Improved Suppression by Dietary Taurine of the Fecal Excretion of Bile Acids from Hypothyroid Rats

Hideki MOCHIZUKI, Jun TAKIDO, and Hidehiko YOKOGOSHI

School of Food and Nutritional Sciences, The University of Shizuoka, 52-1 Yada, Shizuoka 422-8526, Japan

Received September 8, 1998; Accepted December 15, 1998

The effect of dietary taurine, 2-aminoethanesulfonic acid, on hypercholesterolemia caused by thiouracil-induced hypothyroidism was investigated in hypothyroid rats. Serum total- and HDL-cholesterol were significantly increased, and the excretion of fecal bile acids was significantly decreased. Taurine did not change the hypercholesterolemia, but significantly recovered the excretion of bile acids.

Key words: taurine; cholesterol; bile acids; hypothyroid rat

Taurine, 2-aminoethanesulfonic acid, is the major free intracellular amino acid present in many tissues and has various biological and physiological functions, including cell membrane stabilization, antioxidation, detoxification, osmoregulation, neuromodulation, and brain and retinal development. Taurine also plays an important role in lipid metabolism; that is, cholesterol is catabolized to bile acids in the liver which are excreted into the bile after conjugation with glycine (G) or taurine (T). The ratio of these two conjugates of bile acids (G/T) is known to vary depending on the animal species. Rats and mice have a low G/T ratio, and taurine is a major precursor for the conjugation of bile acids in these animals. It is considered that G/T ratio is most important to judge the cholesterol-lowering ability of taurine. Taurine enhanced the serum HDL-cholesterol concentration in rats, and a number of studies concerned with the hypercholesterolemic action of taurine have been conducted on cholesterol-loaded animals. The effect of taurine on the exogenous hypercholesterolemia induced by a high-cholesterol diet has been clearly examined. However, the effect of taurine on the endogenous hypercholesterolemia induced by some kinds of xenobiotics, excess histidine, diabetes, nephritis, and hypothyroidism has not been clearly investigated. The hypercholesterolemia induced by hypothyroidism is characterized by an increase of LDL- and HDL-cholesterol caused by an enhancement of the defective receptor-mediated catabolism of lipoproteins. On the other hand, taurine enhanced the LDL-receptor activity in Hep G2 cells. This evidence has suggested that taurine might possibly improve the hypercholesterolemia induced by hypothyroidism. In this study, we examined whether dietary taurine would have a hypocholesterolemic action on the endogenous hypercholesterolemia induced by hypothyroidism.

Young male rats of the Wistar strain weighing about 90 g (Japan SLC, Hamamatsu, Japan) were maintained at 24°C with a 12-hr light (7:00-19:00 hr) and dark cycle. To accustom the rats to the experimental conditions, they were initially given free access for 2 days to a 20% casein diet (basal diet), before being divided into groups. The composition of the basal diet was (in weight percent): casein, 20.0; mineral mixture (AIN-93G; Nihon Nosan, Yokohama, Japan), 5.0; corn oil, 5.0; vitamin mixture (AIN-76TM; Nihon Nosan), 1.0; choline chloride, 0.15; and a mixture of sucrose and corn starch (1:2, in weight) to 100%. The experimental diets incorporated thiouracil (0.3% in the diet) and/or taurine (0.5% or 3% in the diet) added to the basal diet at the expense of carbohydrate. The animals were individually housed and given free access to the experimental diets and water for 14 days. The rats were killed by decapitation at 2:00 p.m. after 4 hr of fasting on the last day of the experimental period, and blood was collected from the cervical wound. The serum lipids (total- and HDL-cholesterol) were enzymatically determined by using a commercial kit (Cholesterol C-test and HDL-cholesterol-test; Wako Pure Chemicals, Osaka, Japan). Serum thyroid hormones, 3,5,3'-triiodothyronine (T3) and thyroxine (T4), were measured by a radioimmunoassay, using antibody-coated beads (T-3-RIABEAD and T-4-RIABEAD; Dinabot Co. Ltd., Tokyo, Japan, respectively). Liver lipids were extracted by the method of Folch et al., and the concentrations of cholesterol and triglyceride (Triglyceride test; Wako Pure Chemicals) were determined. Fecal bile acids and cholesterol were measured by the enzymatic method and by a kit (Cholesterol C-test, respectively. The experimental procedures used in this study met the guidelines of the Animal Care and Use Committee of The University of Shizuoka. The statistical significance of differences between values was determined by an analysis of variance and Duncan’s multiple-range test.

