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ARTICLE

Effects of *Kimchi* Powder or *Lactobacillus plantarum* Added Fermented Sausages on Serum Lipid and Cholesterol Levels in Rats

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Abstract

This study was performed to investigate the serum lipid and cholesterol lowering effects of *kimchi* powder or *Lactobacillus plantarum* when added to fermented sausage in rats. Male SD (Sprague-Dawley) rats were divided in three groups and administered with the following diets for 6 wk: non-fermented sausage added diets (CON), *kimchi* fermented sausage added diets (KIM), and *Lactobacillus plantarum* fermented sausage added diets (PLA). The CON has significantly ($p<0.05$) higher food intake, body weight gains, and final body weight than the other two groups. The body weight of KIM was significantly ($p<0.05$) lower than that of the other groups. The weights of organs (heart, lung, liver, kidney, spleen, perirenal adipose tissue, and epididymal adipose tissue) were not affected by any treatments. The mean value of serum triglycerides (TG) concentration in KIM and PLA groups was not significantly different compared with CON. Total cholesterol, low density lipoprotein and high density lipoprotein levels in serum of KIM were significantly ($p<0.05$) lower than those of CON. The serum free cholesterol level and atherogenic index (AI) in KIM and PLA were significantly ($p<0.05$) lower than those of CON. In conclusion, dietary supplementations with *kimchi* powder fermented sausages or *L. plantarum* fermented sausages were effective for lowering serum lipid, cholesterol levels and atherogenic index in rats.

Key words: *kimchi* fermented sausage, *Lactobacillus plantarum*, cholesterol, adipose tissue, atherogenic index

Introduction

Cardiovascular disease (CVD) is a leading cause of death in many countries around the world. By the year 2020, up to 40% of all death will be related to cardiovascular or heart disease (WHO, 2002). Higher serum total cholesterol level is generally considered to be a risk factor for coronary heart disease and atherosclerosis. Therefore, the reduction of plasma cholesterol level decreases the incidence and mortality of ischemic heart disease (IHD) and atherosclerosis (Greenwald, 1991).

Meanwhile, consumption of fermented dairy products containing probiotics has been proposed as a means to lower serum cholesterol (Kawase *et al*., 2000; St-Onge *et al*., 2000). This fact stimulates interests in the cholesterol lowering effects of fermented milks and lactic acid bacteria (Mann and Spoerry, 1974). Many studies have been performed with experimental animals and humans to elucidate the effect of fermented dairy products on serum cholesterol, especially with selected strains of lactic acid bacteria (St-Onge *et al*., 2000). Hypocholesterolemic effect of fermented milk products in animals was reported (Rao *et al*., 1981; Rodas *et al*., 1996; Taranto *et al*., 1997). Standard yogurt and *bifidus* yogurt inoculated with *Bifidobacteria* and *Lactobacilli* commonly used as probiotics (Been and Prasad, 1997).

Kimchi is one of traditional functional food from Korea, and its probiotic effect has been reported in many research articles (Chang *et al*., 2010; Lee *et al*., 2005; Lee *et al*., 2011; Wang *et al*., 2010). Lee and Kunz (2005) had reported that when its raw form or freeze-dried powder form has been added in fermented sausages, the antioxidant effect has been investigated.

However, there was not enough information about the probiotics of *Kimchi* added fermented meat products. There-

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fore, the purpose of this study is investigating the plasma lipid and cholesterol levels in fermented sausage-diet rats in order to investigate the probiotic effect of *Kimchi* powder addition.

Materials and Methods

Preparation of *Kimchi* and *L. plantarum* fermented sausage samples

The preparation of *Kimchi* was carried out based on the *Baechu-kimchi* recipe (Lee *et al*., 2005) and the fermentation condition was 10°C for 15 d (Table 1). *Kimchi* was grounded and freeze-dried to prepare the *Kimchi* fermented sausage. Fermented sausage was prepared by modified method of Lee *et al.* (2005). The meat mixture was stuffed in natural casing with minced freeze-dried kimchi powder or *L. plantarum* at levels of 5%, respectively and fermented at 15°C for 15 d.

