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# Effects of structured medium- and long-chain triacylglycerols in diets with various levels of fat on body fat accumulation in rats

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The effects of structured medium- and long-chain triacylglycerols (MLCT) in diets containing 50–200 g fat/kg on body fat accumulation were compared with those of long-chain triacylglycerols (LCT) in rats. In rats fed *ad libitum*, weights of intra-abdominal adipose tissues and carcass fat contents were significantly smaller ($P<0{\cdot}05$) in rats fed the 150–200 g MLCT/kg diet than in rats fed 150–200 g LCT/kg diet. Serum and liver triacylglycerol contents were significantly greater ($P<0{\cdot}05$) in rats fed 200 g MLCT/kg diet, as were hepatic capacities of citrate synthase and cytochrome oxidase ($P<0{\cdot}05$). The effects of MLCT on body fat were also examined in adult rats fed a limited amount of food (approximately 50 % of *ad libitum* intake). Reduction of body fat deposition during the food restriction was the same between in LCT and MLCT groups. These results suggest that accumulation of body fat was less efficient during long-term feeding of MLCT than LCT in rats fed high-fat diets *ad libitum*. The effect of MLCT on body fat might be influenced by the dietary fat content or by energy sufficiency.

**Body fat accumulation: Structured medium- and long-chain triacylglycerols: Dietary fat: Food restriction: Rats**

Obesity is characterized by an increase in lipid stores (Flatt, 1987; Fricker *et al.* 1989). It is generally associated with enhanced lipid consumption, which contributes to its development. In westernized countries, obesity is an important health problem affecting a large proportion of individuals, many of whom seek to prevent further weight gain or decide to counteract the detrimental health consequences of obesity (Atkinson, 1992). To attain these objectives, patients follow a wide variety of preventive or therapeutic methods, taken alone or in combination. Among these approaches, dietary restriction involving lipids is considered most important. The bulk of fatty acids found in most western diets consist of molecules comprising twelve or more C atoms. These long-chain fatty acids (LCFA), either saturated or unsaturated, originate from the long-chain triacylglycerols (LCT) provided by vegetable and/or animal oil and fat sources. They contribute to the supply of energy and fulfil essential fatty acid requirements (Bach *et al.* 1996).

In contrast, medium-chain triacylglyerols (MCT) are edible oils composed of triacylglycerols with saturated medium-chain fatty acids (MCFA) moieties of six to ten C atoms. These have been introduced into clinical nutrition programmes because of their rapid absorption and solubility (Seaton *et al.* 1986). MCT are metabolized differently from LCT. They are transported to the liver directly via hepatic portal circulation and are oxidized to ketones, whereas LCT are absorbed via the intestinal lymphatic ducts and transported in chylomicrons through the thoracic duct to reach the systemic circulation (Bach & Babyan, 1982; Senior 1990).

In animal studies, rats fed MCT do not gain as much weight as rats fed an isoenergetic amount of LCT (Bray *et al.* 1980; Bach & Babyan, 1982; Geliebter *et al.* 1983; Senior, 1990). Fat deposition is diminished, while RMR is increased (Bray *et al.* 1980; Baba *et al.* 1982; Geliebter *et al.* 1983; Senior, 1990). In clinical studies, Seaton *et al.* (1986) reported that mean postprandial $V_{O_2}$ after a meal containing MCT was higher than after a meal containing LCT. These findings suggest that MCT could be useful in the dietary treatment of obesity. However, it is difficult to substitute MCT for LCT in dietary fat for long-term dietary therapy, in part because utilization of MCT as a cooking oil is limited by the lower smoke point (Matsuo *et al.* 2001*a*).

Recently, we developed a new type of cooking oil composed of structured medium- and long-chain triacylglycerols (MLCT) (Matsuo *et al.* 2001*a*,*b*). The MLCT are structured lipids that contain MCFA (20 g/100 g total fatty acids) and LCFA (80 g/100 g total fatty acids) in the same triacylglycerol. They are made by transesterification of MCT and LCT, and are superior for cooking to physical

**Abbreviations:** LCFA, long-chain fatty acid; LCT, long-chain triacylglycerol; MCFA, medium-chain fatty acid; MCT, medium-chain triglycerol; MLCT, medium- and long-chain triacylglycerol.

