

### SIV-3

## **FUNCTIONAL PROPERTIES OF SAENGSIK (SUBSTITUTE MEAL) FORMULATED WITH UNCOOKED CEREALS FERMENTED BY LACTIC ACID BACTERIA**

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### **Abstract**

The objective of this study was to improve the qualities of Saengsik (substitute meal formulated with uncooked cereal powder) such as functionality, safety and sensory preference by using Lactic Acid Bacteria (LAB). The optimum formulation of fermented Saengsik (substitute meal) was verified as 40% of fermented brown rice, 13% of fermented job tears, 5% of yellow soybean, 5% of waxy rice, 5% of sorghum, 3% of Seumok bean, 1% of sesame, 3% of barley, 2% of red bean, 5% of proso millet, 2% of foxtail millet, 9% of black rice, 3% of buckwheat, 1% of chestnut, 2% of pumpkin, 3% of peanut.

Fermented Saengsik decreased significantly serum glucose in STZ-induced diabetic rats. However, serum lipid levels were not affected by fermented Saengsik. In rats fed with high-fat diet, serum triglyceride level was slightly decreased and the contents of liver triglyceride were significantly decreased by supplementation of fermented Saengsik.

**Key words:** Saengsik, Fermented cereals, Lactic acid bacteria, Diabetics, Serum glucose, Serum lipid

### **INTRODUCTION**

The culture of food has been rapidly changed due to the higher economical level of the people and by converting to a very small family in the industrialized society.

The special points of the current culture of the food were higher usage of the processed foods, instant foods and animal foods. These kinds of culture of food ingestion such as hyper nutrition, over ingestion of animal lipid, etc. induce cerebrocardiovascular diseases by obesity. These problems are thought to be a threat in the natural economic due to gigantic medical-cost burden.

By the way, the ingestion of meat was not a bad problem totally. If the ingestion of meat was maintained in the optimal level, it does not a big problem in health as well as it is essential to children and the youth. The suitably balanced menu with meat and vegetables are strongly recommended.

Considering our culture of the food, the balanced healthy food including various grains, beans, vegetable oil etc. is required to people for supplementing the lack of nutritional level of cooked rice.

Brown rice, barley, waxy rice, black bean, job ´ tears, perilla, black sesame, yellow soybean contain most of all essential nutrients. While germinating many useful contents in brown rice such as enzymes, vitamins etc. are produced and activated. When the powder is fermented at optimal condition, microorganisms are grew and produces various enzymes and nutrients. However, the condition for fermentation is not maintained appropriately, spoilage bacteria can grow up and induces food spoilage.

Consequently, development of new nutrient supplemented functional foods by using these kinds of materials is thought to be an important thing for national health.

## **MATERIALS AND METHODS**

### **Materials**

Brown rice and job ´ s tears produced in 1999 were purchased from commercial market, ground to 60 mesh and used for fermentation. Male Sprague-Dawley rats were obtained from DaeHan Biolink Co. LTD. Experimental feeding materials such as casein, mineral mixture, vitamin mixture, cellulose, chloine chloride, methionine were purchased from Teklard Co. and corn starch were purchased commercial market. Streptozotocin was purchased from Sigma Co. and glucose, cholesterol, triglyceride measuring kit were purchased from Shinyang Chemical Co.

### **Methods**

#### **1) Fermentation**

The brown rice and job ´ tears mixture (1:1) containing 2 10% of honey and 5 30% of distilled water were fermented in incubator at 25 35 for 5 days.

Four kinds of Lactic Acid Bacteria (LAB) were obtained from Mediogen co.

The 0.3% of dried LAB powder were fermented in the brown rice and job ´ tears mixture (1:1) containing 5% of honey and 30% of distilled water at 30 for 3 days.

#### **2) Formulation of Fermentated Saengsik**

The formulation was confirmed by experiment on the addition of sugar and water to the traditional formulation of Lifes Co.

#### **3) Animal Study**

##### **Effect of Fermented Saengsik on the Serum Glucose and Lipid Levels of STZ- Induced Diabetic Rats**

Male Sprague-Dawley rats(8weeks) were obtained from Daehan Biolink Co. LTD., and

acclimated to the laboratory for one week. The rats were divided into three groups such as normal group, diabetic control group and fermented Saengsik group. Experimental diets based on AIN 76 composition (Table 1). Diabetes in rats were induced by intramuscular injection of STZ at the level of 50 mg/kg B.W. after 3 weeks of experimental diets. The diabetic rats were fed with experimental diets further 4 weeks and anesthetized with ether, blood was drawn from abdominal vena cave. Serum glucose, total-cholesterol and triglyceride were measured by using enzymatic kits(Shinyang Chemical Co.)

