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Effects of Garlic Vinegar Supplementation on Body Weight, Blood Glucose, and Serum Lipid Profile in Streptozotocin-induced Diabetic Rats-fed High Cholesterol Diet

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Abstract

The effects of garlic vinegar supplementation on body weight, blood glucose, and plasma triglyceride and cholesterol were investigated in streptozotocin-induced diabetic rats fed high-cholesterol (1%) diet for 4 wk. The garlic vinegar was made by fermenting 20% fresh garlic juice. There was no effect of garlic vinegar on body weight, plasma glucose or triglyceride concentrations in the diabetic rats. Plasma total-cholesterol concentrations were unaffected by garlic vinegar supplementation. However, plasma LDL-cholesterol concentrations and atherogenic index were significantly lower in the diabetic rats supplemented with garlic vinegar diet than in the control rats. The diabetic-rats supplemented with garlic vinegar not only had increased HDL-cholesterol levels but had decreased LDL-cholesterol. This alteration in the HDL/LDL-cholesterol ratio in the diabetic rats may decrease the risk of atherosclerosis. Therefore, the over-all effect of garlic vinegar supplement may contribute to the antiatherogenic role in streptozotocin-induced diabetic rats.

Key words: garlic vinegar, plasma glucose, plasma lipids, streptozotocin-induced diabetic rats

INTRODUCTION

Allium sativum, or garlic, is reported to have hypoglycemic (1) and hypolipidemic properties (2) and to have an inhibitory effect on atherosclerosis (3). Atherosclerosis is the most common complication of diabetes. Diabetic men have a two-to threefold higher risk of coronary heart disease, stroke, and peripheral vascular disease, whereas diabetic women have a three to fivefold higher risk than matched nondiabetic individuals.

The most common lipid abnormality in diabetes is hypertriglyceridemia (4). Although mechanisms responsible for accelerated atherosclerosis are not yet understood clearly, abnormalities in low-density lipoprotein (LDL), a decreased serum high-density lipoproteins (HDL), and an increased serum triglycerides contribute to accelerated atherosclerosis in diabetes (5,6). Reducing risk of vascular disease requires improved glycemic control, avoidance of cigarette smoking, normal blood pressure, and desirable serum lipoprotein levels. In comparison of plasma cholesterol levels in diabetic patients and matched controls, cholesterol concentrations in general, are significantly elevated in diabetics compared to those of non-diabetic patients (6,7).

The typical diet in Korea is low in fat, high in carbohydrates, and moderate in protein content (8). Several studies with non-diabetic subjects and non-insulin-dependent diabetes (NIDDM) patients have reported accentuation of hypertriglyceridemia and decreased HDL-cholesterol concentrations with high carbohydrate intakes (9,10). Lipid-lowering effect of allicin extracted from garlic in rats was demonstrated by Augusti and Mathew (11), and the hypolipidemic effect of garlic through the reduction of the elevation of alimentary hyperlipedemia (12). Even the occasional consumption of garlic resulted in significantly lowered levels of serum cholesterol and triglyceride in epidemiological studies (13).

There is no report on the nutritional effects of garlic vinegar administration in diabetics at relatively low doses for extended periods. Therefore, the primary purpose of this study was to determine whether garlic vinegar supplementation induces a hypolipidemic response in rats fed a high-cholesterol diet in diabetic rats. The effects of garlic vinegar supplementation on blood glucose and body weight were also evaluated.

MATERIALS AND METHODS

Twenty-five male Sprague-Dawley rats were obtained from K.L.E.C (Korea Life Engineering Corporation, Seoul, Korea). The average body weight of the animals upon arrival was 141±11 g. Animals were housed individually in stainless steel cages (10″×15″×7″) in a room with controlled temperature and exposed to an alternating twelve hour period of light and dark. All animals were fed a Samyang rat chow diet and water ad libitum for three weeks after arrival. Body weights were recorded weekly. When the animals attained weights of 190 to 210 grams, the animals were made chemically diabetic by an intramuscular injection of streptozotocin (Sigma Chemical Co., St. Louis, MO) at a dose of 50 mg/kg body weight in 0.25 M citrate buffer, pH 4.5 (14).

