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Short communication

Effects of streptozotocin-induced diabetes on acetoacetyl-CoA synthetase activity in rats

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Abstract

In order to investigate the physiological role of acetoacetyl-CoA synthetase (acetoacetate-CoA ligase, EC 6.2.1.16), a cytosolic acetoacetate-activating enzyme, effects of streptozotocin (STZ)-induced diabetes on the enzyme activity was investigated in rats. At 72 hr of the STZ administration (80 mg/kg body weight, injected intravenously), hepatic enzyme specific activity decreased to 23% of its initial activity. However, the enzyme activities in non-hepatic tissues were not significantly affected by the STZ treatment. Feeding of rats with both 4% cholestyramine and 0.4% pravastatin for 3 days remarkably increased the hepatic acetoacetyl-CoA synthetase activity and decreased the plasma ketone bodies level in the diabetic rats. These results suggest that acetoacetyl-CoA synthetase has important roles in the regulation of ketone body utilization in rat liver and that these hypocholesterolemic agents have the ability to remedy the impaired utilization of ketone bodies under the diabetic condition.

Keywords: Acetoacetyl-CoA synthetase; Ketone body; Streptozotocin; Diabetes; Pravastatin; Cholestyramine

1. Introduction

Acetoacetyl-CoA synthetase (acetoacetate-CoA ligase, EC 6.2.1.16) is a cytosolic ketone body-metabolizing enzyme purified from a bacterium, *Zoogloea ramigera* I 16-M [1], and rat liver cytosol [2]. In mammalian tissues, acetoacetate is activated to its CoA ester by two distinct enzymes [3]. In the mitochondria of various tissues, it is converted through the action of 3-oxo-acid CoA-transferase (CoA-transferase) and then utilized as an energy source [4]. On the other hand, in the cytosol of lipogenic tissues, such as liver and brain, it is activated by the action of acetoacetyl-CoA synthetase, and effectively incorporated into cholesterol and fatty acids [5].

As to the regulation of acetoacetyl-CoA synthetase activity, we previously reported that this enzyme is negatively regulated in rats by fatty acyl-CoAs [6] similarly as acetyl-CoA carboxylase, a rate-limiting enzyme for fatty acid synthetase [7]. Furthermore, we showed that not only the activity but also the immunologically determined level of the enzyme in the liver remarkably increased and then decreased during the development [8] and increased upon the administration of hypocholesterolemic compounds, cholestyramine and/or pravastatin, to rats [9]. These results suggest that acetoacetyl-CoA synthetase, which may supply acetyl units directly from acetoacetate in the cytosolic compartment for cholesterol and/or fatty acids biosynthesis, is controlled in a similar manner as in the case of 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase or acetyl-CoA carboxylase. Rous suggested that synthesis of mouse acetoacetyl-CoA synthetase is controlled by insulin [10] as acetyl-CoA carboxylase. However, the physiological role of acetoacetyl-CoA synthetase has not been clarified yet compared to the other acetoacetate-activating enzyme, CoA-transferase. In the present study, we have examined the effects of STZ-induced diabetes, which causes abnormal metabolism of ketone bodies and is known to result in the decreases of HMG-CoA reductase [11] and acetyl-CoA carboxylase [12] activities, on acetoacetyl-CoA synthetase activity in rat liver. Besides, we have also investigated the influence of hypocholesterolemic agent administration on the effects of STZ-induced diabetes, since such agent is known to elevate the synthesis of HMG-CoA reductase [13].

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Abbreviations: HMG-CoA, 3-hydroxy-3-methylglutaryl coenzyme A; CoA-transferase, 3-oxo-acid CoA-transferase; STZ, streptozotocin.

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2. Materials and methods

2.1. Materials

Pravastatin and cholestyramine were kindly donated by Sankyo Co (Japan) and Bristol-Myers Squibb K.K. (Japan), respectively. The following were obtained from the supplier indicated: DL-[3-^{14}C] HMG-CoA (1.48–2.22 GBq/mmol) and [^{14}C] $NaHCO_3$ (1.48–2.22 GBq/mmol) from DuPont/NEN Research Products (USA); and STZ from Sigma (FRG). Other chemicals of reagent grade were purchased from commercial sources.