The results are summarized in Table 1. Body weight gain and food intake were significantly reduced in the hypothyroid rats as compared with the values in normal rats, and taurine did not affect these values. Liver

---

Note

Improved Suppression by Dietary Taurine of the Fecal Excretion of Bile Acids from Hypothyroid Rats

Hideki MOCHIZUKI, Jun TAKIDO, and Hidehiko YOKOGOSHI

School of Food and Nutritional Sciences, The University of Shizuoka, 52-1 Yada, Shizuoka 422-8526, Japan

Received September 8, 1998; Accepted December 15, 1998

The effect of dietary taurine, 2-aminoethanesulfonic acid, on hypercholesterolemia caused by thiouracil-induced hypothyroidism was investigated in hypothyroid rats. Serum total- and HDL-cholesterol were significantly increased, and the excretion of fecal bile acids was significantly decreased. Taurine did not change the hypercholesterolemia, but significantly recovered the excretion of bile acids.

Key words: taurine; cholesterol; bile acids; hypothyroid rat

Taurine, 2-aminoethanesulfonic acid, is the major free intracellular amino acid present in many tissues and has various biological and physiological functions, including cell membrane stabilization, antioxidation, detoxification, osmoregulation, neuromodulation, and brain and retinal development. Taurine also plays an important role in lipid metabolism; that is, cholesterol is catabolized to bile acids in the liver which are excreted into the bile after conjugation with glycine (G) or taurine (T). The ratio of these two conjugates of bile acids (G/T) is known to vary depending on the animal species. Rats and mice have a low G/T ratio, and taurine is a major precursor for the conjugation of bile acids in these animals. It is considered that G/T ratio is most important to judge the cholesterol-lowering ability of taurine. Taurine enhanced the serum HDL-cholesterol concentration in rats, and a number of studies concerned with the hypercholesterolemic action of taurine have been conducted on cholesterol-loaded animals. The effect of taurine on the exogenous hypercholesterolemia induced by a high-cholesterol diet has been clearly examined. However, the effect of taurine on the endogenous hypercholesterolemia induced by some kinds of xenobiotics, excess histidine, diabetes, nephritis, and hypothyroidism has not been clearly investigated. The hypercholesterolemia induced by hypothyroidism is characterized by an increase of LDL- and HDL-cholesterol caused by an enhancement of the defective receptor-mediated catabolism of lipoproteins. On the other hand, taurine enhanced the LDL-receptor activity in Hep G2 cells. This evidence has suggested that taurine might possibly improve the hypercholesterolemia induced by hypothyroidism. In this study, we examined whether dietary taurine would have a hypocholesterolemic action on the endogenous hypercholesterolemia induced by hypothyroidism.

Young male rats of the Wistar strain weighing about 90 g (Japan SLC, Hamamatsu, Japan) were maintained at 24°C with a 12-hr light (7:00-19:00 hr) and dark cycle. To accustom the rats to the experimental conditions, they were initially given free access for 2 days to a 20% casein diet (basal diet), before being divided into groups. The composition of the basal diet was (in weight percent): casein, 20.0; mineral mixture (AIN-93G; Nihon Nosan, Yokohama, Japan), 5.0; corn oil, 5.0; vitamin mixture (AIN-76TM; Nihon Nosan), 1.0; choline chloride, 0.15; and a mixture of sucrose and corn starch (1:2, in weight) to 100%. The experimental diets incorporated thiouracil (0.3% in the diet) and/or taurine (0.5% or 3% in the diet) added to the basal diet at the expense of carbohydrate. The animals were individually housed and given free access to the experimental diets and water for 14 days. The rats were killed by decapitation at 2:00 p.m. after 4 hr of fasting on the last day of the experimental period, and blood was collected from the cervical wound. The serum lipids (total- and HDL-cholesterol) were enzymatically determined by using a commercial kit (Cholesterol C-test and HDL-cholesterol-test; Wako Pure Chemicals, Osaka, Japan). Serum thyroid hormones, 3,5,3'-triiodothyronine (T3) and thyroxine (T4), were measured by a radioimmunoassay, using antibody-coated beads (T-3-RIABEAD and T-4-RIABEAD; Dinabot Co. Ltd., Tokyo, Japan, respectively). Liver lipids were extracted by the method of Folch et al., and the concentrations of cholesterol and triglyceride (Triglyceride test; Wako Pure Chemicals) were determined. Fecal bile acids and cholesterol were measured by the enzymatic method and by a kit (Cholesterol C-test, respectively. The experimental procedures used in this study met the guidelines of the Animal Care and Use Committee of The University of Shizuoka. The statistical significance of differences between values was determined by an analysis of variance and Duncan’s multiple-range test.