Animal experiments

Thirty male SD rats (177±1.0 g body weight, four-wk old) were purchased from Central Laboratory Animal Inc. (Korea). They were housed in air-conditioned room at 22-24°C with 60-70% humidity and 12-h light-dark cycles (07:00-19:00). The base composition of the experimental diet is shown in Table 2. The rats were randomly divided into three groups and fed one of the following diets: non-fermented sausage added normal diet (CON), *Kimchi* fermented sausage added normal diet (KIM), and 5% *Lactobacillus plantarum* fermented sausage added nor- mal diet (PLA). The experimental diets were manufactured based on AIN-76 (Reeves *et al*., 1993). The feed and water were administered *ad libitum*. After 6 wk, the rats were anesthetized by Zoletil (Zoletil 50, Virbac, France) injection and the blood was collected from the abdominal aorta. Subsequently, the liver, heart, kidney and spleen were quickly removed, weighted and frozen in liquid nitrogen in separated portions. Perirenal adipose tissue (PAT) and epididymal adipose tissue (EAT) were also immediately removed and weighed.

Table 1. Composition of *baechu-kimchi* materials

Materials and ingredients	Percentage
Salted Chinese Cabbage	90
Leek	4
Red pepper powder (Korean)	2
Garlic	2
Ginger	1
Sugar	1

(Lee *et al*., 2005)

Body weight and food intake

The total amount of food intake for each rat was recorded daily and their body weight was measured once a week during the raising period.

Serum lipid determination

Serum was acquired by centrifugation of blood samples in the condition of 3,000 *g* for 15 min at 4°C. The level of total cholesterol (TC), high-density lipoprotein (HDL) cholesterol, low-density lipoprotein (LDL) cholesterol, triglyceride (TG), total lipid, free cholesterol, and phospholipids were analyzed by using Cholesterol E Test Wako (Wako Pure Chemical Industries Ltd., Japan), respectively (Friedewald *et al*., 1972).

Atherogenic index calculation

The AI (Atherogenic Index) was calculated as:

AI = (total cholesterol − HDL cholesterol) / HDL cholesterol

Statistical analysis

The calculations were performed using the SAS (statistical analysis system, USA). All data are expressed as mean ±SD (standard deviation). One way ANOVA was used to

Table 2. Composition of experimental diets (%)

Ingredients	Non-fermented sausage	*Kimchi* fermented sausage	*L. plantarum* fermented sausage
Casein	20	20	20
Corn starch	60	60	60
Non-fermented sausage	5	-	-
Kimchi fermented sausage	-	5	-
L. plantarum fermented sausage	-	-	5
Soybean oil	10	10	10
Mineral mixture	4	4	4
Vitamin mixture	1	1	1
Total volume	100	100	100

CON: Meat mixture without *kimchi* or starter culture. KIM: Meat mixture + 10% *kimchi* powder. PLA: Meat mixture + 5% starter mixture with commercial *L. plantarum,* 1.0×10^7 CFU/mL.

Table 3. The change of body weight and food intake for 6 wk

Group	Body weight (g)			Food intake (g/d)
	Initial	Final	Weight gains	
CON	176.3±4.4 ^{NS}	406.1±12.6 ^{a}	229.8±8.2 ^{a}	42.1±5.1 ^{a}
KIM	177.4±2.4	382.8±6.0 ^{b}	205.4±3.6 ^{c}	27.8±5.1 ^{b}
PLA	179.5±5.1	396.2±10.9 ^{ab}	216.7±5.1 ^{b}	33.4±5.2 ^{ab}

All values are mean±SD. (n=10).
CON: non-fermented sausage added AIN-76 diet. KIM: *kimchi* fermented sausage added AIN-76 diet. PLA: *L. plantarum* (1.0×10^7 CFU/mL) fermented sausage added AIN-76 diet.
^{abc}Superscripts with different letters are statistically different ($p<0.05$).
^{NS}not significant.

determine the significance of the differences among the variables of the three experimental groups and followed by Duncan's multiples range test for difference between means. The level of significance was $p<0.05$.

Results and Discussion

Body weight, food intake and organ weights

The initial body weight and the amount of food intake were not significantly different among the groups. CON group has significantly higher food intake, body weight gains, and final body weight than the other groups ($p<0.05$, Table 3). The body weight of KIM was significantly lower than that of the other groups ($p<0.05$). We estimated that the phenomenon of reduced body weight of KIM and PLA was induced by the reduced food intake and by other factors including the probiotic effect of *kimchi* fermented bacteria in the *kimchi* powder. However, there were no significant differences in weights of heart, liver, kidney, spleen, PAT, and EAT among the three groups (Table 4).