2.2. Animals

Female rats of the Sprague–Dawley strain (5 weeks old, obtained from Tokyo Laboratory Animals Science Co, Tokyo, Japan) were fed *ad libitum* on a standard diet (Oriental Yeast Co Ltd., Japan) for 1 week, and then used for the experiments. They were maintained in an air-conditioned room that was artificially illuminated from 8 a.m. to 8 p.m. Diabetes was induced by an intravenous injection of 80 or 60 mg STZ/kg body weight in 0.05 M citrate-Na (pH 4.5). All treatments of the rats were started at 10 a.m. and the sacrifice was also carried out at 10 a.m.

2.3. Preparation of liver subcellular fractions

All the following procedures were carried out at 0–4°. Rat livers were excised and homogenized in 4 vol. of 10 mM Tris–HCl (pH 7.5) containing 10 mM 2-mercaptoethanol, 250 mM sucrose and 1 mM ethylenediamine tetraacetic acid (EDTA) by five strokes in Teflon-glass homogenizer, and then centrifuged at 10,000 *g* for 20 min. The resultant supernatant was further centrifuged at 105,000 *g* for 60 min. The supernatant fraction was used for acetoacetyl-CoA synthetase activity, acetyl-CoA carboxylase activity and immunological assays. The sedimentated microsomal pellet was suspended in the above homogenizing buffer and used for the assay of HMG-CoA reductase activity.

For the assay of 3-oxo-acid CoA-transferase activity, another fraction of the homogenate was centrifuged at 30,000 *g* for 20 min. The particulate fraction thus obtained mainly comprising mitochondria was suspended in 10 mM Tris–HCl (pH 7.5) containing 10 mM 2-mercaptoethanol, 250 mM sucrose and 0.1% Triton X-100, and allowed to stand for 10 min. Then the suspension was centrifuged at 10,000 *g* for 15 min and the supernatant was used for the enzyme assay.

2.4. Enzyme assays

Acetoacetyl-CoA synthetase activity was determined by assaying acetyl-CoA produced from enzymatically formed acetoacetyl-CoA in the presence of CoA with the reaction catalyzed by *Z. ramigera* thiolase [1]. HMG-CoA reductase activity was determined radiochemically by the reduction of DL-[3-^{14}C] HMG-CoA to mevalonate as described by Kuroda and Endo [14]. Acetyl-CoA carboxylase was determined radiochemically by the formation of malonyl-CoA produced from acetyl-CoA and [^{14}C] $NaHCO_3$ as described by Fujioka *et al.* [15]. CoA-transferase was measured as acetoacetate formation by following the succinate-dependent breakdown of acetoacetyl-CoA–Mg^{2+} complex ($\varepsilon_{303} = 21{,}100$ L/mol/cm) as described by Hanson and Carrington [16].

2.5. Determination of mitochondrial HMG-CoA synthase mRNA content

Aliquots of total RNAs (10 μg) prepared from rat liver were resolved on a denaturing agarose gel (1%) containing formaldehyde and transferred to a Immobilon Transfer Membrane (Millipore Co, England) in 20 × SSC (1 × SSC = 0.15 M NaCl/0.015 M sodium citrate) overnight. The membrane was incubated in hybridization solution containing the ^{32}P-labeled rat HMG-CoA synthase probe as described by Hoshikawa *et al.* [17] and analyzed with a radio-imaging analyzer (BAS 2000, Fuji Photo Film Co, Japan). Labeling of HMG-CoA synthase probe was carried out using READY-TO-GO DNA labeling kit (Amersham Pharmacia Biotech., USA) with deoxycystidine [α-^{32}P] dCTP (110 TBq/mmol, Amersham Pharmacia Biotech., England). This probe is a 1554 bp amplification product of the rat liver cDNA between a sense primer (TGATACCTTAGAGATGGAGC) and an antisense primer (ATGGATTGCTCTCCTTAGAC) for rat mitochondrial HMG-CoA synthetase [18].