**Table 1. Composition of experimental diet for diabetic test**

(unit : g)

| Materials          | Normal | White rice | Fermented grain |
|--------------------|--------|------------|-----------------|
| Casein             | 200    | 166        | 140             |
| Corn oil           | 50     | 43.5       | 15              |
| Mineral mix        | 35     | 35         | 35              |
| Vitamin mix        | 10     | 10         | 10              |
| Cholin Cl.         | 2      | 2          | 2               |
| Methionine         | 3      | 3          | 3               |
| Cellulose          | 50     | 50         | 50              |
| Sucrose            | 50     | 50         | 50              |
| Corn starch        | 600    | 140.5      | 195             |
| White rice         | -      | 500        | -               |
| Fermented Saengsik | -      | -          | 500             |

**Effect of Fermented Saengsik on the Lipid Contents of Rats Fed with High-Fat Diets**

Male rats (8 weeks) acclimated to environment for one week were divided into three groups by randomly as follows ; high fat control group, high fat control group with white rice and high fat control group with fermented Saengsik. Table 2 shows the composition of experimental diet based AIN 76 diet. Corn starch, white rice powder and fermented Saengsik were added as a carbohydrate source to each diet. Rats were allowed free access to experimental diets and water for 4 weeks. Total dietary intakes were measured at 2 days interval and the changes in body weight were measured in every week. <sup>6,7)</sup>

Samples and serum were taken by the same above method and the contents of serum lipid were analyzed by the enzymatic kit (Shinyang Co.)

The mean and standard error of every data were acquired and were analyzed by ANOVA and

**Table 2. Composition of experimental diet**

(unit : g)

| Materials          | Control | White rice | Fermented Saengsik |
|--------------------|---------|------------|--------------------|
| Casein             | 200     | 166        | 140                |
| Corn oil           | 50      | 43.5       | 15                 |
| Lard               | 100     | 100        | 100                |
| Mineral mix        | 35      | 35         | 35                 |
| Vitamin mix        | 10      | 10         | 10                 |
| Cholin Cl.         | 2       | 2          | 2                  |
| Methionine         | 3       | 3          | 3                  |
| Cellulose          | 50      | 50         | 50                 |
| Sucrose            | 50      | 50         | 50                 |
| Corn starch        | 500     | 40.5       | 95                 |
| White rice         | -       | 500        | -                  |
| Fermented saengsik | -       | -          | 500                |

group difference were considered statistically significant at  $p < 0.05$  by Duncan's multifal range test.

## RESULTS AND DISCUSSION

### Fermentation Experiment

The analytical results of changes in organic acids, pH and flavor after fermentation by various Lactic Acid Bacteria(LAB) were shown in Table 3.

As a result of Table 3, MG311 produced the most amount of organic acid (0.752%) and generated sour and fresh odor and taste. MG19 produced 0.702% of organic acid and its flavor was good as well. Although MG89 produced a little amount of organic acid (0.501%), the flavor was so good. MG208 produced only 0.301% of organic acid and very low rate of fermentation with spoilage odor. The pH

**Table 3. Changes in organic acids, pH and flavor after fermentation by various Lactic Acid Bacteria(LAB)**

| LAB       | Organic acid(%) | pH   | Flavor                                  |
|-----------|-----------------|------|---|
| MG19*     | 0.702           | 5.38 | good flavor                             |
| MG89**    | 0.501           | 5.99 | good flavor                             |
| MG208***  | 0.301           | 6.49 | a little spoilage flavor                |
| MG311**** | 0.752           | 4.90 | good flavor containing alcoholic flavor |

\*MG19 : *Lactobacillus brevis*

\*\*MG89 : *Streptococcus faecium*

\*\*\*MG208 : *Lactobacillus plantarum*

\*\*\*\*MG311 : *Lactobacillus casei*

ranges of every samples were between 4.9 to 6.49 and were related to contamination and production rate of organic acid.<sup>8)</sup> The number of aerobic bacteria and *E. coli* in fermented product after 4 kinds of treatments were shown in Table 4.