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mixtures of MCT and LCT, because the smoke point of the former is higher than that of the latter. If MLCT had similar nutritional and physiological characteristics to MCT, MLCT could be used in a special cooking-oil for dietary therapy. We previously reported that energy expenditure was higher after MLCT ingestion (43·1 g) than after LCT ingestion (42·6 g) in healthy young women (Matsuo *et al.* 2001*b*), and that body fat accumulation was lower in rats fed 250 g MLCT/kg diet than in rats fed 250 g LCT/kg diet for 6 weeks (Takeuchi *et al.* 2001). However, the amount of MLCT in diets that would be optimum for dietary therapy is not clear.

In the present paper, we report the effects of several levels of MLCT in diets on body fat accumulation in growing rats. Moreover, we demonstrate the effects of MLCT on the decrease of body fat in adult rats fed a limited amount of food, because of the likelihood that MLCT might be useful in low-energy diets for obese people.

## Methods

All procedures involving animals were approved by Experimental Animal Care Committee of the Kagawa University.

### *Expt 1: effect of medium- and long-chain triacylglycerol in diets with various levels of fat on body fat accumulation in rats*

*Animals and diets.* Forty-eight male Wistar rats (3 weeks old) were obtained from Japan SLC, Inc. (Shizuoka, Japan). The rats were fed CE-2 (CLEA Japan, Tokyo, Japan), a commercial rodent diet, and water *ad libitum* to the age of 4 weeks. The rats were randomly assigned to eight groups. Half of the groups were fed diets containing LCT, and the other half were fed diets containing MLCT. The difference in dietary treatment was the level of dietary fat, as follows: 50, 100, 150 and 200 g/kg. The composition of experimental diets is shown in Table 1. Soyabean oil was used as the source of LCT, and was purchased from Nacalai Tesque, Inc. (Kyoto, Japan). MLCT were prepared by transesterification of 200 g MCT and 800 g rapeseed oil (Takeuchi *et al.* 2001), which were purchased from Nisshin Oil Mills (Tokyo, Japan). Composition of fatty acids and triacylglycerols of the test oils have been described previously (Matsuo *et al.* 2001*b*): LCT contained (g/100 g total fatty acids): linoleic acid 53, oleic acid 24, α-linolenic acid 8; MLCT contained (g/100 g total fatty acids): oleic acid 50, linoleic acid 16, caprylic acid 14, capric acid 5.

**Table 1.** Composition of experimental diets (g/kg)

| | Fat in experimental diets (g/kg) | | | |
|---|---|---|---|---|
| Ingredients | 50 | 100 | 150 | 200 |
| Casein | 255·0 | 255·0 | 255·0 | 255·0 |
| DL-Met | 4·0 | 4·0 | 4·0 | 4·0 |
| Maize starch | 460·0 | 410·0 | 360·0 | 310·0 |
| Sucrose | 120·0 | 120·0 | 120·0 | 120·0 |
| Fat* | 50·0 | 100·0 | 150·0 | 200·0 |
| Cellulose | 50·0 | 50·0 | 50·0 | 50·0 |
| Vitamin mixture† | 13·0 | 13·0 | 13·0 | 13·0 |
| Mineral mixture† | 45·0 | 45·0 | 45·0 | 45·0 |
| Choline chloride | 2·5 | 2·5 | 2·5 | 2·5 |
| Butylated hydroxytoluene | 0·50 | 0·50 | 0·50 | 0·50 |
| Total | 1000·0 | 1000·0 | 1000·0 | 1000·0 |

* Soyabean oil and structured medium- and long-chain triacylglycerol were used as experimental fats.
† Based on AIN-76A mineral and vitamin mixtures.

*Experimental design.* The animals were individually housed in an animal room at 24 ± 1°C with lights on from 08.00 to 20.00 hours. Each group of rats was given free access to experimental diet and water for 8 weeks. Body weights and food intakes were monitored daily. On the final day, rats in each dietary group were fasted for 12 h from 22.00 hours, then killed by decapitation. Blood was collected to obtain serum; liver and intra-abdominal adipose tissues (epididymal, perirenal and mesenteric) were quickly removed, weighed, and stored at −40°C until analyses. Carcass samples were obtained by removing the head, tail and splanchnic tissues, and were stored at −20°C until analysis of carcass composition.