2.6. Determination of acetoacetyl-CoA synthetase protein concentration by enzyme immunoassay

A protein sample was subjected to SDS–PAGE (10% acrylamide gel) and the separated proteins were then transferred electrophoretically (Semi-Dry Transfer System; Bio Craft, Tokyo, Japan) to a polyvinylidene difluolide (PVDF) membrane. Acetoacetyl-CoA synthetase protein concentration on the membrane was determined by enzyme immunoassay using anti-rat acetoacetyl-CoA antiserum as described previously [8].

2.7. Serum measurements

Serum glucose concentration was determined with a glucose assay kit (Glucose CII-Test Wako, Wako Pure Chemical Industries, Japan), which was developed from the mutarotase–glucose oxidase method [19]. Serum cholesterol was measured with a cholesterol assay kit (Cholesterol E-test Wako, Wako Pure Chemical Industries, Japan), which was developed from the cholesterol esterase–cholesterol oxidase method [20]. Differential

determination of serum ketone bodies [21] was carried out using ketone body assay kit (Ketone Test Sanwa, Sanwa Kagaku Co, Japan).

3. Results and discussion

3.1. Effect of STZ-induced diabetes on acetoacetyl-CoA synthetase activity in rat liver

Fig. 1A shows that STZ-induced diabetes resulted in a marked decrease in acetoacetyl-CoA synthetase specific activity in the liver of rats as those of HMG-CoA reductase and acetyl-CoA carboxylase (Fig. 1B and C), which have been reported to be STZ-sensitive [11,12]. At 72 hr of the STZ administration (80 mg/kg body weight, injected intravenously), enzyme specific activities of acetoacetyl-CoA synthetase, HMG-CoA reductase and acetyl-CoA carboxylase were 23, 7.3 and 25%, respectively, of those of their individual initial activities. The time-course profile of the decrease in acetoacetyl-CoA synthetase activity was also similar to those of HMG-CoA reductase and acetyl-CoA carboxylase. Hyperglycaemia (Fig. 1D) and hyperketonemia (Fig. 1F) but not hypercholesterolemia (Fig. 1E) were observed during the first 72 hr after the STZ treatment under the experimental condition.

Table 1
Effect of STZ administration on acetoacetyl-CoA synthetase content in rat tissues

Tissues	Acetoacetyl-CoA synthetase protein content (μg/mg protein)	
	Control	Diabetes
Liver	0.507 ± 0.048	0.128 ± 0.017**
Brown adipose tissue	0.090 ± 0.021	0.037 ± 0.002*
White adipose tissue	1.029 ± 0.319	1.127 ± 0.230
Kidney	0.279 ± 0.034	0.273 ± 0.022
Lung	0.330 ± 0.063	0.364 ± 0.049
Brain	0.856 ± 0.236	1.071 ± 0.251

Diabetes was induced by an intravenous injection of 80 mg STZ/kg body weight. After 72 hr rats were sacrificed and supernatant fractions of their tissues were prepared. Acetoacetyl-CoA synthetase protein content in the fractions were measured as described in the text. Values are shown as the mean ± SE (N = 5). Statistical significance of the difference of means was evaluated by Student's *t*-test.
* $P < 0.05$; ** $P < 0.001$.

3.2. Acetoacetyl-CoA synthetase level in the tissues of STZ-induced diabetic rats

As shown in Table 1, immunologically determined acetoacetyl-CoA synthetase level in the control rats was high in liver, white adipose tissue and brain. In the STZ-induced diabetic rats, acetoacetyl-CoA synthetase protein

