The Thermic Effect Is Greater for Structured Medium- and Long-Chain Triacylglycerols Versus Long-Chain Triacylglycerols in Healthy Young Women

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The purpose of this study was to investigate the hypothesis that a single dose of structured medium- and long-chain triacylglycerols (SMLCTs) composed of medium-chain (20%) and long-chain (80%) fatty acids would increase the metabolic rate more than a dose of long-chain triacylglycerols (LCTs) in 15 healthy young women aged 18 to 28 years. The effects on postingestive energy expenditure were compared for SMLCTs versus LCTs. On the experimental days, the subjects fasted overnight and then ingested 1,680 kJ SMLCTs or LCTs each day. Energy expenditure and the respiratory quotient (RQ) were measured before and after SMLCT and LCT ingestion by indirect calorimetry. Blood samples were collected before and after ingestion to obtain plasma and serum. Postingestive total energy expenditure (PTEE) was significantly higher after SMLCT ingestion versus LCT ingestion (26.9 ± 1.0 *v* 25.5 ± 1.1 kJ/kg/6 h, $P < .05$). The thermic effects of the test oil were also significantly greater after SMLCT ingestion compared with LCT ingestion (3.02 ± 0.49 *v* 1.47 ± 0.82 kJ/kg/6 h, $P < .01$). Plasma glucose and serum triacylglycerol concentrations were not significantly different. Serum free fatty acid and 3-hydroxybutyric acid concentrations were higher after SMLCT ingestion versus LCT ingestion. These results suggest that long-term substitution of SMLCTs for LCTs will produce body fat loss if energy intake remains constant.

OBESITY IS CHARACTERIZED by an increase in lipid stores. It is generally associated with enhanced lipid consumption, which contributes to its development.[1,2] In Western countries, obesity is an important health problem affecting a large proportion of individuals who seek to prevent further weight gain or decide to counteract the detrimental health consequences of obesity.[3] To attain these objectives, patients use a wide variety of preventive or therapeutic methods alone or in combination. Among these approaches, dietary restrictions involving lipids are considered most important. The bulk of fatty acids found in normal Western diets consist of molecules comprising 12 or more carbon atoms. These long-chain fatty acids (LCFAs), either saturated or unsaturated, originate from the long-chain triacylglycerols (LCTs) provided by vegetable and/or animal oil and fat sources. They contribute to the supply of energy and fulfill essential fatty acid requirements.[4]

In contrast, medium-chain triacylglycerols (MCTs) are edible oils composed of triacylglycerols with saturated medium-chain fatty acid (MCFA) moieties of 6 to 10 carbon atoms. These were introduced to clinical nutrition in the 1950s for dietary treatment of malabsorption syndromes because of their rapid absorption and solubility.[5] MCTs and LCTs are metabolized differently. MCTs are transported to the liver directly via the hepatic portal circulation and are oxidized to ketones, whereas LCTs are absorbed via the intestinal lymphatic ducts and transported in chylomicrons through the thoracic duct to reach the systemic circulation.[6,7]

In animal studies, rats fed MCTs do not gain as much weight as rats fed an isocaloric amount of LCTs.[6-9] They show a diminished fat deposition[7,8] and an increased resting metabolic rate (RMR).[8-10] In a clinical study, Seaton et al[5] reported that mean postprandial oxygen consumption was higher after a MCT meal versus a LCT meal. These results suggest that MCTs could be useful in the dietary treatment of obesity. However, it is difficult to substitute MCTs for LCTs in dietary fat for long-term dietary therapy, largely because the lower smoke point makes MCTs difficult to use as cooking oil.[11]

Recently, we invented a new type of cooking oil composed of structured medium- and long-chain triacylglycerols (SMLCTs).[11] SMLCTs are structured lipids that contain MCFA and LCFA in the same triacylglycerol. They are made by transesterification of MCT and LCT. SMLCTs are superior for cooking versus the physical mixtures of MCTs and LCTs because the smoke point of the former is higher than that of the latter. If SMLCTs are similar biochemically and physiologically to MCTs, SMLCTs could be used in special cooking oils for dietary therapy.