*Analysis.* Concentrations of serum glucose and triacylglycerols were determined by methods reported previously (Fletcher, 1968; Bergmeyer & Bent, 1974). Serum insulin concentration was determined by a rat insulin enzyme immunoassay system purchased from Amersham Biosciences Inc. (Tokyo, Japan). Liver total lipid was extracted by the method of Folch *et al.* (1957) and liver triacylglycerol was measured by the method of Fletcher (1968). Carcass fat and protein were analysed using the method reported by Mickelsen & Anderson, (1959). For assays of hepatic capacities of citrate synthase and cytochrome oxidase, the frozen tissues were homogenized at 0–4°C in 0·1 M-phosphate buffer (pH 7·5) containing Triton X-100 (10 g/l) with three 20 s bursts of a motor-driven homogenizer (Ultra-Turrax T8; IKA Labortechnik, Staufen, Germany). Tissue capacity of the cytochrome oxidase was measured by the method of Smith (1955) using the homogenate. The rest of the homogenate was centrifuged at 0–4°C for 15 min at 600 ***g***. The supernatant fraction was used for measuring hepatic capacity of the citrate synthase by the method described by Srere (1969). The hepatic capacity of the enzymes were measured at 25°C and expressed as units per g wet weight. One unit of the enzyme catalysed the formation of 1 μmol free CoA/min for citrate synthase, and 1 μmol oxidized cytochrome *c*/min for cytochrome oxidase.

### *Expt 2: effect of medium- and long-chain triacylglycerol in diets with various levels of fat in rats fed a limited amount of food for weight reduction*

*Animals and experimental design.* Fifty-four male Wistar rats (10 weeks old) were obtained from Japan SLC, Inc. The rats were fed CE-2 diet (CLEA Japan) and water *ad libitum*, to the age of 15 weeks. After the pre-feeding period, six rats were killed as a control before initiation of food restriction, and each factor described later was analysed. The rest of the rats were randomly divided into eight groups. Half of the groups were fed diets containing LCT, and the other half were fed diets containing MLCT.

The difference in dietary treatment was the level of dietary fat as described in Expt 1. Diet composition and housing conditions were the same as in Expt 1. Each group of rats was fed 6 g diet/d (approximately 50 % of the meal observed in Expt 1) for weight reduction. The food restriction regimen was carried out for 21 d. On the final day of the experiment, rats in each group were killed by decapitation at 10.00 hours. Blood, organs, and carcass samples were collected and stored as described in Expt 1.

*Analysis*. Serum components, liver triacylglycerol, carcass compositions and enzyme capacities were assayed as in Expt 1.

### *Data analysis*

All data were analysed by a factorial ANOVA and *post hoc* Scheffé's test. Differences were considered statistically significant at $P<0{\cdot}05$.

## Results

### *Expt 1*

*Body weight, food intake and food efficiency*. In the LCT groups, final body weight and weight gain were significantly lower ($P<0{\cdot}05$) in rats fed 50 and 100 g fat/kg diet than in rats fed 150 and 200 g fat/kg diet (Table 2). In the 150 and 200 g fat/kg diets groups, final body weight and weight gain were significantly lower ($P<0{\cdot}05$) in the MLCT groups than in the LCT groups (Table 2). Food intake was more suppressed by the feeding of a high-fat diet, and food efficiency increased significantly ($P<0{\cdot}05$) with increasing dietary fat both in the LCT and MLCT groups (Table 2). In the 150 and 200 g fat/kg diet groups, food efficiency was significantly lower ($P<0{\cdot}05$) in the MLCT groups (Table 2).

*Tissue weights and liver triacylglycerol*. Liver weight and triacylglycerol content were not influenced by the levels of LCT and MLCT (Table 3). In the 200 g fat/kg diet group, liver triacylglycerol content was significantly higher ($P<0{\cdot}05$) in rats fed MLCT diets than in rats fed LCT diets (Table 3). Weights of intra-abdominal adipose tissues increased with increasing dietary fat in the LCT group (Table 3). In the 150 and 200 g fat/kg diet groups, perirenal adipose tissue weight and total intra-abdominal adipose tissue weight were significantly lower ($P<0{\cdot}05$) in rats fed MLCT diets than in rats fed LCT diets (Table 3).

*Carcass fat and protein*. Carcass weight and carcass fat content increased with increasing dietary fat in the LCT group (Table 4). In the 150 and 200 fat/kg diet groups, carcass weight and carcass fat content were significantly lower ($P<0{\cdot}05$) in rats fed MLCT diets than in rats fed LCT diets (Table 4). Carcass protein content was not affected by dietary fat level or dietary fatty acid composition (Table 4).