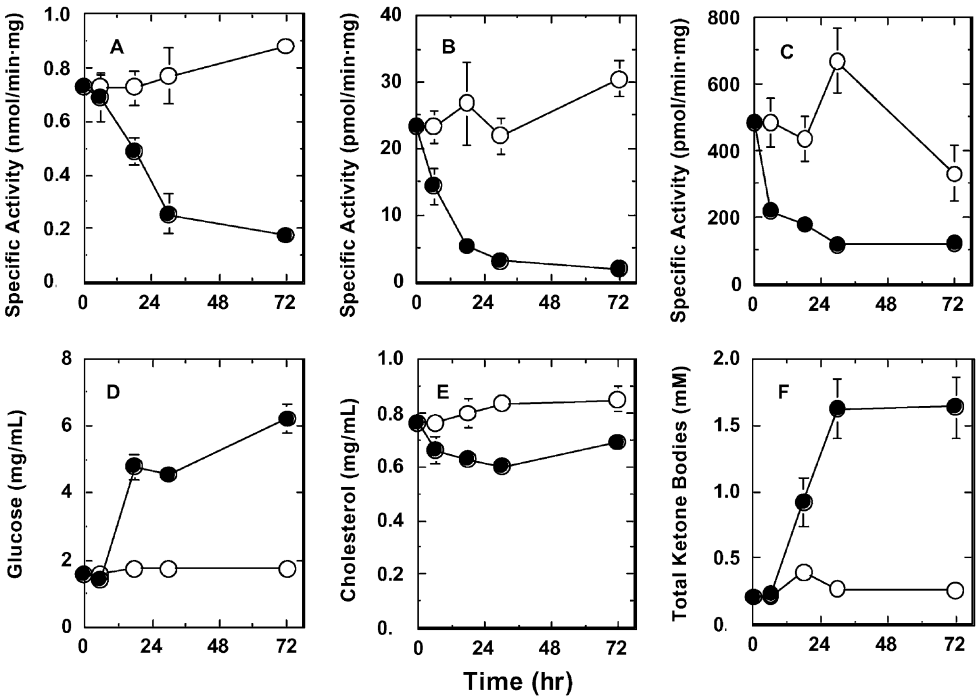

Fig. 1. Changes of acetoacetyl-CoA synthetase, HMG-CoA reductase, and acetyl-CoA carboxylase activities in the liver of STZ-treated rats. Groups of five female rats of Sprague–Dawley strain were induced diabetes by an intravenous injection of 80 mg STZ/kg body weight and sacrificed at the time indicated. Supernatant fractions of the livers were prepared. (A) Acetoacetyl-CoA synthetase, (B) HMG-CoA reductase, (C) acetyl-CoA carboxylase activities in the fractions, (D) glucose concentration, (E) cholesterol concentration, and (F) total ketone bodies (acetoacetate + 3-hydroxybutyrate) concentration in the blood serum of the rats were measured as described in the text. (○) Control rat; (●) STZ-treated rat. Each point represents the mean ± SE.

level in the liver decreased to about 25% of that in the control rats. The extent of the decrease is proportional to that in the enzyme activity (Fig. 1A), suggesting that the decrease in acetoacetyl-CoA synthetase activity induced by the STZ-treatment is not due to the alteration of catalytic efficiency of the enzyme. In contrast, such decrease was not observed in other main acetoacetyl-CoA synthetase-containing tissues (Table 1). In brown adipose tissue, which contained acetoacetyl-CoA synthetase protein at the level of less than one-tenth of that in white adipose tissue, the enzyme content considerably decreased by the STZ treatment.

3.3. Effect of hypocholesterolemic agents on ketone body metabolism

We previously showed that feeding of rats with two different hypocholesterolemic agents, cholestyramine, an anion exchanger and accelerator of bile acid excretion, and pravastatin, a competitive inhibitor of HMG-CoA reductase which is rate-limiting enzyme of cholesterol biosynthesis, remarkably elevated the liver acetoacetyl-CoA synthetase activity by increasing the enzyme amount [9]. As to the effect of pravastatin on plasma ketone body level, there is a report showing that it decreased the level in the patients with non-insulin dependent diabetes [22]. However, its effect on the ketone body metabolism in any mammalian tissues is entirely unknown. Therefore, we examined effects of hypocholesterolemic agents administration to the STZ-induced diabetic rats on their activities of the main enzymes responsible for ketone body metabolism. In order to observe pronounced effect of the agents on acetoacetyl-CoA synthetase activity, we used milder diabetic rats (60 mg STZ/kg body weight, injected intravenously) than those used in the experiments of Fig. 1 and Table 1.