The purpose of this study was to investigate the hypothesis that a single dose of SMLCTs will increase the metabolic rate more than a dose of LCTs in healthy young women.

SUBJECTS AND METHODS

Subjects

Fifteen young Japanese women (aged 18 to 28 years) who did not customarily exercise daily were recruited from Sanyo Women's College and Hiroshima University of Economics (Hiroshima, Japan) to participate in the study. All procedures were approved in advance by the Human Use Committee of Sanyo Women's College and are in accordance with the Helsinki Declaration of 1975, as revised in 1983. After a detailed explanation of the study, each subject provided informed written consent. Subjects were examined and found to be free of disease before the study. The physical characteristics of the subjects are shown in Table 1. The height and weight, from which the body mass index (BMI) was calculated as weight in kilograms divided by height in meters squared, were measured by conventional methods. The percent body fat and fat-free mass (FFM) were measured with a bioelectric impedance analyzer (model TBF-102; Tanita, Tokyo, Japan). All subjects had a normal menstrual cycle of 28 to 32 days. The

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Table 1. Characteristics of the Subjects

Subject No.	Age (yr)	Height (cm)	Weight (kg)	BMI (kg/m^2)	Body Fat (%)	FFM (kg)
1	20	161.0	56.6	21.8	29.2	40.1
2	20	163.5	64.1	24.0	30.4	44.6
3	28	156.0	49.5	20.3	21.8	38.7
4	18	155.0	46.4	19.3	26.0	34.3
5	19	159.0	52.5	20.8	27.0	38.3
6	19	160.0	55.0	21.5	30.6	38.2
7	20	163.0	50.7	19.1	23.3	38.9
8	18	157.0	53.7	21.8	33.9	35.5
9	18	159.0	43.3	17.1	19.0	35.1
10	18	166.0	56.4	20.5	25.6	42.0
11	18	158.0	49.4	19.8	20.9	39.1
12	18	159.0	50.2	19.9	21.6	39.4
13	18	162.0	47.2	18.0	19.3	38.1
14	20	164.0	52.0	19.3	24.3	39.4
15	19	149.0	44.6	20.1	25.2	33.4
Mean	19.4	159.4	51.4	20.2	25.2	38.3
SD	2.5	4.2	5.3	1.7	4.4	2.9

phase of the menstrual cycle was determined as described previously[12] (follicular phase, days 6 to 10; luteal phase, days 21 to 25).

Test Oils

LCTs (soybean oil), MCTs, and rapeseed oil were purchased commercially (Nisshin Oil Mills, Tokyo, Japan). SMLCTs were prepared by transesterification of MCTs and rapeseed oil (Fig 1). The presence of residual monoacylglycerols and free fatty acids was less than 1% of SMLCTs. The composition of fatty acids and triacylglycerols of the test oils are shown in Tables 2 and 3. SMLCTs contain about 20% MCFAs. The smoke points of MCTs, rapeseed oil, physical mixtures of MCTs and rapeseed oil, and SMLCTs were 143°, 230°, 160°, and 210°C, respectively.

Experimental Design

During the period of study, each subject maintained a normal lifestyle and ate ad libitum except for the day before the experiment. On that day, each subject ate the same supper (50 kJ/kg body weight) at 7:00 PM. The subjects fasted overnight at their homes and were brought

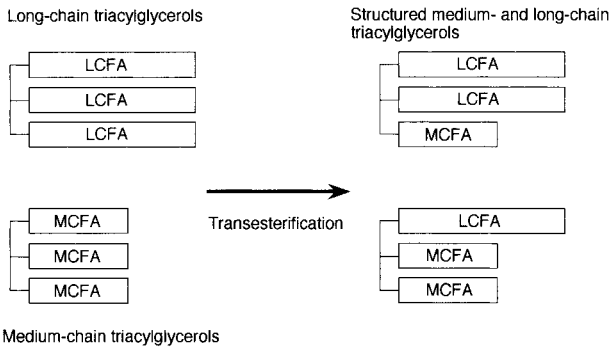

Fig 1. Preparation of structured medium- and long-chain triacylglycerols (SMLCTs). After mixing 800 g rapeseed oil and 200 g medium-chain triacylglycerols, SMLCTs were prepared by transesterification using sodium methoxide as a catalyst. SMLCTs were bleached by activated clay and deodorized by steam distillation.