The results are summarized in Table 1. Body weight gain and food intake were significantly reduced in the hypothyroid rats as compared with the values in normal rats, and taurine did not affect these values. Liver

---

To whom correspondence should be addressed. Tel: +81-54-264-5559; Fax: +81-54-263-7079; E-mail: yokogosi@fnsl.u-shizuoka-ken.ac.jp

Abbreviations: HDL, high-density lipoproteins; LDL, low-density lipoproteins; VLDL, very-low-density lipoproteins; CYP7A1, cholesterol 7α-hydroxylase; Tau, taurine

---
Table 1. Effect of Taurine on Lipid Metabolism in Thiouracil-induced Hypothyroid Rats

<table>
<thead>
<tr>
<th></th>
<th>Normal</th>
<th>Hypothyroidism</th>
<th>Hypothyroidism +0.5% Taurine</th>
<th>Hypothyroidism +3% Taurine</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body weight gain (g/14 days)</td>
<td>82.3 ± 2.0b</td>
<td>54.4 ± 2.6c</td>
<td>49.0 ± 2.6c</td>
<td>48.3 ± 2.2a</td>
</tr>
<tr>
<td>Final body weight (g)</td>
<td>178 ± 3.0b</td>
<td>149 ± 3.0a</td>
<td>145 ± 3.2a</td>
<td>142 ± 4.7b</td>
</tr>
<tr>
<td>Liver weight (% of body weight)</td>
<td>4.51 ± 0.16b</td>
<td>8.40 ± 0.13a</td>
<td>4.71 ± 0.08b</td>
<td>4.68 ± 0.05a</td>
</tr>
<tr>
<td>Food intake (g at day 10)</td>
<td>16.5 ± 1.3b</td>
<td>9.0 ± 0.7a</td>
<td>8.9 ± 0.8a</td>
<td>10.2 ± 0.6b</td>
</tr>
<tr>
<td>Serum thyroid hormones</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>T3 (x10⁻¹ μg/dl)</td>
<td>1.12 ± 0.08b</td>
<td>0.43 ± 0.03a</td>
<td>0.51 ± 0.03a</td>
<td>0.50 ± 0.06a</td>
</tr>
<tr>
<td>T4 (μg/dl)</td>
<td>7.16 ± 0.36b</td>
<td>2.04 ± 0.10a</td>
<td>1.98 ± 0.11a</td>
<td>1.90 ± 0.15a</td>
</tr>
<tr>
<td>Serum lipids (mg/dl)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total cholesterol</td>
<td>97.2 ± 4.7b</td>
<td>130 ± 6.2a</td>
<td>135 ± 12a</td>
<td>127 ± 11b</td>
</tr>
<tr>
<td>HDL-cholesterol</td>
<td>70.3 ± 3.7b</td>
<td>82.1 ± 5.3b</td>
<td>95.9 ± 5.3b</td>
<td>87.0 ± 4.7b</td>
</tr>
<tr>
<td>LDL + VLDL-cholesterol</td>
<td>26.9 ± 3.8b</td>
<td>47.9 ± 7.1b</td>
<td>30.9 ± 2.9b</td>
<td>40.3 ± 6.1b</td>
</tr>
<tr>
<td>Liver lipids (mg/g of liver)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cholesterol</td>
<td>5.37 ± 0.51b</td>
<td>3.87 ± 0.26b</td>
<td>3.48 ± 0.35b</td>
<td>3.54 ± 0.48b</td>
</tr>
<tr>
<td>Triglyceride</td>
<td>19.7 ± 3.7b</td>
<td>8.4 ± 1.4a</td>
<td>11.1 ± 1.1a</td>
<td>7.2 ± 5.5a</td>
</tr>
<tr>
<td>Dry fecal weight (g/2 days)</td>
<td>2.83 ± 0.18a</td>
<td>1.82 ± 0.17a</td>
<td>1.82 ± 0.09a</td>
<td>1.92 ± 0.05a</td>
</tr>
<tr>
<td>Fecal lipids (mg/2 days)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bile acids</td>
<td>8.25 ± 0.28c</td>
<td>5.10 ± 0.45a</td>
<td>6.34 ± 0.51b</td>
<td>7.94 ± 0.30c</td>
</tr>
<tr>
<td>Cholesterol</td>
<td>8.03 ± 0.51b</td>
<td>3.58 ± 0.54a</td>
<td>4.02 ± 0.54b</td>
<td>4.21 ± 0.03c</td>
</tr>
</tbody>
</table>