**Table 4. The number of aerobic bacteria and *E. coli* in fermented product after 4 kinds of treatments**

| LAB   | <i>E. coli</i> /g | Aerobic plate count/g | Contamination |
|-------|-------------------|-----------------------|---------------|
| MG19  | Negative          | 1.6 10 <sup>7</sup>   | No            |
| MG89  | Negative          | 5.4 10 <sup>8</sup>   | No            |
| MG208 | Negative          | 5.0 10 <sup>7</sup>   | Yes           |
| MG311 | Negative          | 5.3 10 <sup>8</sup>   | No            |

As a result of Table 4, *E. coli* were all negative. Aereobic plate counts (APC) of MG89 and MG311 were similar (5 10<sup>8</sup>/g) but much lower than MG19 (1.6 10<sup>7</sup>/g) as well as MG208 (5.0 10<sup>7</sup>/g) which were contaminated.

These results shows that the production of organic acid was not correlated to

APC and are thought to be due to the characteristics of LAB. The best LAB which produced the highest amount of organic acid and good flavor was verified as MG311. Therefore, the single use of MG311 as well as the mixture with MG19 was recommended.

### **Formulation of Fermented Saengsik**

The optimum formulation of fermented Saengsik was as in Table 5.

### **Animal Study**

#### **Effect of Fermented Saengsik on the Serum Glucose and Lipid Levels of STZ-Iduced Diabetic Rats**

Although the body weight of normal group was continuously increased, the body weights of diabetic groups were drastically decreased after induction of diabetes. The body weights of both diabetic control group and fermented Saengsik group were not significantly different. The weights of both liver and spleen of diabetic groups were significantly lower than the normal. This is thought to be a decrease in body weight of the diabetic group.<sup>1)</sup> Although the weight of kidney of both the normal and the diabetic group were almost similar, the weight of kidney/100g of body weight of the diabetic group were much higher than the normal. This means unusual enlargement of kidney.

The changes in contents of serum triglyceride and total cholesterol of experimental group were shown in Table 6. The contents of serum triglyceride and total cholesterol in the normal group were

**Table 5. The optimum formulation of fermented Saengsik**

| Raw materials           | Usage |      |
|-------------------------|-------|------|
|                         | (%)   | (g)  |
| Fermented brown rice    | 40    | 16.0 |
| Fermented job ´ s tears | 13    | 5.2  |
| Yellow soybean          | 5     | 2.0  |
| Waxy rice               | 5     | 2.0  |
| Sorghum                 | 3     | 1.2  |
| Seumok bean             | 3     | 1.2  |
| Sesame                  | 1     | 0.4  |
| Barley                  | 3     | 1.2  |
| Red bean                | 2     | 0.8  |
| Prosmillet              | 5     | 2.0  |
| Foxtail millet          | 2     | 0.8  |
| Black rice              | 9     | 3.6  |
| Buckwheat               | 3     | 1.2  |
| Chestnut                | 1     | 0.4  |
| Pumpkin                 | 2     | 0.8  |
| Peanut                  | 3     | 1.2  |
| Total                   | 100   | 40   |

(40g / 1Pouch)

**Table 6. The changes in contents of serum triglyceride and total cholesterol of each treated group**

| Treatments               | Serum triglycerides(mg/dl) | Total cholestrol(mg/dl)  |
|--------------------------|----------------------------|--------------------------|
| Normal group             | 180.93±33.42 <sup>a</sup>  | 78.84± 5.88 <sup>a</sup> |
| Diabetic control group   | 63.77±29.12 <sup>b</sup>   | 48.23±11.58 <sup>b</sup> |
| Fermented Saengsik group | 48.51± 9.72 <sup>b</sup>   | 52.20± 5.02 <sup>b</sup> |

significantly higher than diabetic group. There was no significant difference between diabetic control group and fermented Sangsik group.<sup>2)</sup>

Table 7 showed the changes in serum glucose level during experiment of the diabetic states.