*Relationship between fat intake and body fat deposition*. Intra-abdominal adipose tissues and carcass fat content were positively correlated with LCT intake (epididymal $r$ 0·677, $P<0{\cdot}001$; perirenal $r$ 0·679, $P<0{\cdot}001$; mesenteric $r$ 0·582, $P<0{\cdot}01$; carcass fat $r$ 0·487, $P<0{\cdot}05$), but were not correlated with MLCT intake (epididymal $r$ 0·204, $P=0{\cdot}34$; perirenal $r$ 0·18, $P=0{\cdot}40$; mesenteric $r$ 0·326, $P=0{\cdot}12$; carcass fat $r$ 0·044, $P=0{\cdot}84$). Liver triacylglycerol content was not correlated with LCT or MLCT intake.

*Serum glucose, insulin and triacylglycerol concentrations*. Serum glucose and insulin concentrations were significantly lower ($P<0{\cdot}05$) in the 50 g fat/kg diet group than in 100, 150, and 200 g fat/kg diet groups in rats fed LCT and MLCT diets (Table 5). In the 200 g dietary fat/kg diet group, serum triacylglycerol concentration was significantly higher ($P<0{\cdot}05$) in rats fed MLCT diet than in rats fed LCT (Table 5).

**Table 2.** Body weight, food intake and food efficiency in rats fed experimental diets (Exp 1)†
(Mean values with their standard errors for six rate per group)

| | | Fat in experimental diets (g/kg) | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | 50 | | 100 | | 150 | | 200 | |
| | | Mean | SEM | Mean | SEM | Mean | SEM | Mean | SEM |
| Body weight (g) | | | | | | | | | |
| Initial | L | 87 | 2 | 88 | 1 | 88 | 1 | 89 | 1 |
| | ML | 88 | 1 | 88 | 1 | 88 | 1 | 88 | 1 |
| Final | L | 273$^{b}$ | 9 | 275$^{b}$ | 5 | 293$^{a}$ | 5 | 297$^{a}$ | 6 |
| | ML | 272 | 7 | 272 | 8 | 272* | 6 | 278* | 6 |
| Gain | L | 186$^{b}$ | 8 | 187$^{b}$ | 5 | 205$^{a}$ | 5 | 208$^{a}$ | 5 |
| | ML | 185 | 7 | 185 | 7 | 184* | 5 | 190* | 6 |
| Food intake (g) | | | | | | | | | |
| | L | 14·3$^{a}$ | 0·4 | 13·7$^{ab}$ | 0·2 | 13·6$^{ab}$ | 0·2 | 13·1$^{b}$ | 0·2 |
| | ML | 14·7$^{a}$ | 0·3 | 13·8$^{ab}$ | 0·4 | 12·9$^{b}$ | 0·4 | 12·5$^{b}$ | 0·4 |
| Food efficiency (mg/g) | | | | | | | | | |
| | L | 236$^{d}$ | 4 | 248$^{c}$ | 3 | 274$^{b}$ | 4 | 289$^{a}$ | 4 |
| | ML | 229$^{d}$ | 4 | 245$^{c}$ | 4 | 259$^{bc}$ | 3 | 276$^{ab}$ | 8 |

L, soyabean-oil diet; ML, structured medium- and long-chain triacylglrylcerol diet.
$^{a,b,c,d}$Mean values within a row with unlike superscript letters were significantly different ($P<0{\cdot}05$).
Mean values were significantly different from those of the L group: *$P<0{\cdot}05$.
† For details of diets and procedures, see Table 1 and p. 220.