Rats were fed on the appropriate test diet for 14 days. Hypothyroidism was induced by thiouacil (0.3% in the diet).
Each value is the mean ± SEM of five rats per group; values with different superscripts (a–c) in a row are significantly different at p < 0.05.

weight did not differ among the four groups. Serum thyroid hormones (T3 and T4) were significantly decreased in the hypothyroid rats, and taurine did not affect the concentrations of the thyroid hormones. The concentrations of total- and HDL-cholesterol in the serum were significantly higher in the hypothyroid rats than in the normal rats. In the hypothyroid rats, taurine did not exhibit any hypocholesterolemic action toward this hypercholesterolemia. On the contrary, serum LDL + VLDL-cholesterol was significantly elevated by hypothyroidism, and taurine acted to slightly decrease this elevation. The hepatic concentrations of cholesterol and triglyceride were significantly decreased in the hypothyroid rats, and the supplementation of taurine did not affect the reduction of these lipids by hypothyroidism. The fecal excretion of bile acids and cholesterol was significantly depressed by the hypothyroidism, and the supplementation of taurine significantly increased the bile acid excretion.

In the hypothyroid rats, the hypercholesterolemia was caused by an enhancement of the intestinal absorption of cholesterol, suppression of the transformation of cholesterol into bile acids, and reduction in the number of LDL receptors. Considering the hypocholesterolemic action of some nutrients, the fecal excretion of neutral steroids is also an important factor, like bile acid excretion. For example, Nagata et al. have reported that the hypocholesterolemic action of soy protein compared with casein was partly caused by a reduction in the intestinal absorption of cholesterol and an increase in fecal steroid excretion. On the other hand, in the case of hypercholesterolemia caused by a high-cholesterol diet, taurine reduced the serum cholesterol levels, but did not affect fecal steroid excretion. These results correspond with the present results. The ability of bile acid synthesis is dependent on the activity of cholesterol 7α-hydroxylase (CYP7A1) which is the rate-limiting enzyme for cholesterol degradation. It is known that taurine enhanced the CYP7A1 activity and its gene expression (unpublished data), so it was expected that taurine might stimulate this enzyme activity and consequently increase the fecal excretion of bile acids from hypothyroid rats. Our results prove the enhanced fecal excretion of bile acids. On the other hand, it has been demonstrated that taurine enhanced the activity of LDL receptors in HepG2 cells and the hypercholesterolemia induced by hypothyroidism was also characterized by reduced LDL-catabolism. Although we consequently expected a hypocholesterolemic action from taurine in this hypercholesterolemia induced by hypothyroidism, our results show that taurine had no hypocholesterolemic effect. However, as taurine significantly improved the reduced excretion of fecal bile acid caused by hypothyroidism, taurine might be expected to increase the activity of CYP7A1. Yagasaki et al. have reported that, in hypothyroid rats, methionine improved the hypercholesterolemia, but promoted decreased body weight gain and food intake. It is suggested that the effect of taurine differs from the effect of methionine on hypothyroid animals, because taurine did not change the body weight and food intake. It remains unclear whether taurine can directly affect the activity of the LDL-receptor in vivo; therefore, we will determine the gene expression of the LDL-receptor and the activity of CYP7A1 in the liver of hypothyroid rats in the near future.

References
Taurine and Cholesterol in Hypothyroid Rats