and after consumption of soybean oil or triacylglycerols containing medium- and long-chain fatty acids (TML).

|                        | 0 wk        | 1 wk        | 2 wk        | 3 wk        |
|------------------------|-------------|-------------|-------------|-------------|
| Body weight (kg)       |             |             |             |             |
| Soybean oil diet       | 67.3±3.7    | 67.7±3.6    | 67.8±3.7    | 68.2±3.8    |
| TML diet               | 66.6±3.1    | 67.1±3.4    | 67.7±3.5    | 67.2±3.5    |
| Body-mass index (kg/m²)|             |             |             |             |
| Soybean oil diet       | 22.0±0.7    | 22.2±0.6    | 22.2±0.7    | 22.3±0.7    |
| TML diet               | 21.9±0.4    | 22.0±0.5    | 22.2±0.6    | 22.0±0.6    |

Values are means±SE.

Windows (Healthy Diet; Tokyo Shoseki, Tokyo).

Statistical analysis. Data were expressed as means±SE and analyzed by Duncan’s multiple-range test following ANOVA by the use of a commercial software package (EXCEL TOUKEI version 3.0 for Windows; Esumi, Tokyo). Differences with p<0.05 were considered significant.

RESULTS

Energy intake, lipid intake, body weight, and body mass index

The energy and lipid intakes during the consumption of the liquid diets containing soybean oil or TML are shown in Table 3. No significant differences in energy and lipid intakes were found during the consumption of either soybean oil liquid diet or TML liquid diet. The liquid diets had no effect on body weight or body mass index (Table 4).

Serum lipids and body fat

The changes in rate of variation of serum triacylglycerol concentration, cholesterol concentration, and body-fat mass during the consumption of the soybean oil and TML liquid diets are shown in Fig. 1. The serum triacylglycerol concentration at 0 wk did not differ significantly between treatments: 0.82±0.12 and 0.85±0.11 mmol/L for the soybean oil and TML, respectively. The serum cholesterol concentration at 0 wk did not differ significantly between treatments: 4.03±0.27 and 4.11±0.17 mmol/L for the soybean oil and TML, respectively. The rate of variation of serum triacylglycerol concentration was significantly lower during the consumption of the TML liquid diet for 3 wk compared with the soybean oil liquid diet for 3 wk. However, serum cholesterol concentration did not change significantly during either the soybean oil or TML diet periods.

The body-fat mass at 0 wk did not differ significantly between treatments: 12.9±1.7 and 13.3±1.8 kg for the soybean oil and TML, respectively. The rate of variation of body-fat mass was significantly lower during the consumption of the TML liquid diet for 3 wk compared with the soybean liquid diet for 3 wk.

Results of plasma and serum analyses

The results of plasma and serum analyses before and after consumption of the liquid diets containing soybean oil or TML are shown in Table 5. Neither liquid diet had significant effects on the concentrations of plasma glucose, serum insulin, serum acetoacetic acid, serum 3-hydroxybutyric acid, or serum total ketone bodies. The activities of serum GOT and GPT remained constant throughout both experimental periods. The concentrations of serum nonesterified fatty acids and glycerol did not change significantly during the study.

DISCUSSION

In this study, the effects of TML, which is more suitable for cooking with heat than MCT is, on serum lipid concentration and body fat accumulation were investigated in college athletes. For 3 wk the subjects ingested a liquid diet containing 20 g/d of soybean oil or TML in addition to their regular diet. The serum triacylglycerol level was significantly increased by the ingestion of soybean oil, but no significant changes from the ingestion of TML were observed. The rate of variation of serum triacylglycerol level after 3 wk of TML diet was significantly lower compared with the soybean diet. This finding suggests that the effect of TML on serum triacylglycerol level is similar to that of MCT rather than to that of
Fig. 1. Changes in rate of variation of serum triacylglycerol concentration (A), serum cholesterol concentration (B), and body-fat mass (C) before and after the consumption of soybean oil or triacylglycerols containing medium- and long-chain fatty acids (TML). Six young men consumed liquid diets containing 20 g/d of soybean oil (●) or TML (□) for 3 wk in a double-blind crossover design. Values are means±SE. Data were analyzed by Duncan’s multiple-range test following ANOVA. Values with different superscripts are significantly different (p<0.05).