Table 2. Fatty Acid Composition of the Test Oils (g/100 g)

Fatty Acid	Soybean Oil	SMLCT
8:0	—	13.7
10:0	—	4.7
16:0	10.4	3.6
16:1 (n-9)	0.1	0.2
18:0	4.0	1.8
18:1 (n-9)	23.9	50.1
18:2 (n-6)	52.9	16.1
18:3 (n-3)	7.8	7.4
20:0	0.3	0.5
20:1 (n-9)	0.2	1.1
22:0	0.4	0.3
22:1 (n-9)	—	0.3
24:0	—	0.1
24:1 (n-9)	—	0.1
Total	100.0	100.0

by car to the laboratory at 8:00 AM, where they rested until the start of the experiment at 9:00 AM. All experiments were performed in the preovulatory phase on day 8 to day 12 after the onset of menstruation.[13] The experimental sessions were divided into two types, SMLCT and LCT ingestion. There were at least 2 days between sessions, and the study was performed in a randomized order. The energy content of the test oils was 39.0 and 39.4 kJ/g for SMLCT and LCT, respectively.

On the days of the experiments, the subjects ingested 1,680 kJ test oil at 9:30 AM. They then rested for 6 hours (9:30 AM to 3:30 PM). While resting, the oxygen consumption and nonprotein respiratory quotient (RQ) were measured (9:00 AM to 3:30 PM). Blood samples were collected from the cephalic vein in the forearm to obtain serum and plasma at 9:30, 10:30, and 11:30 AM and 12:30, 1:30, and 3:30 PM. All procedures were performed in the laboratory under the same conditions (temperature 22° ± 1°C and humidity 60%).

Measurements

To measure the oxygen uptake and RQ, the subjects wore a face mask (Takei, Tokyo, Japan) continuously from 30 minutes before ingestion of the test oil to 60 minutes after ingestion and for 30 min/h for the next 5 hours. All expired gas was collected in a Douglas bag (Takei) and the bag was changed every 15 minutes while the subjects were resting. The concentrations of oxygen and carbon dioxide in the expired collected gas were immediately analyzed by an oxygen analyzer (scale range, 0.00% to 25.00%, model RAS-30; AIC, Tokyo, Japan) and a carbon dioxide analyzer (scale range, 0.00% to 6.00%, model RAS-31; AIC). The analyzers were calibrated with dried standard gas mixture and dried filtered fresh air. In the course of the measurements, the span of the analyzers was controlled once every 60 minutes. The accuracy of the measurements was ±0.03% of full scale for oxygen analysis and ±0.01% of full scale for carbon dioxide

Table 3. Triacylglycerol Composition of the Test Oils (g/100 g)

Components	Soybean Oil	SMLCT
L,L,L	100.0	38.4
L,L,M	—	44.2
L,M,M	—	15.9
M,M,M	—	1.5
Total	100.0	100.0

Abbreviations: L, LCFAS; M, MCFAS.

analysis. The RQ was calculated from oxygen consumption, carbon dioxide production, and urinary nitrogen loss at standard temperature and pressure–dry as an index of fat utilization.[14] Energy expenditure was calculated by an equation described previously,[14] metabolic rate (kJ/min) = $4.184 \cdot ([4.686 + 1.096 \cdot (RQ - 0.707)] \cdot V_{O_{2\,np}} + 4.60 \cdot V_{O_{2p}})$, where np is nonprotein, p is protein, and V_{O_2} is oxygen consumption. The thermic effect of the test oils was determined by the RMR, which was used as the baseline, and postingestive total energy expenditure per 6 hours (PTEE) using the formula, PTEE − RMR · 6 h.