**Table 3.** Liver weight, liver triacylglycerol weight and intra-abdominal adipose tissue weights in rats fed experimental diets (Exp 1)†

(Mean values with their standard errors for six rate per group)

| | Fat in experimental diets (g/kg) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 50 | | 100 | | 150 | | 200 | |
| | Mean | SEM | Mean | SEM | Mean | SEM | Mean | SEM |
| Liver weight (g) | | | | | | | | |
| L | 11·2 | 0·5 | 11·1 | 0·4 | 11·7 | 0·2 | 11·8 | 0·4 |
| ML | 11·3 | 0·4 | 11·8 | 0·4 | 11·8 | 0·3 | 11·7 | 0·6 |
| Liver triacylglycerol (mg/g) | | | | | | | | |
| L | 20·8 | 3·0 | 21·5 | 2·2 | 24·9 | 1·4 | 24·6 | 1·6 |
| ML | 25·4 | 2·6 | 27·3 | 1·2 | 28·6 | 1·7 | 30·8* | 3·1 |
| Epididymal adipose tissue (g) | | | | | | | | |
| L | 7·0[b] | 0·2 | 8·2[a] | 0·2 | 8·6[a] | 0·4 | 8·8[a] | 0·4 |
| ML | 8·1 | 0·3 | 8·0 | 0·4 | 7·7 | 0·2 | 7·5 | 0·5 |
| Perirenal adipose tissue (g) | | | | | | | | |
| L | 7·6[b] | 0·2 | 8·0[b] | 0·2 | 9·1[a] | 0·2 | 8·9[a] | 0·3 |
| ML | 7·9 | 0·5 | 8·3 | 0·3 | 7·8* | 0·2 | 7·5* | 0·4 |
| Mesenteric adipose tissue (g) | | | | | | | | |
| L | 5·8[b] | 0·3 | 6·0[a] | 0·2 | 6·8[a] | 0·3 | 6·9[a] | 0·4 |
| ML | 6·6 | 0·4 | 6·1 | 0·3 | 5·8 | 0·4 | 5·7* | 0·3 |
| Total intra-abdominal adipose tissue (g) | | | | | | | | |
| L | 20·4[c] | 0·7 | 22·2[b] | 0·2 | 24·5[a] | 0·7 | 24·6[a] | 0·9 |
| ML | 22·6 | 1·2 | 22·4 | 0·9 | 21·3* | 0·9 | 20·7* | 1·0 |

L, soyabean-oil diet; ML, structured medium- and long-chain triacylglycerol diet.
[a,b,c]Mean values within a row with unlike superscript letters were significantly different ($P<0·05$).
Mean values were significantly different from those of the L group: *$P<0·05$.
† For details of diets and procedures, see Table 1 and p. 220

*Hepatic capacity of the enzymes.* Hepatic capacities of the citrate synthase and cytochrome oxidase were not influenced by level of LCT, but were increased by increased MLCT in diets (Fig. 1). In the 150 and 200 g fat/kg diet groups, hepatic capacities of the citrate synthase and cytochrome oxidase were significantly higher ($P<0·05$) in rats fed MLCT than in rats fed LCT (Fig. 1).

*Expt 2*

*Body weight.* Rat body-weights reached approximately 280 (range 282–286) g before initiation of the food-restriction regimen. Food restriction for 21 d resulted in a loss of body weight to 223–240 g in all the groups, and the resulting body weights of rats fed both LCT and MLCT were the same. Total loss of body weight was less as both LCT and

**Table 4.** Carcass composition in rats fed experimental diets (Exp 1)†

(Mean values with their standard errors for six rats per group)

| | Fat in experimental diets (g/kg) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 50 | | 100 | | 150 | | 200 | |
| | Mean | SEM | Mean | SEM | Mean | SEM | Mean | SEM |
| Carcass weight (g) | | | | | | | | |
| L | 164[b] | 4 | 160[b] | 2 | 180[a] | 4 | 181[a] | 3 |
| ML | 161 | 4 | 163 | 3 | 160* | 5 | 165* | 1 |
| Carcass fat (g) | | | | | | | | |
| L | 36·7[b] | 1·9 | 37·3[b] | 1·9 | 43·6[a] | 2·4 | 43·2[a] | 2·9 |
| ML | 36·8 | 1·7 | 37·1 | 1·1 | 35·7* | 1·8 | 36·5* | 1·5 |
| Carcass fat (%) | | | | | | | | |
| L | 22·4 | 1·3 | 23·3 | 1·3 | 24·2 | 0·9 | 23·8 | 1·5 |
| ML | 22·9 | 0·9 | 22·8 | 0·5 | 22·3 | 1·0 | 22·9 | 1·0 |
| Carcass protein (g) | | | | | | | | |
| L | 38·0 | 2·1 | 35·8 | 1·5 | 40·3 | 1·2 | 39·0 | 1·5 |
| ML | 39·3 | 1·6 | 37·8 | 1·2 | 40·0 | 1·5 | 38·5 | 1·4 |
| Carcass protein (%) | | | | | | | | |
| L | 23·1 | 0·9 | 22·4 | 1·1 | 22·4 | 0·4 | 21·6 | 0·8 |
| ML | 24·4 | 0·5 | 23·2 | 0·4 | 24·9* | 0·3 | 24·1* | 0·9 |

L, soyabean-oil diet; ML, structured medium- and long-chain triacylglycerol diet.
[a,b]Mean values within a row with unlike superscript letters were significantly different ($P<0·05$).
Mean values were significantly different from those of the L group: *$P<0·05$.
† For details of diets and procedures, see Table 1 and p. 220.

