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学術論文

Effects of Dietary Apios (*Apios americana* Medikus Tuber) on Plasma and Liver Lipid Levels in Rats[*1]
ラットの血漿および肝臓脂質レベルに対する食餌アピオスの作用

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The effects of dietary apios (*Apios americana* Medikus tuber) ingestion on plasma and liver lipid levels were studied in rats by comparing them to the effects of the potato, the potatoes most commonly eaten in Japan. Rats were fed the apios and potato in amounts based on the human ingestion of energy from potatoes and starches as a fraction of all food intake. Apios feeding enhanced the plasma high-density lipoprotein (HDL)-cholesterol concentration. On the other hand, potato ingestion did not affect the plasma HDL-cholesterol concentration. Plasma (very-low-density lipoprotein plus low-density lipoprotein)-cholesterol concentration was not changed by either the apios or potato diet. These results suggest that the apios may exert an antiatherosclerotic action along with an increase in the plasma HDL-cholesterol concentration.

Key words: apios (*Apios americana* Medikus tuber), liver lipid, plasma lipid
アピオス，肝臓脂質，血漿脂質

INTRODUCTION

Potatoes are an important food and energy source throughout the world. Most potatoes are utilized as food, as an ingredient of starch and as animal feed. The main ingredient of potatoes is carbohydrate consisting of starch. The potatoes which are most important as food throughout the world are the potato, *Solanum tuberosum*, sweet potato, *Ipomoea batatas*, cassava, *Manihot esculenta*, taro, *Colocasia esculenta*, and yam, *Dioscorea* spp. Other than the potato, these potatoes originally come from the western part of Africa, South-East Asia or the tropical region of the Americas, and have been cultivated in tropical, subtropical and temperate zone areas since ancient times. In Japan, the potatoes cultivated now are classified as potatoes, sweet potatoes, taroes and yams.

Apios (*Apios americana* Medikus tuber) is a potato, and an edible tuberous legume that originated in the eastern part of North America. The subterranean stem is partially massive, and has been eaten since ancient times. Apios was imported into Japan in the middle of the Meiji era, and is cultivated now in the northern part of the Tohoku region. Apios is rich in protein, lipid, iron, sodium and calcium, and the total energy per weight of apios is 2.6-fold that of the potato.

The present study examined the effects of dietary apios ingestion on plasma and liver lipid levels in rats by comparing them to the effects of potato ingestion. One group of rats was fed the apios in amounts based on the human ingestion of energy from potatoes and starches as a fraction of all food intake. The potatoes most commonly eaten in Japan are the potato, *S. tuberosum*, so another group of rats was fed this potato in amounts based on the human ingestion of energy from potatoes and starches as a fraction of all food intake.

MATERIALS AND METHODS

Animals and diets. This animal experiment was conducted with the approval of the Ethics Committee of Animal Study of Morioka Junior College, Iwate Prefectural University.

Male Wistar rats (3 wk old, Charles River Japan Inc., Kanagawa, Japan) were individually housed in stainless steel cages with wire bottoms in an air-conditioned room at a temperature of 22 ± 2 ℃, a relative humidity of 60 ± 5 %, and a 12-h light cycle (8:00-20:00). They were fed a stock pellet diet (MF; Oriental Yeast Co., Tokyo, Japan) followed by a basal diet for 4 d. Subsequently, the rats were divided into three groups with similar body weights and were fed the basal (Control group) or experimental diets containing the apios or potato. The compositions of the basal[1)] and experimental diets

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Abbreviations: HDL, high-density lipoprotein; LDL, low-density lipoprotein; NEFA, nonesterified fatty acid; VLDL, very-low-density lipoprotein.

Table 1. Composition of experimental diets (g / 100 g).

Ingredients	Control	Apios	Potato
Casein [1]	20	19.66	19.73
α-Cornstarch [1]	13.2	13.2	13.2
Cornstarch [1]	39.75	37.78	37.33
Sucrose [2]	10	10	10
Cellulose powder [1]	5	4.89	4.95
Soybean oil [1]	7	6.86	6.97
Mineral mixture (AIN93G composition) [1]	3.5	3.5	3.5
Vitamin mixture (AIN93 composition) [1]	1	1	1
Choline bitartrate [3]	0.25	0.25	0.25
L-Cystine [3]	0.3	0.3	0.3
Apios [4]	-	2.56	-
Potato	-	-	2.77

[1] Oriental Yeast Co., Tokyo, Japan.