Plasma glucose, serum immunoreactive insulin, triacylglycerol, free fatty acid, glycerol, and 3-hydroxybutyrate were determined by methods reported previously.[15-20]

Statistical Analysis

Statistical analysis was performed using a personal computer (Power Macintosh G3 400; Apple Japan, Tokyo, Japan) with a statistical program package (StatView; SAS Institute, Cary, NC). The statistical significance of differences between SMLCT and LCT treatments was tested by Student's paired *t* test with a confidence level of 95%. The differences in data over time with SMLCT or LCT treatment were not analyzed. All descriptive statistics were computed as the mean ± SEM.

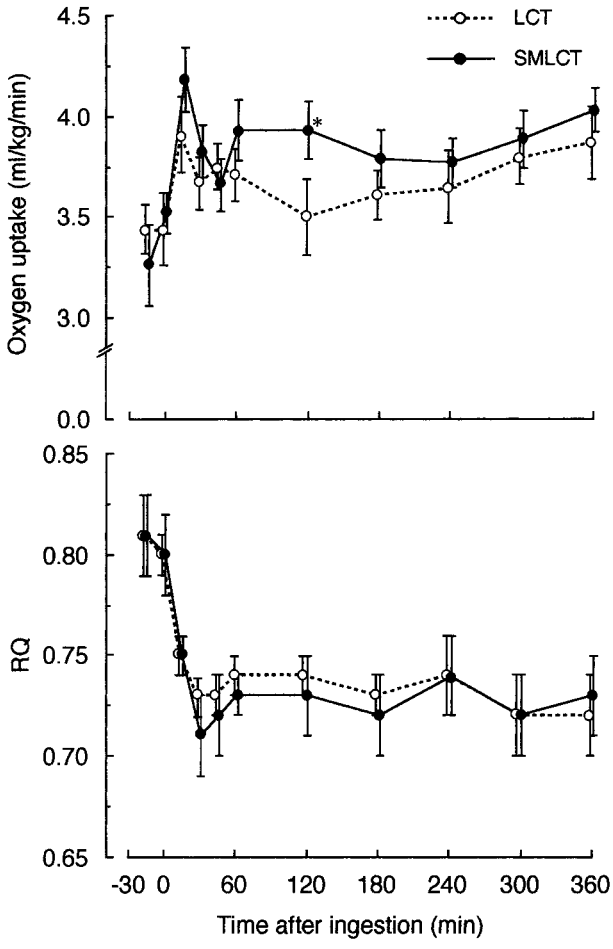

Fig 2. Oxygen consumption and RQ after ingestion of 400 kcal LCTs (○) or SMLCTs (●). Oxygen consumption and carbon dioxide production were measured for each session, and the RQ was calculated from these values. Each point represents the mean ± SEM for 15 subjects. *$P < .05$ *v* LCT ingestion, Student's paired *t* test.

