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Effect of Chicory Root Extract on Cholesterol Metabolism in Rats

Jae-Young Cha, Soon-Jae Jeong and Young-Su Cho*

Faculty of Natural Resources and Life Science, Dong-A University, Busan 604-714, Korea

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Effects of water-soluble extract from roasted-chicory root on the cholesterol metabolism in rats fed cholesterol diet were investigated. Sprague-Dawley rats received a hypercholesterolemic diets without (control group) or with 5.0% water-soluble extract from roasted chicory root for 2 weeks. Roasted chicory extract group showed significantly higher body weight gain and food intake compared with the control group. The concentrations of total cholesterol and LDL+VLDL cholesterol in serum were significantly lower in rats fed roasted chicory extract diet. However, HDL-cholesterol concentration, and atherogenic index were not significantly affected by the dietary roasted chicory extract. Fecal net weight, fecal cholesterol, and bile acid excretion were significantly higher in the chicory extract group. The results suggest that the hypocholesterolemic effect in rats fed roasted chicory extract may be caused by an alteration in the absorption of cholesterol by an increase in the fecal excretion of cholesterol and bile acid.

Key words: *roasted-chicory extract, hypocholesterolemic effect, rat, bile acid.*

Elevated plasma cholesterol is regarded as a primary risk factor for the coronary heart disease.[1] Because plant food ingredients are generally known to be safe and have few side effects. The hypolipidemic effects have been studied.[2,3] Recently, the action mechanism of novel compounds derived from natural products on lipid metabolism has been intensively investigated in experimental animals and human subjects.[3,4] Chicory (*Chicorium intybus*) containing non-digestible oligosaccharides is recognized as a natural food ingredient in most European countries, Japan, and Korea, but little attention has been paid until recently on their potential physiological effects.[5,6] Because chicory extracts containing inulin or oligosaccharides are water-soluble, they are expected to behave like soluble fibers, which have a hypolipidemic effect.[7,8] In recent animal studies, hypotriglycerolemic effect through the inhibition of triglyceride synthesis by dietary chicory components has been demonstrated.[8,9] It was also reported that chicory extract in rats fed with cholesterol-enriched diet influences the concentration of serum lipids and fecal lipid excretion.[10] However, the possible mechanism for cholesterol decrease by chicory extract has not been well elucidated. We previously confirmed that water-soluble extracts from unroasted and roasted chicory roots exert different effects on serum lipid levels in rats fed a normal diet, which dose not contain cholesterol.[3] However, no study has been reported on the water-soluble extract derived from roasted chicory root on lipid metabolism in animals fed diet containing cholesterol.

*Corresponding author
Phone: 82-51-200-7586; Fax: 82-51-200-7505
E-mail: choys@mail.donga.ac.kr.

Abbreviations: LDL, low density lipoprotein; VLDL, very low density lipoprotein; HDL, high density lipoprotein.

This study was conducted to examine the effect of water-soluble extract from roasted chicory root on the cholesterol concentrations in serum and liver and fecal lipid excretions.

Materials and Methods

Materials. Chicory extract was provided by Korea Ginseng and Tobacco Research Institute (Taejon, Korea). All other chemicals and reagents were of the best commercial grade available.

Diets and animal experiments. Male Sprague-Dawley rats aged four weeks were housed individually in suspended wire-mesh stainless cages in a temperature controlled animal room (21~24°C) with a 12 hours light/dark cycle (07 : 00~19 : 00). Rats were maintained on a basal semisynthetic diet (control group) without chicory extract for 1 week before the experimental start and were then divided into two groups of six each. The experimental diets were prepared by supplementing roasted chicory extract at a level of 5.0% (w/w) at the expense of sucrose to the hypercholesterolemic diet (Table 1). Food and water were provided *ad libitum* for 2 weeks. Food intake amount was recorded daily, and the body weight was recorded every other day during the experimental period. The feces of each rat were collected during the feeding period from day 12 to 13. At the end of the feeding period, the rats were killed by withdrawing blood from the abdominal aorta under light diethylether anesthesia after 12 hours of starvation. Tissues were excised from each rat, weighed, and stored at −80°C before analysis.