[2] Nissin Sugar Manufacturing Co., Tokyo, Japan.

[3] Wako Pure Chemical Industries, Osaka, Japan.

[4] JA Tohoku Tenma, Aomori, Japan.

are shown in Table 1. In the present study, rats were fed these two kinds of potatoes in amounts based on the human ingestion of energy from potatoes and starches as a fraction of all food intake. Humans ingest about 2.5 % of their food energy from potatoes and starches in Japan. The energy of the basal diet is 416 kcal/100 g in the present study, so the caloric intake from potatoes and starches is equal to 10.4 kcal/100 g. Therefore, the experimental diets of the apios and potato groups were supplemented with 2.56 g of apios and 2.77 g of potato per 100 g diet, respectively, at the expense of cornstarch. The apios and potato were boiled, left at room temperature, then freeze-dried and powdered. The energy of the apios (tuber, raw) and potato (tuber, raw) is 198 and 77[2)] kcal/100 g edible portion, respectively. All of the diets contained the same levels of protein (20 %), lipid (7 %) and dietary fiber (5 %). The rats were kept for an additional 28 d. The diet and water were available at all times. Animals were deprived of their diet at 9:00 on the 28th day but allowed free access to water until killing, which was performed 4 h later. Blood was collected from the heart and left to clot at room temperature so that plasma could be obtained. The liver was quickly removed, washed with cold 0.9 % NaCl, blotted on filter paper, and weighed. The plasma and liver were stored at -80 ℃ until being analyzed. Aliquots of the liver were also preserved in methanol and stored at 4 ℃ until lipid content analyses were performed.

Lipid analyses. Plasma total cholesterol, HDL-cholesterol, triglyceride, phospholipid, and nonesterified fatty acid (NEFA) concentrations were determined by an enzymatic method using a Cholesterol E-test Wako, HDL-Cholesterol E-test Wako, Triglyceride E-test Wako, Phospholipid C-test Wako, and NEFA C-test Wako, respectively. All test kits were obtained from Wako Pure Chemical Industries, Osaka, Japan. The difference between total cholesterol concentration and HDL-cholesterol concentration was regarded as (VLDL+LDL)-cholesterol concentration. The ratio of (VLDL+LDL)-cholesterol concentration to HDL-cholesterol concentration was designated as the atherogenic index. The ratio of HDL-cholesterol concentration to total cholesterol concentration was estimated as the HDL-cholesterol ratio.

Total lipids from the liver were extracted according to the procedure described by Folch *et al.*[3)] After portions of the chloroform phase had been dried under nitrogen, cholesterol,[4)] triglyceride,[5)] and phospholipid[6)] contents were determined.

Statistical analyses. Results were expressed as means ± standard errors. Statistical analysis was carried out by one-way analysis of variance followed by Fisher's protected least significant difference (PLSD) test. A significance level of $p < 0.05$ was used for all the comparisons.

RESULTS

Table 2 shows the initial body weight, food intake and body weight gain over the 28 d of experimental feeding and the relative liver weight at the end of the experimental feeding. Food intake, body weight gain and liver weight were not significantly different among the three groups.

Table 2. Initial body weight, food intake, body weight gain and liver weight in rats and rats fed on apios or potato diet.

Measurement	Control	Apios	Potato
Initial body weight (g)	73.4 ± 1.3	74.2 ± 0.9	73.4 ± 1.5
Food intake (g/28d)	501.9 ± 13.8	536.0 ± 9.1	527.3 ± 8.3
Body weight gain (g/28d)	206.4 ± 7.6	212.7 ± 3.7	216.5 ±7.0
Liver weight (g/100g body weight)	4.30 ± 0.11	4.23 ± 0.09	4.27± 0.11

Each value represents the mean ± standard error for five rats.

Plasma cholesterol concentrations are shown in Fig. 1. Plasma total cholesterol concentration in the groups fed both apios and potato was not significantly different from that of the control group. In terms of lipoprotein cholesterol concentration, the apios ingestion significantly enhanced the plasma HDL-cholesterol concentration. Plasma (VLDL+LDL)-cholesterol concentration was not changed by the apios diet. On the other hand, potato ingestion did not affect either the plasma HDL- or (VLDL+LDL)-cholesterol concentration. Atherogenic index and HDL-cholesterol ratio were not significantly different among the three groups.