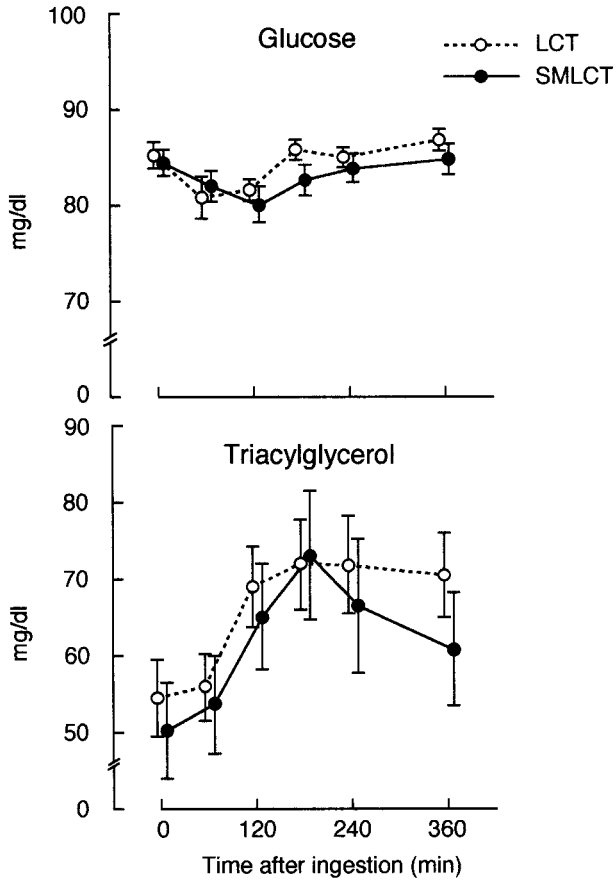

Fig 3. Plasma glucose and serum triacylglycerol after ingestion of 400 kcal LCTs (○) or SMLCTs (●). Each point represents the mean ± SEM for 15 subjects.

RESULTS

Oxygen Consumption and RQ

Oxygen consumption and the RQ at rest were measured for 6.5 hours to assess the thermic effect of test oil ingestion (Fig 2). The mean baseline oxygen consumption and RMR were identical before each test ingestion (3.46 ± 0.15 mL/kg/min, 66.8 ± 1.9 J/kg/min). Oxygen consumption tended to increase more after SMLCT ingestion than after LCT ingestion during the 6-hour experimental period. The high response to SMLCTs at 120 minutes was especially significant ($P < .05$). The RQ decreased immediately after each test oil ingestion. Postprandial RQs were lower with SMLCTs versus LCTs at 30 to 240 minutes, but the differences were not significant. PTEE was significantly higher after SMLCT ingestion versus LCT ingestion (26.9 ± 1.0 *v* 25.5 ± 1.1 kJ/kg/6 h, $P < .05$). The thermic effect of the test oil was also significantly higher after SMLCT ingestion versus LCT ingestion (3.02 ± 0.49 *v* 1.47 ± 0.82 kJ/kg/6 h, $P < .01$) (Fig 2).

Substrate Concentrations in Serum and Plasma

Plasma glucose concentrations did not change, but serum triacylglycerol increased after both SMLCT and LCT ingestion.

Serum triacylglycerol tended to decrease 150 to 360 minutes after SMLCT ingestion, but did not change after LCT ingestion. The differences in glucose and triacylglycerol concentrations were negligible (Fig 3).

Small increases in serum insulin were found after SMLCT (0 to 120 minutes) and LCT (0 to 60 minutes) ingestion. The insulinemic response to SMLCTs at 120 minutes was significantly higher ($P < .05$). Serum glycerol decreased 60 to 150 minutes after SMLCT ingestion, but did not change after LCT ingestion. Glycerol concentrations were significantly lower with SMLCTs at 60, 120, and 150 minutes ($P < .05$) (Fig 4).

Serum free fatty acid and 3-hydroxybutyrate concentrations increased after ingestion of each test oil. The mean values for serum free fatty acids were higher after SMLCT ingestion, with a significant difference at 120 minutes ($P < .05$). The mean 3-hydroxybutyrate concentrations were also higher after SMLCT ingestion, with a significant difference at 60 to 240 minutes ($P < .01$ to $.05$) (Fig 5).