Lipid analysis. The serum was separated through the centrifugation of the blood at 3,000 rpm for 15 min. Serum total cholesterol (Cholesterol C-test) and high-density lipoprotein (HDL)-cholesterol (HDL-cholesterol E-test) were measured

Table 1. Composition of experimental diets. (Unit: %)

Ingredients	Control	Roasted-chicory
Casein	20.0	20.0
α-Corn starch	15.0	15.0
Palm oil	10.0	10.0
Cellulose	5.0	5.0
AIN-93 mineral mixture	4.0	4.0
AIN-93 vitamin mixture	1.0	1.0
L-Methionine	0.3	0.3
Choline bitartrate	0.2	0.2
Cholesterol	0.5	0.5
Sodium cholate	0.125	0.125
Roasted-chicory	-	5.0
Sucrose	to make 100	

-: not supplemented in diet.

Table 2. Effects of roasted chicory extract on body weight, food intake, and liver weight in rats.

Ingredient	Control	Roasted-chicory
Initial body weight (g)	151.7 ± 1.48	151.9 ± 1.03
Final body weight (g)	261.1 ± 2.36	281.5 ± 3.58**
Food intake (g/day)	21.78 ± 0.23	22.57 ± 0.11*
Liver total weight (g)	15.82 ± 0.70	15.95 ± 0.72
Relative liver weight (g/100 g B.W.)	5.80 ± 0.22	5.67 ± 0.27

Rats were fed with the hypercholesterolemic diet containing roasted chicory extract at a 5.0% (w/w) level for 2 weeks.
Values are means ± SE of six rats per group.
*p<0.05, **p<0.01. correspondence to control group.

Table 3. Effects of roasted chicory extract on the cholestrol concentrations of serum and liver in rats.

Ingredient	Control	Roasted-chicory
Liver cholesterol (mg/g)	41.66 ± 3.51	36.14 ± 1.70
Serum lipids (mg/100 ml)		
Total cholesterol	276.23 ± 21.8	191.80 ± 9.9*
HDL-cholesterol	27.29 ± 1.2	24.51 ± 1.0
LDL+VLDL cholesterol[1)]	206.93 ± 30.12	162.29 ± 6.11***
Atherogenic index[2)]	7.74 ± 1.39	6.62 ± 0.01

Rats were fed with the hypercholesterolemic diet containing roasted chicory extract at a 5.0% (w/w) level for 2 weeks.
Values are means ± SE of six rats per group.
[1)] LDL+VLDL cholesterol=Total cholestrol−HDL cholestrol.
[2)]Atherogenic index was expressed as LDL+VLDL cholestrol/HDL cholestrol.
*p<0.05, ***p<0.001 correspondence to control group.

Table 4. Effects of roasted chicory extract on dietary cholesterol intake, fecal cholesterol, and bile acid excretion in rats.

Ingredient	Control	Roasted-chicory
Fecal net weight (g/day)	3.69 ± 0.12	5.54 ± 0.14***
Cholesterol intake (mg/day)	115.8 ± 4.21	123.8 ± 1.01
Fecal cholesterol excretion (mg/day)	8.87 ± 0.67	12.59 ± 1.34*
Fecal bile acid excretion (mg/day)	34.04 ± 3.30	49.57 ± 1.16**

Rats were fed with the hypercholesterolemic diet containing roasted chicory extract at a 5.0% (w/w) level for 2 weeks.
Values are means ± SE of six rats per group.
*p<0.05, **p<0.01, ***p<0.001 correspondence to control group.

enzymically using commercial kits from Wako Pure Chemical Ind. (Osaka, Japan). The liver lipids were extracted with chloroform-methanol (2 : 1, v/v) using the method of Folch *et al.*[11)] The concentration of cholesterol in the lipid extracts was measured using the method of Sperry andWebb. [12)]

Dietary and fecal lipid analysis. Feces and food were extracted using the method of Tokunaga *et al.*,[13)] and the extracted solutions were used to determine the concentrations of fecal and dietary cholesterol and fecal bile acid. The fecal cholesterol and bile acid were measured with commercially available Cholesterol E-test and Bile acid-test kit (Woko Pure Chemical Inc., Osaka, Japan) after those dissolving in the isopropyl alcohol after evaporating the extracted solution.