**Table 5.** Serum glucose, insulin and triacylglycerol concentrations in rats fed experimental diets (Exp 1)† (Mean values with their standard errors for six rats per group)

| | Fat in experimental diets (g/kg) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 50 | | 100 | | 150 | | 200 | |
| | Mean | SEM | Mean | SEM | Mean | SEM | Mean | SEM |
| Glucose (mg/ml) | | | | | | | | |
| L | 1·69[b] | 0·04 | 1·80[a] | 0·05 | 1·86[a] | 0·05 | 1·85[a] | 0·05 |
| ML | 1·73[b] | 0·08 | 1·94[a] | 0·07 | 1·90[a] | 0·03 | 1·97[a] | 0·10 |
| Insulin (ng/ml) | | | | | | | | |
| L | 73·1[b] | 0·6 | 77·5[a] | 1·0 | 78·4[a] | 1·5 | 77·7[a] | 0·9 |
| ML | 74·4[b] | 0·7 | 77·8[a] | 1·1 | 79·2[a] | 1·2 | 78·1[a] | 1·9 |
| Triacylglycerol (mg/ml) | | | | | | | | |
| L | 1·87 | 0·17 | 2·04 | 0·36 | 1·93 | 0·15 | 1·55 | 0·22 |
| ML | 2·16 | 0·30 | 2·40 | 0·30 | 2·17 | 0·25 | 2·52* | 0·40 |

L, soyabean-oil diet; ML, structured medium- and long-chain triacylglycerol diet.
[a,b]Mean values within a row with unlike superscript letters were significantly different ($P<0·05$).
Mean values were significantly different from those of the L group: *$P<0·05$.
† For details of diets and procedures, see Table 1 and p. 220.

MLCT were increased in diets (mean values 57, 55, 48 and 46 g for 50, 100, 150 and 200 g fat/kg diet groups respectively).

*Tissue weights and liver triacylglycerol*. The weights of liver, epididymal, perirenal and mesenteric adipose tissues reached 10·8, 6·2, 5·4 and 4·4 g respectively before initiation of the food-restriction regimen. All of the weights of liver and adipose tissues were decreased by the food restriction (weight loss (%): liver 43–48, epididymal fat 35–60, perirenal fat 55–80, mesenteric fat 52–82). Liver triacylglycerol and weights of intra-abdominal adipose tissues had increased with increase in both LCT and MLCT in diets, but the differences were not observed between in the LCT and MLCT groups.

*Carcass fat and protein*. Carcass fat and protein content were 25·3 and 48·0 g respectively before initiation of the food-restriction regimen. On food restriction, the decrease in carcass fat was less as the dietary fat content increased, for both LCT and MLCT diets (mean values 17·0, 15·2, 13·1 and 12·6 g for 50, 100, 150 and 200 g fat/kg groups, respectively). There was no difference between LCT and MLCT groups. Carcass protein content and decrease in carcass protein were not influenced by dietary fat content or fatty acid composition (range of mean values (g): carcass protein 38·6–41·5, decrease in carcass protein 6·5–9·4).

*Serum glucose, insulin and triacylglycerol concentrations*. Serum triacylglycerol concentration was dramatically decreased by food restriction (mean values (mg/ml): control group 2·25, restrictive-diet groups 0·53–0·74). The concentrations of serum glucose, insulin and triacylglycerol were not affected by dietary fat content or fatty acid composition (range of mean values (mg/ml): serum glucose 1·51–1·73, serum insulin 69·6–77·1).

*Hepatic capacity of the enzymes*. Hepatic activities of citrate synthase and cytochrome oxidase were not influenced by dietary fat content or fatty acid composition (range of mean values (U/g): citrate synthase 29·2–32·8, cytochrome oxidase 46·7–51·2.