As shown in Table 2, plasma glucose level in the diabetic rats fed with both 4% cholestyramine and 0.4% pravastatin for 3 days was somewhat lower than that in the rats fed with the normal diet although the level in the control rats was not affected by the feeding. Plasma total ketone bodies level in the control rats fed with the hypocholesterolemic agents was considerably higher than that in the rats fed with the normal diet. This difference was mainly due to the increase in β-hydroxybutyrate level. On the contrary, the increase in the plasma total ketone bodies level caused by STZ administration was almost eliminated to reach the level in the control rats fed with the agents. Hepatic acetoacetyl-CoA synthetase activity in the control or diabetic rats fed with the hypocholesterolemic agents was 8.0- or 8.9-fold higher, respectively, than that in the control or diabetic rats fed with the normal diet (Table 2). In contrast, activity of the other acetoacetate-activating enzyme, CoA-transferase, in various non-hepatic tissues was not significantly affected by the administration of hypocholesterolemic agents and/or STZ. Hepatic content of mRNA for mitochondrial HMG-CoA synthase, which is suggested to be a key enzyme in regulating ketogenesis, increased about 2-fold by the STZ treatment, as observed by Casals *et al*. [23]. However, the administration of hypocholesterolemic agents caused no significant change of the mRNA content in STZ-treated or control rats.

Endemann *et al*. [24] showed that in rat liver, which contains virtually no activity of CoA-transferase, ketone bodies contributed 25% of the carbon incorporated into sterols and 14% of that incorporated into fatty acids using liver perfusion [5]. They also reported that the incorporation

Table 2
Effect of hypocholesterolemic agents administration on the activities of ketone body-metabolizing enzymes in the tissues and the blood parameters of STZ-treated rats

Treatments	Acetoacetyl-CoA synthetase activity (nmol/min mg)	CoA-transferase activity (nmol/min mg)				HMG-CoA synthase mRNA (% of control)		
		Liver	Brown adipose tissue	White adipose tissue	Kidney	Lung	Brain	Liver
Control	0.569 ± 0.049	ND	79.6 ± 22.7	28.4 ± 0.3	84.9 ± 11.0	7.04 ± 1.02	26.0 ± 0.9	100 ± 8
STZ	0.276 ± 0.063**	ND	96.1 ± 13.9	29.8 ± 1.8	64.4 ± 3.2	5.87 ± 0.37	23.1 ± 1.2	192 ± 21**
PC	4.54 ± 0.33***	ND	71.6 ± 5.6	25.7 ± 1.1	70.8 ± 8.4	10.0 ± 2.0	21.3 ± 0.6**	107 ± 11
STZ + PC	2.45 ± 0.37**	ND	109 ± 15	19.5 ± 0.8***	74.2 ± 5.1	5.33 ± 0.96	23.5 ± 1.5	174 ± 5***
	Glucose (mg/mL)	Cholesterol (mg/mL)	Total ketone bodies (mM)	Acetoacetate (mM)	β-Hydroxybutyrate (mM)			
Control	1.30 ± 0.01	0.883 ± 0.044	221 ± 54	90.3 ± 21.7	131 ± 34			
STZ	3.41 ± 0.63*	0.964 ± 0.044	524 ± 114*	231 ± 68	294 ± 59*			
PC	1.30 ± 0.04	0.924 ± 0.058	285 ± 13	96.4 ± 14.2	189 ± 11			
STZ + PC	2.81 ± 0.43*	0.735 ± 0.036*	299 ± 37	117 ± 19	182 ± 28			

Rats were induced diabetes by an intravenous injection of 60 mg STZ/kg body weight, and then fed a standard diet or a standard diet supplemented with 0.4% pravastatin and 4% cholestyramine. After 72 hr rats were sacrificed and supernatant fraction of their liver, mitochondrial and total RNA fractions of their tissues were prepared. Acetoacetyl-CoA synthetase activity in the liver supernatant, CoA-transferase activities in the mitochondrial fractions, mitochondrial HMG-CoA synthase mRNA levels in the total RNA fractions, and blood parameters were measured as described in the text. (Control), standard diet-fed rats; (STZ), diabetic rats; (PC), hypocholesterolemic agents-fed rats; and (STZ + PC), hypocholesterolemic agents-fed diabetic rats. ND: not detected. Values are shown as the mean ± SE (N = 5). Statistical significance of the difference of means was evaluated by Student's *t*-test.