Soybean oil, which consists of long-chain triacylglycerols. However, the serum cholesterol level did not differ significantly between the soybean oil diet and TML diet periods. This finding suggests that the effects of TML on serum triacylglycerol and cholesterol levels are different. The mechanism by which TML inhibits the elevation of serum triacylglycerol level is unknown, but it might be related to differences in chylomicron formation in the small intestine (9) and β-oxidation in the liver (15).

We recently observed that body-fat accumulation was significantly lower in rats fed TML than in those fed soybean oil (14). In this study of college athletes, the rate of variation of body-fat mass after 3 wk of TML diet was significantly lower than after the soybean diet period. Energy intake did not differ significantly between the soybean diet and TML diet periods. These findings suggest that TML is less likely to accumulate as body fat than soybean oil is, not only in laboratory animals, but also in humans. We have reported that diet-induced thermogenesis after the consumption of TML was greater than that after the consumption of LCT in young women (13). The intake of TML diet, compared with the intake of soybean oil diet, might increase diet-induced thermogenesis, resulting in less body-fat accumulation, because diet-induced thermogenesis plays an important role in the regulation of energy balance (16, 17). There is currently great interest in the regulation mechanism of body-fat accumulation by leptin (18) and uncoupling proteins (19, 20). The mechanism of less body-fat accumulation in subjects ingesting a TML diet might be related to the regulation by these factors. To clarify the participation of leptin and uncoupling proteins, much more study is needed.

In this study, body-fat mass was determined by the bioelectrical impedance method. There was a significant positive correlation between percent of body fat determined by the bioelectrical impedance method and the underwater weighing method (r=0.885, p<0.001) (21). Lakaski et al. studied the reliability of bioelectrical impedance method in 14 men on five successive days (22). The individual coefficients of variation ranged from 0.9–3.4%, and the average precision was 2%. The test-retest correlation coefficient was 0.99, and the reliability coefficient for a single measurement over 5 d was 0.99. The bioelectrical impedance method is noninvasive and convenient for the participants and is a useful and reproducible method of measuring body-fat percentage (21–24). However, it has the disadvantage that the results vary because of differences in body sizes and times of measurement. The variations resulting from differences in body sizes could be suppressed by using the crossover method. The measurement was performed at a fixed time to decrease variation in this study.

In a single oral dose study (13), serum insulin and ketone body levels increased significantly at 2 h after an administration of TML compared with soybean oil. In this study, serum glucose, plasma insulin, and plasma ketone body levels did not differ between the soybean oil
and TML liquid diets. The increases in the serum insulin and ketone body levels caused by TML may have been transient effects. Medium-chain fatty acids absorbed from the small intestine are transported to the liver via the portal vein. If these fatty acids accumulate without being β-oxidized, fatty liver and hepatopathy may occur (25). Serum GOT and GPT activities, indices of hepatopathy (26), were not increased by an ingestion of the TML diet. This finding excludes the possibility of TML-induced hepatopathy and demonstrates the safety of TML in men. As an index of lipolysis, the serum levels of nonesterified fatty acids and glycerol were measured. No significant differences were found in the serum levels of nonesterified fatty acids or glycerol between the different types of liquid diets.

In this study, the liquid diet containing 20 g of soybean or TML was added to the regular diet for 3 wk, and the effects of TML on the serum triacylglycerol concentration and body-fat accumulation were investigated in a double-blind crossover study. The serum triacylglycerol concentration and body fat were less increased by the intake of TML diet than by the intake of soybean oil diet. These results suggest that TML, compared to soybean oil, may have the potential to prevent the hypertriglyceridemia and obesity resulting from a high-fat diet. The subjects ingested a liquid diets containing 506 kcal/d in addition to their regular diets for 3 wk. However, the liquid diets had no effect on body weight. The reason of this discrepancy is not clear. Another study that used more subjects and a stricter method of body fat analysis is needed to clarify the preventive effect of TML against hypertriglyceridemia and obesity.

Acknowledgments
We thank Mr. T. Nagasawa, Mr. I. Hidaka, Mr. A. Nagatoishi, Ms. T. Ito, Mr. O. Noguchi, Ms. M. Itakura, and Ms. E Kubota for their assistance in preparing the triacylglycerols containing medium- and long-chain fatty acids and the liquid diets.

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