## Discussion

Many studies have reported that body fat accumulation is lower in animals fed MCT *v*. LCT diets (Bray *et al.* 1980; Bach & Babyan, 1982; Geliebter *et al.* 1983; Senior, 1990). However, the effect of dietary MLCT on body fat accumulation is unclear. Moreover, the effects of dietary MLCT level or food restriction on rat body fat deposition have not been examined. The present study was undertaken to investigate the effect of several level of MLCT in diets on body fat accumulation under growth and food restriction conditions. We have shown here that body fat accumulation was less in rats fed MLCT diets than in rats fed high-fat (150–200 g fat/kg) LCT diets (Expt 1). Moreover, intra-abdominal adipose tissues and carcass fat content were positively correlated with LCT intake, but not with MLCT intake (Expt 1). On the other hand, reduction of body fat deposition during the food restriction was the same between in LCT and MLCT groups (Expt 2). These results suggest that the MLCT diet would effectively inhibit body fat deposition in rats with high-fat feeding under energy sufficiency. These findings are consistent with the results of our previous report, in which body fat accumulation was greater in rats fed high-LCT diets (250 g/kg) *v*. rats fed high-MLCT diets for 6 weeks (Takeuchi *et al.* 2001). The present study also supports our clinical reports concerning MLCT (Matsuo *et al.* 2001*a*) and MCT (Tsuji *et al.* 2001).

MCT and LCT have different metabolic fates, which may account for the difference in postprandial thermogenesis. MCT are rapidly absorbed in the small intestine and transported to the liver as NEFA via hepatic portal circulation (Hashim & Tantibhedyangkul, 1987; Linscheer *et al.* 1970; Odle *et al.* 1991). MCFA enter the mitochondria of liver cells independently of fatty acyl-CoA-carnitine transferase, which is necessary for the transport of LCFA into mitochondria (Senior, 1990; Papamandjaris *et al.* 1998). Acetyl-CoA formed by β-oxidation can be oxidized further via the Krebs cycle to $CO_2$ and water. In the present study, hepatic capacities of the citrate synthase and cytochrome oxidase, key enzymes in the mitochondrial Krebs cycle

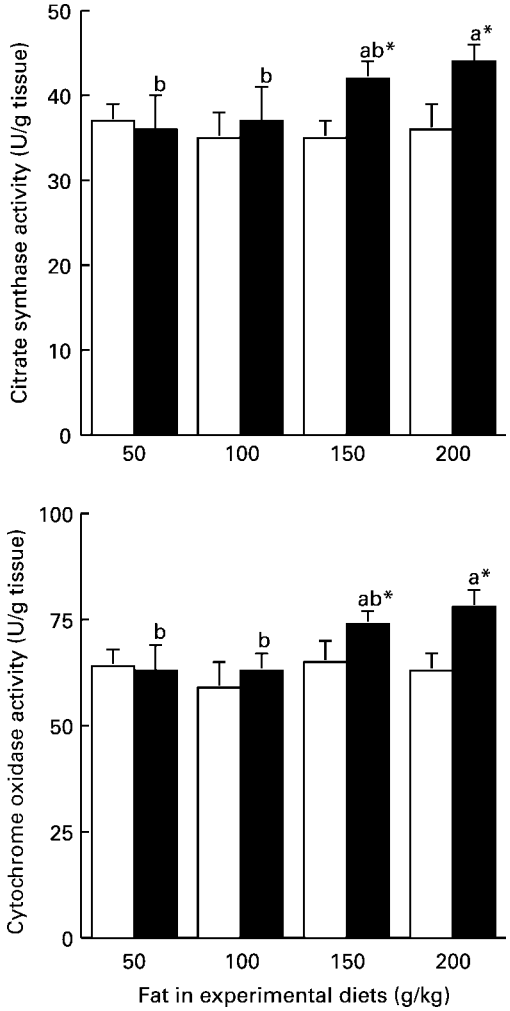


**Fig. 1.** Hepatic activities of the oxidative enzymes in rats fed experimental diets containing various levels of long-chain triacylglycerols (LCT, □) or medium- and long-chain triaclyglycerols (MLCT, ■) (Expt 1). One unit (U) of the enzyme catalysed the formation of 1 μmol free CoA/min for citrate synthase or 1 μmol oxidized cytochrome *c*/min for cytochrome oxidase. Values are means with their standards errors shown by vertical bars (six rats per group). For details of diets and procedures, see Table 1 and p. 220. [a,b]Mean values within the MLCT groups with unlike superscript letters were significantly different ($P<0{\cdot}05$). Mean values were significantly different from the those of the LCT group: *$P<0{\cdot}05$.