Fig. 2 shows the plasma triglyceride, phospholipid and NEFA concentrations. Neither the apios nor the potato diet affected the plasma triglyceride or NEFA concentration; however, these concentrations in the apios diet group tended to be lower than those in the potato diet group. There was no significant difference in the plasma phospholipid concentration among the three groups.

Liver lipid contents are shown in Fig. 3. There were no significant differences in the liver cholesterol and phospholipid contents among the three groups. Liver triglyceride content was also not affected by either apios or potato ingestion; however,

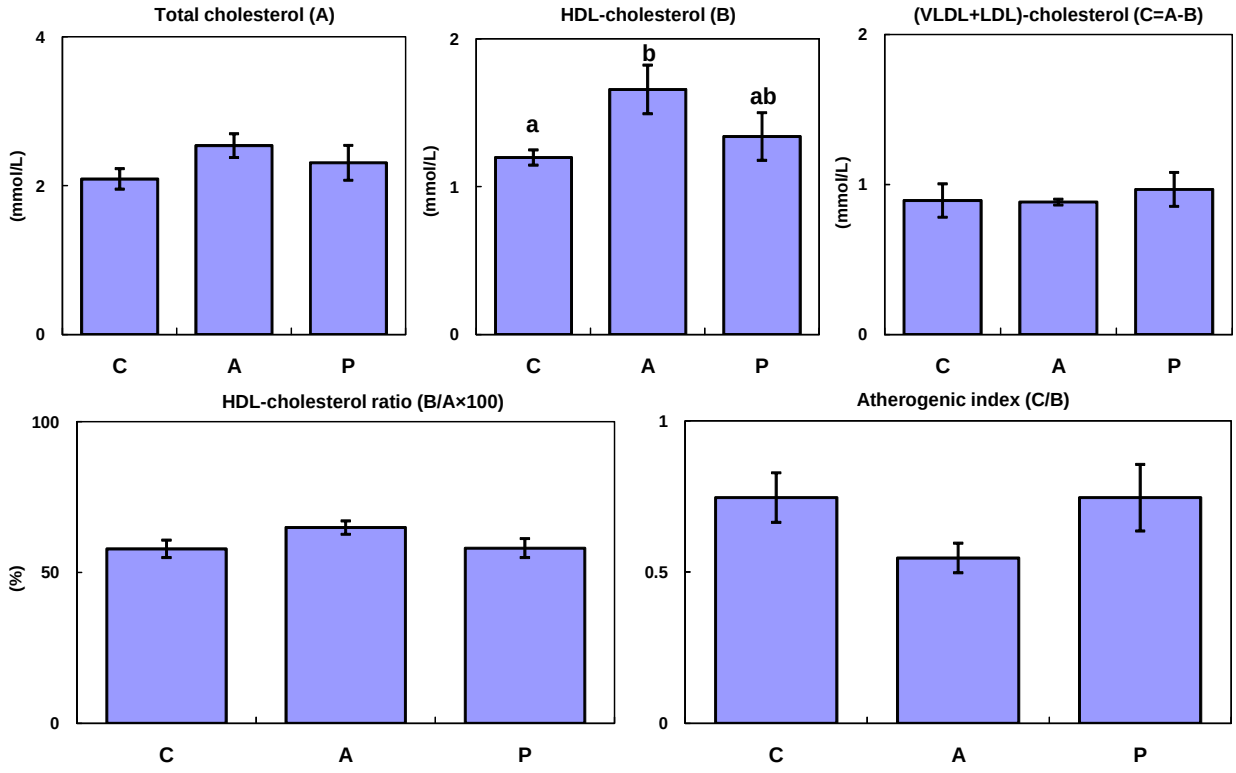

Fig. 1. Effects of dietary apios or potato on plasma cholesterol concentration, atherogenic index and high-density lipoprotein (HDL)-cholesterol ratio in rats. Measurement of the plasma cholesterol concentration was carried out as described in the MATERIALS AND METHODS section. Each value and vertical bar represents the mean and standard error for five rats. Values not sharing a common letter are significantly different at $p < 0.05$ by one-way analysis of variance followed by Fisher's protected least significant difference (PLSD) test. C, basal diet (Control) group; A, apios diet group; P, potato diet group.