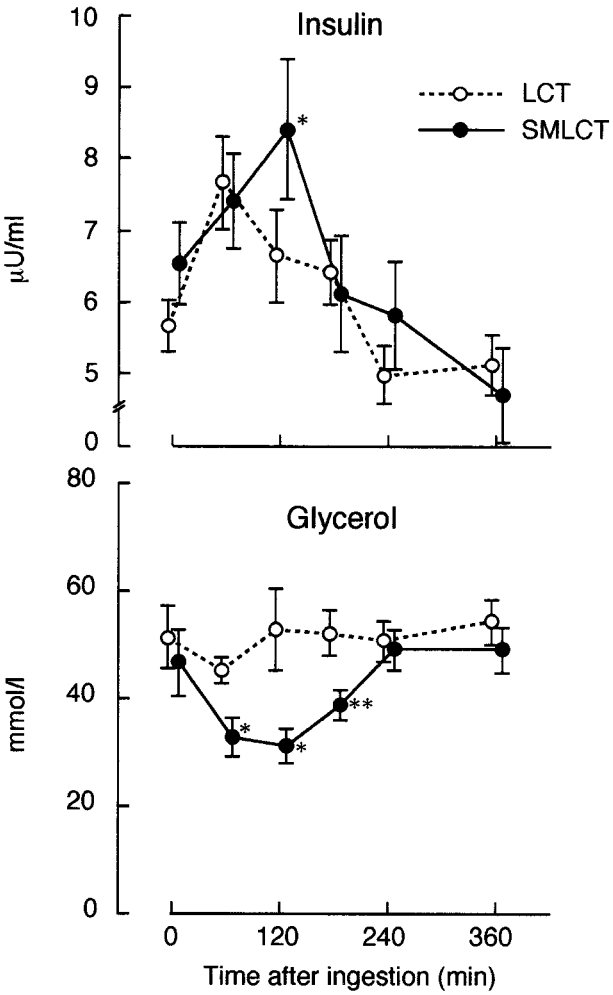

Fig 4. Serum insulin and glycerol after ingestion of 400 kcal LCTs (○) or SMLCTs (●). Each point represents the mean ± SEM for 15 subjects. **$P < .01$, *$P < .05$ *v* LCT ingestion, Student's paired *t* test.

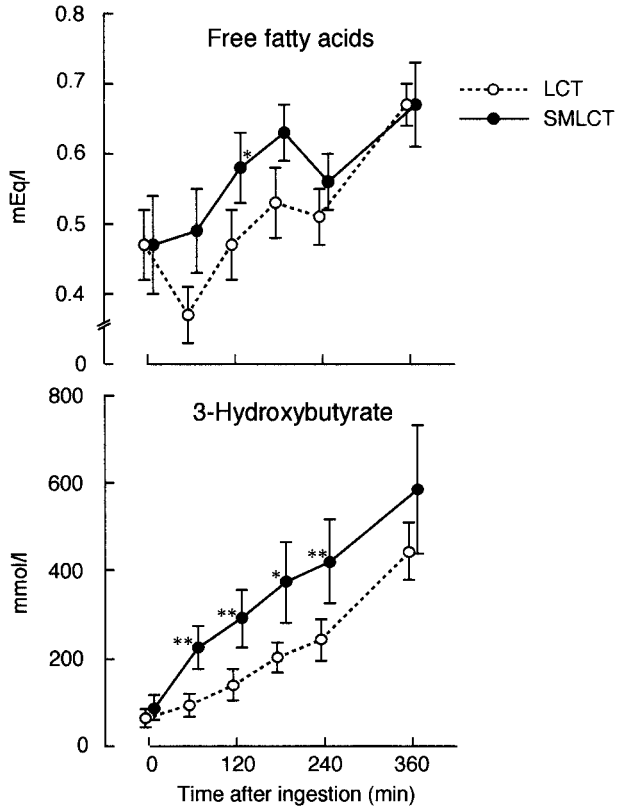

Fig 5. Serum free fatty acids and 3-hydroxybutyrate after ingestion of 400 kcal LCTs (○) or SMLCTs (●). Each point represents the mean ± SEM for 15 subjects. **$P < .01$, *$P < .05$ *v* LCT ingestion, Student's paired *t* test.

DISCUSSION

This study shows a mean 9% increase above the RMR over 6 hours following SMLCT ingestion. This increase is equivalent to 9.3% of the energy contained in the SMLCTs. The small increase in the metabolic rate after LCT ingestion is equivalent to 4.4% of the energy contained in the oil.