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

of ketone bodies into fatty acids and sterols was markedly inhibited in the perfused livers from STZ-induced diabetic rats despite the stimulation of ketogenesis by diabetes [25]. The marked decrease in acetoacetyl-CoA synthetase activity in the diabetic rats demonstrated here seems to be quite consistent with their results. Since the specific activity of acetoacetyl-CoA synthetase in the liver is much lower than those of CoA-transferase in non-hepatic tissues and HMG-CoA synthase in the liver, it is not clear yet to what extent the pronounced increase in acetoacetyl-CoA synthetase activity specifically induced by the hypocholesterolemic agents contributed to the decrease in plasma ketone bodies level in the diabetic rats. Furthermore, ketone body levels in the control rats appear to be even raised by the treatment of hypocholesterolemic agents despite the increase in acetoacetyl-CoA synthetase activity. Since HMG-CoA reductase inhibitor is known to increase the activity of mitochondrial carnitine palmitoyltransferase [26,27], it is possible that pravastatin caused the enhancement of mitochondrial β-oxidation. However, further investigation is required to elucidate the effects of hypocholesterolemic agents on the ketone body metabolism. In any case, the present results suggest that acetoacetyl-CoA synthetase has important roles in the regulation of ketone body utilization for the formation of biologically important lipidic substances in rat liver and that these hypocholesterolemic agents have the ability to remedy the impaired utilization of ketone bodies caused by diabetes-induced decrease in hepatic acetoacetyl-CoA synthetase activity.