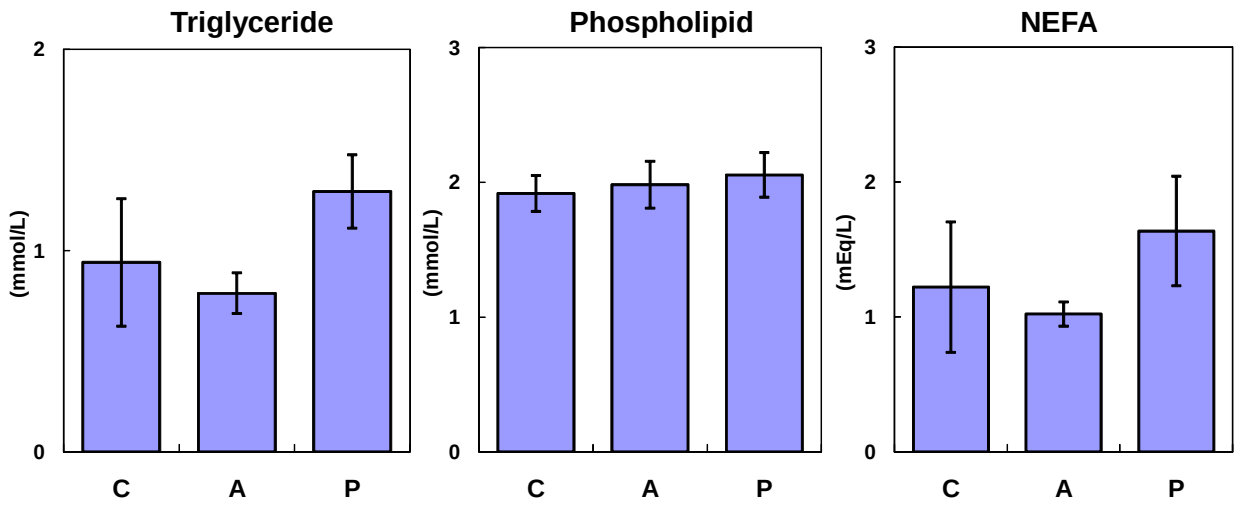

Fig. 2. Effects of dietary apios or potato on plasma triglyceride, phospholipid and nonesterified fatty acid (NEFA) concentrations in rats. Measurements of the plasma triglyceride, phospholipid and NEFA concentrations were carried out as described in the MATERIALS AND METHODS section. Each value and vertical bar represents the mean and standard error for five rats. C, basal diet (Control) group; A, apios diet group; P, potato diet group.

the liver triglyceride content in the apios diet group tended to be lower than that of the potato group.

DISCUSSION

This study was performed to evaluate the effects of dietary apios on plasma and liver lipid levels in rats. Apios feeding enhanced the plasma HDL-cholesterol concentration without increasing the total cholesterol concentration, and the HDL-cholesterol-lifting effect induced by dietary apios was stronger than that induced by potato. The mechanism of the plasma HDL-cholesterol-lifting action might be the specificity of the apios protein. Dietary protein and peptide are known to change the cholesterol concentration in the serum or plasma. For example, dietary proso millet protein concentrate increased the plasma HDL-cholesterol concentration in comparison to the casein diet in mice.[7,8] Dietary fish protein ingestion increased the serum HDL-cholesterol concentration as compared with the casein diet in rabbits.[9,10] Potato and soy peptide diets increased the serum HDL-cholesterol concentration as compared with the casein diet in rats.[11] There is a possibility that the apios protein