According to some reports, MCT and LCT produce similar thermic effects,[21,22] while other studies have found that MCT has a greater effect than LCT.[5,23-25] Flatt et al[21] compared the effect of a 3,595-kJ test meal containing 40 g MCTs versus 40 g LCTs over 9 hours. Energy expenditure due to consumption of the test meals was similar and equivalent to 11.2% and 12.5% of the energy contained in the LCT and MCT meals, respectively. Conversely, Scalfi et al[23] examined the diet-induced thermogenesis response to the consumption of a 5,447-kJ test meal containing 30 g MCTs or LCTs in lean subjects. Total energy expenditure increased and the RQ decreased after the MCT test meal, resulting in a significantly elevated thermogenic response. Hill et al[24] reported that the thermic response to ingestion of a 4,190-kJ test meal containing 40% MCTs was significantly higher compared with LCTs. Dulloo et al[25] found a 5% increase in 24-hour energy expenditure when humans were fed a diet containing 15 to 30 g MCTs. The discrepancies among these findings may be partly ascribed to differences in the composition of the test meals. Because these studies tested

mixed MCT or LCT meals, the protein or carbohydrate contained in the test meals may have affected the thermic effects of MCTs or LCTs.

On the other hand, Seaton et al[5] compared the thermic effect of meals consisting almost entirely of 48 g MCTs or 45 g corn oil. The MCT meal produced a significant increase in postprandial oxygen consumption compared with the LCT meal, resulting in an increase of energy expenditure over the basal level of 222 kJ/h and 71 kJ/h. These changes in energy expenditure were equivalent to 13% and 4% of the energy contained in the MCT and LCT meals, respectively. The results obtained in the present study are consistent with these findings, at least in part. SMLCTs containing 20% MCFAs may have thermic effects similar to MCTs. Sandstrom et al[26] suggested that SMLCTs are more rapidly oxidized than LCTs in postoperative patients and are associated with no side effects. Our results in this study support these findings.

MCTs and LCTs have different metabolic fates, which may account for the difference in postprandial thermogenesis. MCTs are rapidly absorbed in the small intestine and transported to the liver as free fatty acids via the hepatic portal circulation.[27-29] MCFAs enter the mitochondria of liver cells independently of fatty acyl-coenzyme A (CoA)-carnitine transferase, which is necessary for the transport of LCFAs into mitochondria.[7,30] Acetyl-CoA formed by β-oxidation can be oxidized further via the Krebs cycle to carbon dioxide and water or used in the synthesis of LCFAs and cholesterol. Two molecules of acetyl-CoA can condense to form ketones. The utilization of ketones by peripheral tissues is concentration-dependent, and oxidation can cause a significant increase in oxygen consumption if the oxidation of other substrates is not reduced appropriately.[31] The increase in serum 3-hydroxybutyrate found in this study is consistent with reports that MCT feeding produces hyperketonemia.[10,27,31] The increased thermic effect of SMLCTs would be related to the production and oxidation of ketone bodies. LCFA synthesis from acetyl-CoA in the liver requires large amounts of energy.[24,30] However, the concentration of triacylglycerol did not differ between SMLCTs and LCTs, suggesting that there is no increased export of triacylglycerol from the liver after SMLCT ingestion.

The higher increase in serum insulin following SMLCT ingestion compared with LCT ingestion agrees with MCT studies reported previously.[32] This increase in insulin might account for the suppression of lipolysis after SMLCT ingestion, as indicated by reduced glycerol concentrations.

The increase in thermogenesis following SMLCT ingestion suggests that the liver may play an important role in postprandial thermogenesis, as proposed previously.[4,30,32] Several hypothetical mechanisms may be propounded to explain the increased thermic effect of SMLCTs: a specific regulatory thermogenesis dependent on peroxisomal β-oxidation in brown adipose tissue[33]; a partial uncoupling of oxidative phosphorylation[34]; and a retroconversion of some adenosine triphosphate (ATP) molecules produced during the accelerated oxidation of MCFAs to adenosine diphosphate ([ADP] to restore a normal ATP/ADP ratio).[35]

SMLCTs are better for cooking than MCTs or a physical mixture of MCTs and LCTs because of their higher smoke point, which allows the use of larger amounts of cooking oil, eg, for deep-frying. It is not clear whether the effect of SMLCT remains when this oil is ingested with other macronutrients in regular meals. Further clinical studies are needed to clarify the impact of SMLCT ingestion on body fat during dietary therapy.

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