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References

[1] Fukui T, Ito M, Tomita K. Purification and characterization of acetoacetyl-CoA synthetase from *Zoogloea ramigera* I-16-M. Eur J Biochem 1982;127:423–8.
[2] Ito M, Fukui T, Kamokari M, Saito T, Tomita K. Purification and characterization of acetoacetyl-CoA synthetase from rat liver. Biochim Biophys Acta 1984;794:183–93.
[3] Robinson AM, Williamson DH. Physiological roles of ketone bodies as substrates and signals in mammalian tissues. Physiol Rev 1980;60:143–87.
[4] Tildon JT, Sevdalian DA. CoA-transferase in the brain and other mammalian tissues. Arch Biochem Biophys 1972;148:382–90.
[5] Endemann G, Goetz PG, Edmond J, Brunengraber H. Lipogenesis from ketone bodies in the isolated perfused rat liver. Evidence for the cytosolic activation of acetoacetate. J Biol Chem 1982;257:3434–40.
[6] Ito M, Fukui T, Saito T, Tomita K. Inhibition of acetoacetyl-CoA synthetase from rat liver by fatty acyl-CoAs. Biochim Biophys Acta 1987;922:287–93.
[7] Bloch K, Vance D. Control mechanisms in the synthesis of saturated fatty acids. Ann Rev Biochem 1977;46:263–98.
[8] Nakamoto M, Takahashi N, Iwahori A, Sato H, Fukui T. Effects of development on acetoacetyl-CoA synthetase biosynthesis in rat liver. Biol Pharm Bull 1999;22:981–3.
[9] Iwahori A, Takahashi N, Nakamoto M, Iwama M, Fukui T. cDNA-derived amino acid sequence of acetoacetyl-CoA synthetase from rat liver. FEBS Lett 2000;466:239–43.
[10] Rous S. Fasting and insulin regulation of the utlization of acetoacetate for fatty acid synthesis. Arch Biochem Biophys 1977;179:328–33.
[11] Easom RA, Zammit VA. Effects of diabetes on the expressed and total activities of 3-hydroxy-3-methylglutaryl-CoA reductase in rat liver *in vivo*. Biochem J 1985;230:747–52.
[12] Pape ME, Lopez-Casillas F, Kim KH. Physiological regulation of acetyl-CoA carboxylase gene expression: effects of diet, diabetes, and lactation on acetyl-CoA carboxylase mRNA. Arch Biochem Biophys 1988;267:104–9.
[13] Liscum L, Luskey KL, Chin DJ, Ho YK, Goldstein JL, Brown MS. Regulation of 3-hydroxy-3-methylglutaryl coenzyme A reductase and its mRNA in rat liver as studied with a monoclonal antibody and a cDNA probe. J Biol Chem 1983;258:8450–5.
[14] Kuroda M, Endo A. Inhibition of *in vitro* cholesterol synthesis by fatty acids. Biochim Biophys Acta 1976;486:70–81.
[15] Fujioka T, Tsujita Y, Shimotsu H. Induction of fatty acid synthesis by pravastatin sodium in rat liver and primary hepatocytes. Eur J Pharmacol 1997;328:235–9.
[16] Hanson PJ, Carrington JM. Activity of 3-oxo-acid CoA-transferase, D-3-hydroxybutyrate dehydrogenase, hexokinase and carnitine palmitoyltransferase in the stomach and small and large intestine of the rat. Biochem J 1981;200:349–55.
[17] Hoshikawa M, Ohbayashi N, Yonamine A, Konishi M, Ozaki K, Fukui S, Itoh N. Structure and expression of a novel fibroblast growth factor, FGF-17, preferentially expressed in the embryonic brain. Biochem Biophys Res Commun 1998;244:187–91.
[18] Ayté J, Gil-Gomez G, Haro D, Marrero PF, Hegardt FG. Rat mitochondrial and cytosolic 3-hydroxy-3-methylglutaryl-CoA synthases are encoded by two different genes. Proc Natl Acad Sci USA 1990;87:3874–8.
[19] Miwa I, Okuda J, Maeda K, Okuda G. Mutarotase effect on colorimetric determination of blood glucose with D-glucose oxidase. Clin Chim Acta 1972;37:538–40.
[20] Allain CC, Poon LS, Chan CS, Richmond W, Fu PC. Enzymatic determination of total serum cholesterol. Clin Chem 1974;20:470–5.
[21] Harano Y, Kosugi K, Hyosu T, Uno S, Ichikawa Y, Shigeta Y. Sensitive and simplified method for the differential determination of serum levels of ketone bodies. Clin Chim Acta 1983;134:327–36.
[22] Sato T, Oouchi M, Nagakubo H, Chiba T, Ogawa S, Sato C, Sugimura K, Fukuda M. Effect of pravastatin on plasma ketone bodies in diabetics with hypercholesterolemia. Tohoku J Exp Med 1998;185:25–9.
[23] Casals N, Roca N, Guerrero M, Gil-Gomez G, Ayte J, Ciudad CJ, Hegardt FG. Regulation of the expression of the mitochondrial 3-hydroxy-3-methylglutaryl-CoA synthase gene. Its role in the control of ketogenesis. Biochem J 1992;283:261–4.
[24] McGarry JD, Foster DW. Regulation of hepatic fatty acid oxidation and ketone body production. Ann Rev Biochem 1980;49:395–420.
[25] Freed LE, Endemann G, Tomera JF, Gavino VC, Brunengraber H. Lipogenesis from ketone bodies in perfused livers from streptozocin-induced diabetic rats. Diabetes 1988;37:50–5.
[26] Cook GA, Khan B, Heimberg M. Feeding of lovastatin to rats increases the activity of the hepatic mitochondrial outer carnitine palmitoyltransferase. Biochem Biophys Res Commun 1988;150:1077–82.
[27] Bhuiyan J, Seccombe DW. The effects of 3-hydroxy-3-methylglutaryl-CoA reductase inhibition on tissue levels of carnitine and carnitine acyltransferase activity in the rabbit. Lipids 1996;31:867–70.