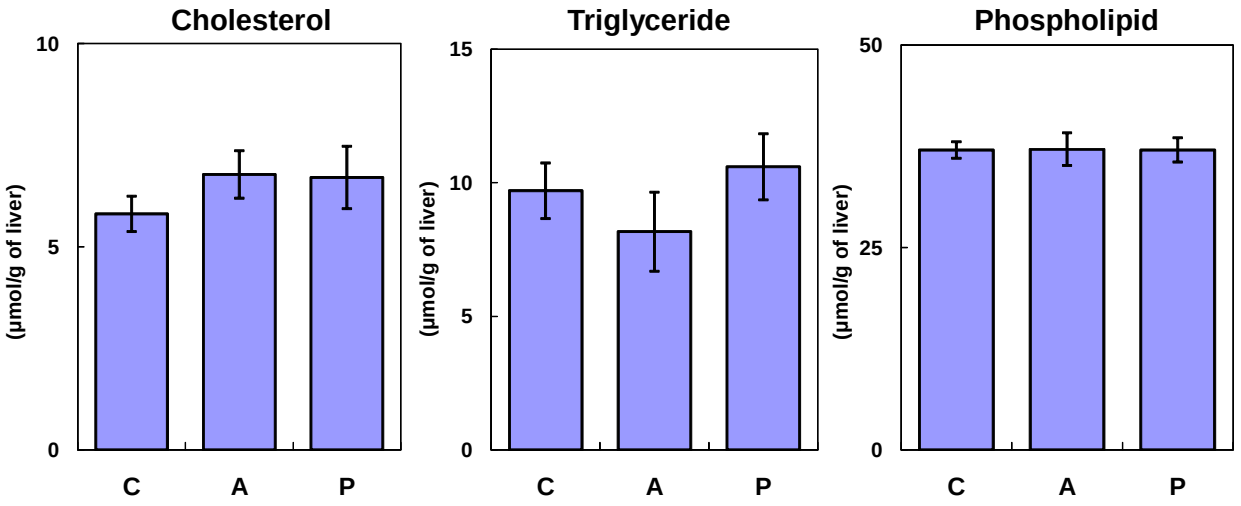

Fig. 3. Effects of dietary apios or potato on liver lipid contents in rats. Measurements of the liver lipid contents were carried out as described in the MATERIALS AND METHODS section. Each value and vertical bar represents the mean and standard error for five rats. C, basal diet (Control) group; A, apios diet group; P, potato diet group.

might also play a role in the plasma HDL-cholesterol-lifting action seen in the present study.

In the present study, neither the apios nor the potato diet changed the plasma (VLDL+LDL)-cholesterol concentration. It is reported that resistant starch of beans[12] and retrograded starch of potato pulps[13] lowered the serum total cholesterol and (VLDL+LDL)-cholesterol concentrations in rats. Retrograded starch is a kind of resistant starch that has resistance against amylase, and may act like dietary fiber in the digestive tract. The apios and potato samples used in the present study were prepared by the same method used in the above-mentioned study, although the apios and potato contents in the present study (about only 2.6-2.8 g per 100 g diet) were lower than those of the potato pulps in the above-mentioned study (15 g per 100 g diet). Thus, it is possible that no effect could be seen in the plasma total and (VLDL+LDL)-cholesterol concentrations due to apios and potato ingestion.

Cholesterol is an animal sterol and plays many important roles in the body. For example, cholesterol is a constituent of cell membrane and a precursor of steroid hormones, cholecalciferol (vitamin D_3) and bile acids. Cholesterol transport to extrahepatic tissues is primarily ensured by LDL, while HDL retrieves cholesterol from extrahepatic tissues and carries it back to the liver in the blood. The concentration of HDL-cholesterol is inversely related to the risk of coronary heart disease. The results seen in the present study suggest that dietary apios might have a beneficial effect on the cholesterol metabolism.

In conclusion, apios exerts an antiatherosclerotic action and results in an increase in the plasma HDL-cholesterol concentration. These findings suggest that the ingestion of apios could have a beneficial effect on the lipid metabolism.

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和文要旨　　ラットにおける血漿および肝臓脂質レベルに対する食餌アピオス（アメリカホドイモ根茎）摂取の作用をわが国で最も一般的に摂取されているイモ類であるジャガイモ摂取時と比較検討した。ラットにはアピオスならびにジャガイモをヒトのイモ及びデンプン類エネルギー摂取量に基づいて与えた。アピオスの摂取により血漿高密度リポタンパク質 (HDL) コレステロール濃度が有意に上昇した。その一方で，ジャガイモの摂取では有意な影響はみられなかった。血漿超低密度リポタンパク質および低密度リポタンパク質コレステロール濃度にはアピオスならびにジャガイモ摂取による変動はみられなかった。これらの結果より，アピオスは血漿 HDL コレステロール濃度を上昇させることにより抗動脈硬化的に作用する可能性が示唆された。