At the same time, 0.25 M citrate buffer, pH 4.5, was injected intramuscularly into the non-diabetic rats. At three days

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after streptozotocin treatment, and the appearance of glucose in the urine Multistix (Ames Company Division Miles Laboratory. Inc., Elkhart, IN) was used to confirm the diabetic state. Blood samples were taken from the tail vein of unanesthetized rats using heparinized, microhematocrit capillary tubes, and the plasma glucose concentration was analyzed using GLUCOPAT (Kyoto Daiichi Kagaku Co., LTD Japan). Animals were diagnosed as being diabetic if they had a non-fasting blood glucose concentration greater than 300 mg/dl in whole blood. Then these diabetic rats were randomly assigned to two experimental groups (control or vinegar supplemented diet). The diet containing garlic was passed through a sieve to avoid forming lumps. The composition of the diets are presented in Table 1. At the end of 28 days of the experimental diets, the rats were sacrificed with light anesthesia (ethyl ether) following a 16 hr fast. Blood samples were taken from the inferior vena cava and were then centrifuged for 20 minutes at 3000 rpm at 4°C to obtain plasma. The plasma was frozen at -20°C for later determination of concentrations of total cholesterol, HDL-cholesterol, triglyceride and glucose. Concentrations of blood glucose and plasma total cholesterol, high-density lipoprotein (HDL) cholesterol, triglyceride were determined by enzymatic methods using a commercial kit (Yeongdong Pharm. Corp, Seoul, Korea). Concentration of plasma low-density lipoprotein (LDL) cholesterol was calculated by the Friedwald method (15).

Statistical analysis

Data were analyzed by the statistical analysis system (SAS) program. Values are expressed as mean ± SD. Statistical analysis for the comparison for the two different diet groups was performed using the students' t-test.

RESULTS AND DISCUSSION

This study was performed to determine the effects of garlic vinegar supplementation on body weight, plasma glucose, and plasma lipids in streptozotocin-diabetic rats. Weights of the two groups were not statistically different at the beginning of experiment (Table 2). The garlic vinegar supplementation did not have a significant effect on weight gain in the diabetic rats. The result of the present study are in agreement with our previous study (14). Also, there was no significant difference between the mean food efficiency ratio (FER) in the two diabetic groups. However, there was a significant difference between the mean FER when the garlic vinegar was supplemented to non-diabetic rats in our other study (14).

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

Ingredient	Control	Garlic vinegar[1)]
Corn starch	65.2	65.2
Casein	20	20
Corn oil	5.0	5.0
α-cellulose	3.8	3.8
Min. mix	3.5	3.5
Vit. mix	1.0	1.0
Choline	0.2	0.2
DL-methionine	0.3	0.3
Cholesterol	1	1
Garlic vinegar	0 ml	35 ml

[1)]Garlic vinegar fermented with a 20% fresh garlic juice

Table 2. Effects of garlic vinegar supplementation on body weight, food intakes, and FER[1)] in diabetic rats fed cholesterol

Variables	Diabetic control	Diabetic garlic vinegar
Body weight at beginning (g)	218.23 ± 5.76[2)]	217.07 ± 9.50[NS3)]
Body weight at sacrifice (g)	212.73 ± 29.65	215.77 ± 30.20[NS]
Food intake (g/day)	28.21 ± 2.73	30.59 ± 3.13[NS]
FER[1)]	-0.070 ± 0.044	-0.001 ± 0.041[NS]

[1)]$FER = \frac{\text{weight gain for 4 wks}}{\text{food intake for 4 wks}}$

[2)]Mean ± SD

[3)]Not significantly different at $p<0.05$

The effects of garlic vinegar supplementation on the concentrations of blood glucose, plasma cholesterol, and triglyceride in chemically-induced diabetic rats are shown in Table 3. In the present study, there were no significant effects of the garlic vinegar on plasma glucose concentration in diabetic rats. The results of the present study are consistent with an earlier study which showed that there were no significant difference in plasma glucose concentration between animals fed a garlic vinegar and control diet in non-diabetic rats (16).

In non-diabetic high-cholesterol fed rats, the plasma triglyceride concentration of rats supplemented garlic vinegar was lower than those fed a control diet (14). However, there were no changes in plasma triglyceride concentration in diabetic rats supplemented with garlic vinegar. In the U.S., Chang and Johnson (1) studied the effects of garlic on lipids synthesis in rats fed an experimental diet containing 1% cholesterol. Feeding of garlic equivalent to 5g of fresh bulbs per day for seven days resulted in a 56% decrease in total serum lipid. The results of this study does not support the earlier study (1) of the hypotriglyceridemic effect of dietary garlic in rats.