Table 5. Serum lipid level in rats

Group	Triglyceride	Total lipid	Phospholipid
CON	56.0±2.00^{NS}	224.4±18.54^{a}	96.3±9.32^{NS}
KIM	52.4±7.62	190.2±14.92^{b}	95.6±3.42
PLA	54.4±7.70	218.5±9.53^{ab}	98.8±4.58

All values are mean±SD. (n=10).
CON: non-fermented sausage added AIN-76 diet. KIM: *kimchi* fermented sausage added AIN-76 diet. PLA: *L. plantarum* (1.0×10^7 CFU/mL) fermented sausage added AIN-76 diet.
^{abc}Superscripts with different letters are statistically different ($p<0.05$).
^{NS}not significant.

Plasma lipids, cholesterol level and atherogenic index

The serum lipid composition was presented in Table 5. The mean value of serum TG concentration in KIM and PLA groups was not significantly different. Serum total lipid in KIM group was significantly reduced than that of the CON group ($p<0.05$).

The concentration of plasma TC, HDL, LDL, free cholesterol, and AI in CON were significantly higher than those of KIM group ($p<0.05$). Especially, free cholesterol level of KIM and PLA was significantly ($p<0.05$) lower than that of CON group. These results suggest that the *kimchi* powder or *L. plantarum* consumption may adjusts the serum lipid and cholesterol levels in rats, as well as the increased risk of senile arteriosclerosis which was induced by meat product consumption might by also reduced.

L. plantarum is one of the most well studied probiotics with hypocholesterolemic effects (Jeun *et al.*, 2010). The mechanism of lipid regulation associated with *L. plan-*

Table 4. The weight of organs in rats

Group	Heart	Liver	Kidney	Spleen	Peri-renal adipose tissue	Epididymal adipose tissue
CON	1.15±0.08	10.9±0.68	2.67±0.21	0.66±0.05	7.2±1.85	7.8±0.54 ^{NS}
KIM	1.14±0.07	10.2±0.55	2.67±0.23	0.75±0.01	7.5±0.77	7.9±0.40
PLA	1.13±0.09	11.1±0.36	2.56±0.08	0.71±0.11	6.2±1.28	6.6±1.60

All values are mean±SD. (n=10).
CON: non-fermented sausage added AIN-76 diet. KIM: *kimchi* fermented sausage added AIN-76 diet. PLA: *L. plantarum* (1.0×10^7 CFU/mL) fermented sausage added AIN-76 diet.
^{NS}not significant.

Table 6. The cholesterol level and atherogenic index of serum in rats

Group	Total cholesterol	HDL cholesterol	LDL cholesterol	Free Cholesterol	AI
CON	70.1±7.67^{a}	52.7±8.54^{a}	13.0±2.62^{a}	3.42±0.97^{a}	0.33±0.10^{a}
KIM	54.6±4.62^{b}	42.8±3.92^{b}	9.4±1.52^{b}	2.42±0.97^{b}	0.17±0.19^{b}
PLA	61.3±8.81^{ab}	49.1±9.53^{ab}	10.8±1.58^{ab}	1.50±0.55^{c}	0.18±0.07^{b}

All values are mean±SD. (n=10).
CON: non-fermented sausage added AIN-76 diet. KIM: *kimchi* fermented sausage added AIN-76 diet. PLA: *L. plantarum* (1.0×10^7 CFU/mL) fermented sausage added AIN-76 diet. AI: Artherogenic Index.
^{abc}Superscripts with different letters are statistically different ($p<0.05$).
^{NS} not significant.

tarum has been described as cholesterol assimilation by the bacteria and bile salt hydrolase deconjugate of bile salt (Jeun *et al.*, 2010). Also Cha *et al.* (2012) reported that *L. plantarum* CIB 001 from *kimchi* decreased serum triglyceride, total cholesterol and LDL cholesterol levels in hypercholesterolemic rats. These similar results suggest that *kimchi* fermented sausage also has serum lipid and cholesterol lowering effects in animal models.

Conclusions

This study was designed for investigating the plasma lipid and cholesterol lowering effect of *Kimchi* powder or *L. plantarum* added fermented sausage in rats. The present results suggested that *Kimchi* powder intake may very effective to decrease serum lipid, cholesterol levels and AI in rats. Therefore, *Kimchi* powder fermented sausage plays very important roles to produce the low cholesterol functional meat products.

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