Statistical analysis. Data from the animal experiments were statistically analyzed through Student's t-test, and significant differences in the means were inspected at $p<0.05$, $p<0.01$, and $p<0.001$.

Results and Discussion

Body weight gain, food intake, and liver weight. Initial body weights of rats randomly assigned to the two experimental groups were similar. However, final body weights of rats fed roasted chicory extract were significantly greater ($p<0.01$) than those fed the control diet (Table 2). Food intake was also significantly greater($p<0.05$) in the roasted chicory extract-fed group. This result also agreed with a previous observation that rats fed 5% chicory extract or 10% chicory oligofructose showed significantly greater food intake than those fed the control diet.[9,10)] However, no significant differences were observed in their liver total and relative weights (g/100 g body weight) during the feeding period (Table 2). It was also reported that liver weights of hamsters fed inulin-supplemented diets did not differ from those of the controls.[14)] Therefore, present study appears to suggest that dietary supplementation of roasted chicory extract does not exert any harmful effects under the present experimental condition.

Serum cholesterol concentrations. Concentrations of total cholesterol and LDL+VLDL-cholesterol in serum were significantly lower in roasted chicory extract-fed group than the control group (Table 3). Our results were in agreement with those of other studies, which reported a significant reduction in serum cholesterol concentration in inulin,[14,16)] soluble fibers,[15)] and oligofructose.[9)] However, no significant differences have been reported in serum cholesterol concentrations between rats fed 10% fructooligosaccharides (Neosugar) or 5% chicory and the controls.[10,13)] In our previous study, the concentration of serum total cholesterol was not significantly affected by 5% roasted or 5% unroasted chicory extract in normocholesterolemic rats.[3]

Differences in the cholesterolemic effect with similar dietary fibers among different studies may have stemmed from the level of supplemented cholesterol, the presence/absence of cholate, the level of dietary fiber or animal species.[3,7,10] The experimental diet included 0.5% cholesterol and 0.125% sodium cholate, which are experimentally used widely to induce a mild hypercholesterolemia in rats.[3,17,18] This study also added 5.0% roasted chicory extract in to the experimental diet. Other investigators have generally chosen 5~10% of dietary fibers for various nutritional studies.[10,16,19] The amount of 5% chicory extract used in this study would be equivalent to daily consumption of about 25 g dietary fiber in humans.[20] Dietary recommendations for nonstarch polysaccharides are 12~24 g/day as recommended by WHO.[19]

Liver cholesterol concentration. Concentrations of liver cholesterol were not significantly different between the two groups (Table 3). Other studies also observed that the liver cholesterol concentration was not significantly different between hypercholesterolemic rats or hamsters fed inulin or chicory diets and those fed the control diet.[10,15,21]

Fecal lipid concentrations. Fecal net weight (g/day) was significantly greater (p<0.01) in rats fed roasted chicory extract diet compared with those fed the control diet (Table 4). Fecal cholesterol and bile acid excretions were greater in rats fed roasted chicory extract diet (Table 4). This result was in agreement with those of other investigators[10,21] who found higher fecal cholesterol and bile acid excretion in rats fed 6.0% raw inulin or 6.0% baked inulin extracts, and 5% chicory extract. Several hypotheses concerning the mechanism by which dietary fibers elicit their hypocholesterolemic effect have been proposed. The most frequently suggested mechanism is the interference with intestinal cholesterol and bile acid absorption, leading to an increase in fecal neutral sterol and bile acid excretion. It was reported that dietary fibers might exert their hypocholesterolemic effect by increasing the excretion of fecal neutral sterols[22] The reduced cholesterol absorption may result in lower cholesterol concentration in serum. Anderson[15] and Story[23] also reported that most soluble fiber intakes increased fecal bile acid excretion, which probably contributed to the cholesterol-lowering effect.

The present study suggests that the hypocholesterolemic effect of hypercholesterolemia rats fed roasted chicory extract may have been caused by an alteration in the absorption of neutral sterols by an increase in the fecal excretion of neutral sterols.

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