Studies both in man and experimental animals have shown that the concentration of serum cholesterol can be greatly affected by dietary garlic (1-3). In a comparison of plasma cholesterol levels in diabetic rats with garlic vinegar and matched controls, cholesterol concentrations are not significantly decreased in garlic vinegar supplemented group com-

Table 3. Effect of garlic vinegar supplementation on concentrations of plasma glucose and plasma lipidsl in diabetic rats fed a high-cholesterol diet

Variables	Diabetic control	Diabetic garlic vinegar
Glucose (mg/dl)	356.30 ± 263.25[2)]	343.71 ± 143.40[NS3)]
Triglyceride (mg/dl)	69.85 ± 21.26	76.45 ± 40.75[NS]
Total-cholesterol (mg/dl)	144.55 ± 32.56	139.89 ± 26.77[NS]
HDL-cholesterol (mg/dl)	64.63 ± 11.96	85.87 ± 21.51*
LDL-cholesterol (mg/dl)	78.70 ± 17.83	37.95 ± 20.33*
Atherogenic index[1)]	1.24	0.63*

[1)](Total Cholesterol-HDL cholesterol)/HDL cholesterol

[2)]Mean ± SD

[3)]Not significantly different at $p<0.05$ from control group.

*Significantly different at $p<0.05$ from control group.

pared to those of the control group. The results of the present study are consistent with a previous study which showed that serum cholesterol levels in garlic vinegar-fed rats were not significantly different than in control diet-fed rats (8). It is reasonable to conclude that the different effects of dietary garlic on plasma cholesterol level are due to, in part, the differences in the quantity of the garlic in the diets. Studies both in humans and experimental animals have shown that the concentration of serum cholesterol level can be greatly affected by the proportion of garlic and period of intake. The lack of effect on total cholesterol concentrations in the garlic vinegar is in agreement with others who found that low dosage of garlic had little effect in hypercholesterolemic individuals (8). In our present results, there were no effects on plasma total cholesterol concentration by garlic vinegar supplementation in diabetic rats. However, plasma LDL-cholesterol concentrations were lower in the diabetic rats supplemented with garlic vinegar (37 mg/dl) than in the diabetic control rats (78 mg/dl). And plasma HDL-cholesterol concentrations were significantly higher in the diabetic rats fed the garlic diet (85 mg/dl) than in the rats fed the control diet (64 mg/dl).

The results of the present study are in agreement with Chang and Johnson's study (1) that reports the effect of garlic on lipoproteins in patients with coronary heart disease. The group taking essential oil of garlic, 0.25 mg/kg for 10 months, had steady decrease of LDL-cholesterol accompanied by a progressive rise of HDL-cholesterol. Lowering of LDL-cholesterol has also been reported in healthy individuals ingesting raw garlic (17). Abnormalities in the lipoprotein profile contribute to accelerated atherosclerosis in diabetics (5). Diabetic-rats supplemented with garlic vinegar not only had increased HDL-cholesterol levels but had an alteration in the HDL/ LDL-cholesterol ratio in favor of a decreased athrogenic index compared to that of the control rats. Results suggest that garlic vinegar supplementation is beneficial in reducing the risk of developing atherosclerosis in diabetic rats.

SUMMARY AND CONCLUSION

Effects of garlic vinegar supplementation on blood glucose, triglyceride, cholesterol, and body weight in streptozotocin-diabetic rats were studied. Sprague-Dawley rats were fed an experimental (control or garlic vinegar supplemented) diet for 4 weeks after induction of diabetes by streptozotocin at a dose of 50 mg/kg body weight. On the basis of previously discussed results, the following conclusions could be drawn:

1. Garlic vinegar supplementation had no effect on body weight gain in diabetic rats.
2. There was no significant effect of garlic vinegar supplementation on plasma glucose and triglyceride concentrations in diabetic rats.
3. Concentrations of plasma HDL-cholesterol fed garlic vinegar supplemented diet were significantly higher ($p < 0.05$) than that of those fed control diet. And the opposite is true for plasma LDL-cholesterol.

In conclusion, this study demonstrates that garlic vinegar supplementation has marked effects on plasma LDL-cholesterol and HDL-cholesterol concentrations. Results suggest that garlic vinegar may be of potential value in antiatherogenic diets for diabetic individuals. Also these data showed that the beneficial effects of garlic vinegar in streptozotocin-induced diabetic rats can still be maintained when garlic powder is replaced by garlic vinegar as a garlic source.

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