The glucose levels in diabetic groups were significantly higher than normal group, and increased continuously during experimental period . After 3 weeks of the experimental diets, the contents of serum glucose in the fermented Saengsik group were significantly decreased compared to diabetic control group.<sup>3,4)</sup>

**Table 7. The changes in serum glucose levels of each treated group**

| Treatments               | 1 <sup>st</sup> week      | 2 <sup>nd</sup> week      | 3 <sup>rd</sup> week       | 4 <sup>th</sup> week       |
|--------------------------|---------------------------|---------------------------|----------------------------|----------------------------|
| Normal group             | 83.38± 5.43 <sup>b</sup>  | 91.88± 4.13 <sup>b</sup>  | 101.75± 7.29 <sup>b</sup>  | 147.69±16.43 <sup>b</sup>  |
| Diabetic control group   | 346.57±13.73 <sup>a</sup> | 267.25±28.51 <sup>a</sup> | 323.42±33.58 <sup>a</sup>  | 362.87±33.80 <sup>a</sup>  |
| Fermented Saengsik group | 324.86± 9.60 <sup>a</sup> | 204.00±31.82 <sup>a</sup> | 227.00±51.61 <sup>ab</sup> | 238.18±46.10 <sup>ab</sup> |

### Effect of Fermented Saengsik on Lipid Content of Experimental Rats

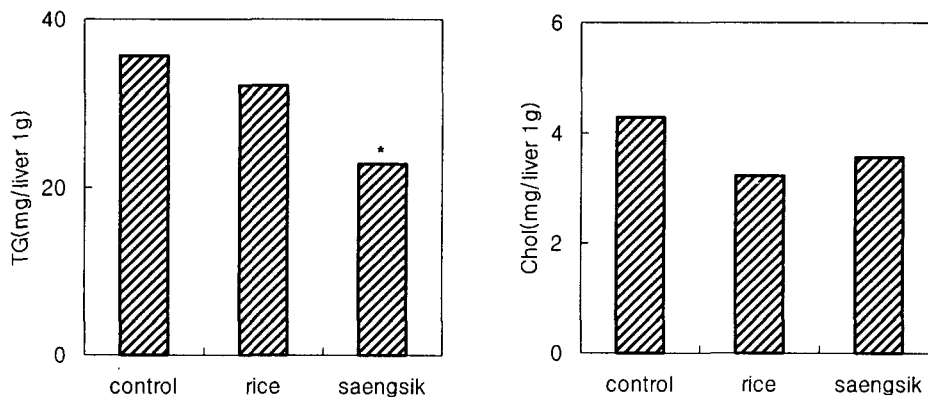
Weight gains in the fermented Saengsik group were slightly increased compared to other groups and a little higher than the normal. Daily total dietary intake of all treated groups were not significantly different. There was no significant difference in the weights of liver, kidneys and spleen among experimental groups. The effects of fermented Saengsik on the contents of serum triglyceride and total cholesterol in each treated group were shown in table 8. The contents of serum triglyceride of fermented Saengsik group were also slightly decreased compared to white rice group and significantly lower than high-fat control group . The contents of total cholesterol of both the high fat control group and the white rice group were 73mg/dl and 75mg/dl, respectively and were not significantly different. However, the fermented Saengsik group showed about 14% lower than other groups but were not significantly different. <sup>5,6)</sup>

The effects of fermented Saengsik on the contents of liver triglyceride and total cholesterol in each treated group were shown in figure 1. The results of liver triglyceride showed the similar with the serum triglyceride data. The contents of liver triglyceride of the high fat control group and the white rice group were 35 mg/g and 32 mg/g, respectively and were significantly higher than the fermented Saengsik group (22 mg/g). The contents of total cholesterol of the high fat control group (4.3mg/g liver) was higher than the white rice group (3.2mg) and the fermented Saengsik group (3.6mg/g) but these were not significantly different. <sup>7)</sup>

**Table 8. The contents of serum triglyceride and total cholesterol in each treated group**

| Treatments               | Triglycerides (mg/dl)    | Total cholesterol (mg/dl) |
|--------------------------|--------------------------|---------------------------|
| High-fat control group   | 85.18±3.44 <sup>a</sup>  | 73.24±3.48 <sup>ns</sup>  |
| White rice group         | 76.34±2.66 <sup>ab</sup> | 75.32±3.09                |
| Fermented Saengsik group | 65.21±2.73 <sup>b</sup>  | 65.90±4.32                |

Therefore, the supplementation of fermented Saengsik is thought to be an useful mean for decrease of serum and liver triglyceride contents in the rat fed with high-fat diet.



**Figure 1. The contents of liver triglyceride and total cholesterol in each treated group**

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