and electron transport chain, are used as markers for mitochondrial oxidative capacity. The capacities of these enzymes were higher in rats fed 150–200 g MLCT/kg diets under *ad libitum* conditions. These findings suggest that the higher capacity for mitochondrial oxidation might be related to the mechanism of lower body fat accumulation induced by the greater thermic effects of MLCT (Seydoux *et al.* 1986; Surette *et al.* 1992). The increase in thermogenesis following MLCT ingestion suggests that the liver may play an important role in postprandial thermogenesis, as proposed previously (Ruderman & Goodman, 1973; Berry *et al.* 1986; Bach *et al.* 1996). Several hypothetical mechanisms may be propounded to explain the increased thermic effect of MLCT: a specific regulatory thermogenesis dependent on peroxizomal β-oxidation in brown adipose tissue (Rothwell & Stock, 1987); a partial uncoupling of oxidative phosphroylation (Baba *et al.* 1987); a retroconversion of some ATP molecules produced during the accelerated oxidation of MCFA to ADP to restore a normal ATP:ADP ratio (Johnson & Cotter, 1986). Another possibility is that heat is produced in the liver induced by uncoupling protein-2 (Ricquier & Bouilla, 1998; Viguerie-Bascand *et al.* 1999).

In the present study, serum triacylglycerol levels and liver triacylglycerol contents were significantly greater in rats fed 200 g MLCT/kg diet than in rats fed 200 g LCT/kg diet under the *ad libitum* feeding conditions (Expt 1). Excess acetyl-CoA produced by β-oxidation of MCFA, and not oxidized in liver mitochondria, was used in the synthesis of LCFA (Senior, 1990; Papamandjaris *et al.* 1998). Hill *et al.* (1989, 1990) reported that 6 d of overfeeding with MCT significantly increased blood triacylglycerol concentration compared with LCT in non-obese male subjects. They suggested that changes in blood lipids with MCT feeding are consist with the hypothesis that excess dietary MCT causes a significant increase in the hepatic synthesis and/or chain elongation and desaturation. These processes could account for the higher rate of postprandial thermogenesis with MCT as compared with LCT (Hill *et al.* 1989, 1990). These results are supported by our present findings.

2-Monoacylglycerols released in the intestinal lumen are reconverted to triacylglycerols via the monoacylglycerol pathway, which could make the MCFA included in 2-monoacylglycerols less available for absorption via the portal vein (Ikeda *et al.* 1991; Christensen *et al.* 1995). However, medium-chain 2-monoacylglycerols are absorbed much less through lymph than long-chain 2-monoacylglycerols (Ikeda *et al.* 1991). Since a large proportion of 2-monoacylglycerols is expected to be hydrolysed to MCFA and glycerols by mucosal lipase (Hashim & Tantibhedyangkul, 1987), production of chylomicron from medium-chain 2-monoacylglycerols may be limited.

MLCT are better for cooking than MCT because of their higher smoke point, which allows the use of larger amounts in cooking oil. In the present study, we demonstrated that body fat accumulation was greater in rats fed LCT diets than in rats fed MLCT diets under the high-fat conditions. However, reduction of body fat content during food restriction was the same in LCT and MLCT groups. It is not entirely clear that the effects of MCLT are retained when this oil is used in cooking regular meals or for long-term diets. Further clinical studies are needed to clarify the impact of MLCT ingestion on human body fat during free-living and dietary therapy.

In summary, effects of MLCT in 50–200 g fat/kg diets on body fat accumulation were compared with those of LCT in rats. In rats fed *ad libitum*, weights of intra-abdominal adipose tissues and carcass fat content were significantly greater in rats fed the 150–200 g MLCT/kg diet. Reduction of body fat content during the food restriction (approximately 50 %) was the same in LCT and MLCT groups. These results suggest that accumulation of body fat was less efficient on long-term feeding of MLCT than on LCT in rats fed high-fat diets *ad libitum*. The effect of MLCT on body fat might be influenced by % dietary fat or energy sufficiency.

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