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Effect of soybean oligosaccharides on blood lipid, glucose levels and antioxidant enzymes activity in high fat rats

Hua Chen [a], Liu Li-jun [a,*], Zhu Jian-jun [a], Xu Bo [a], Li Rui [b]

[a] Department of Emergency, The Second Affiliated Hospital of Soochow University, Suzhou 215004, China
[b] Department of Gastroenterology, The First Affiliated Hospital of Soochow University, Key Laboratory of Medicine and Clinical Immunology of Jiangsu Province, Suzhou 215006, China

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ABSTRACT

The effect of soybean oligosaccharides on blood lipid levels and oxidative stress in rats fed on high-fat diet was investigated. Rats were divided into five groups of 10 animals each. The high-fat group received a high-fat diet containing 18% (w/w) lipid in the diet (36% of total energy). Animals allocated to the soybean oligosaccharides-treatment groups (I, II and III) received the high-fat diet and orally fed with soybean oligosaccharides at a single dose of 150, 300 and 450 mg/kg body weight, respectively. Control rats received basic diet. Results showed that soybean oligosaccharides significantly reduced abnormal blood glucose, lipid level and oxidative stress in animal models at all doses examined. Soybean oligosaccharides were able to reduce oxidative stress and improve abnormal blood lipid levels induced by high-fat diets. In summary, the present study may be important for reverse cardio-cerebrovascular disease.

1. Introduction

Soybeans are unique foods because of their rich nutrient content. They contain complex carbohydrates, protein, dietary fibre, oligosaccharides, phytochemicals (especially the isoflavones in soy) and minerals (Refstie, Storebakken, & Roem, 1998). Carbohydrates are the second largest component in soybeans. Their complex carbohydrates and dietary fibre contents contribute to their low glycemic index, which benefits diabetic individuals (Jenkins, Wolever, & Taylor, 1981) and reduces the risk of developing diabetes (Salmeron et al., 1997). Soybean seed is a rich source of oligosaccharides, namely raffinose and stachyose: raffinose is a trisaccharide containing galactose linked α-(1–6) to the glucose unit of sucrose; stachyose is a tetrasaccharide containing a galactose linked α-(1–6) to the terminal galactose unit of raffinose (Kim, Kim, & Hwang, 2003). Other reported major sugar of soybeans is sucrose with lower amounts of the monosaccharides, fructose, rhamnose and arabinose; significant levels of glucose occurred only in immature seeds (Van der Riet, Wight, Cilliers, & Datel, 1989).

High-fat diets are reported to increase oxidative stress in a variety of tissues, which may result in many degenerative diseases (Chen, Zhong, Zeng, & Ge, 2008; Lieber et al. 2007; Ma, Liu, Yu, Chen, & Zhang, 2009; Schreibelt et al., 2007). Antioxidant supplementation prevents many diseases attributed to high-fat diet (Chen, Shen, & Chen, in press; Chen, Zhong, Zhu, Zeng, & Dai, 2009; Hong, Wu, Ma, Liu, & He, 2009; Zhu, Wang, Zhang, Pei, & Fen, 2008). There have been several reports describing the biological activities of soybean oligosaccharides such as antioxidant, blood pressure lowering and antidiabetic activities (Huang et al., 2006; Zhao & Yang, 2007). Soybean oligosaccharides and various derivatives can stabilise lipids in formulated foods. Huang et al. (2006) have reported the effect of soybean oligosaccharides on antioxidant enzymes and the immunity activity of broilers. Deng, Mai, Ai, and Zhang (2007) reported that dietary soybean oligosaccharides (SBOS) could decrease the incidences of fatty liver of the fish fed soy protein isolate (SPI)-based diets.

In this work, the effect of soybean oligosaccharides on blood lipid, glucose levels and antioxidant enzyme activity in HF rats was studied.

2. Materials and methods

2.1. Materials

Soybean oligosaccharides (SBOS) were purchased from XiAn ChenXing Plant Science Technology Co. Ltd. (XiAn, China).

Abbreviations: SBOS, soybean oligosaccharides; TC, total cholesterol; LDL-c, low-density lipoprotein cholesterol; TG, triacylglycerols; HDL-c, high density lipoprotein cholesterol; HF, high fat; SOD, superoxide dismutase; CAT, catalase; GSH-Px, glutathione peroxidase; AI, atherosclerosis index; MDA, malondialdehyde.

* Corresponding author. Tel./fax: +86 0512 68282030.
E-mail address: liu_ljdr@yahoo.com.cn (L.-j. Liu).

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2.2. Animals, diets and experimental design

All animal protocols were approved by the animal care and use committee of Suzhou University (Suzhou City, China). Wistar rats, aged 4 weeks of age, were obtained from the Laboratory Animal Center of our university. The rats were preferred as a model of spontaneous human obesity over other genetically altered animals because when wistar rats are fed high-fat diets (Table 1), they become obese, hyperinsulinemic, and hyperlipidemic and display characteristics of metabolic syndrome. The rats were kept at a constant temperature of 22 °C and exposed to a 12/12 h (light/dark) cycle. After adaptation for 1 week, rats were divided into five groups of 10 animals each: control group; HF model group; oligosaccharides-treated group (I); oligosaccharides-treated group (II); and oligosaccharides-treated group (III).

Rats in control group were allowed free access to basic diet, water and orally administrated with the same volume of physiological saline for 45 consecutive days.

Rats in HF model group were allowed free access to high-fat diet, water and treated with the same volume of physiological saline for that same period.

Rats of oligosaccharides-treated group (I) were allowed free access to high-fat diet, water and were treated by oral infusion with oligosaccharides at a dose of 150 mg/kg BW/day dissolved in physiological saline for that same period.

Rats of oligosaccharides-treated group (II) were allowed free access to high-fat diet, water and were treated by oral infusion with oligosaccharides at a dose of 300 mg/kg BW/day dissolved in physiological saline for that same period.

Rats of oligosaccharides-treated group (III) were allowed free access to high-fat diet, water and were treated by oral infusion with oligosaccharides at a dose of 450 mg/kg BW/day dissolved in physiological saline for that same period.

Body weight was recorded weekly. On completion of the experiment, all rats were weighed and blood was collected via the postcaval vein from anesthetised animals into blood collection tubes between 09:30 and 10:00 h after 12 h of food deprivation. Plasma was prepared by centrifugation of blood at 1000×*g* for 15 min at 4 °C and stored at −80 °C until analysed. Immediately after blood collection, rats were killed by decapitation and livers were then removed, weighed and stored at −80 °C until used for preparation of biochemical analysis.

2.3. Biochemical analysis

2.3.1. Blood lipid profile analysis

Plasma total cholesterol (TC), triacylglycerols (TG), low-density lipoprotein cholesterol (LDL-c), and high-density lipoprotein cholesterol (HDL-c) levels were measured by enzymatic and colorimetric methods, using assay kits from Sigma Diagnostics (St. Louis, MO, USA).

2.3.2. Antioxidant enzymes activities analysis

The SOD activity in the liver and blood was assayed by the inhibition of xanthine/xanthine oxidase mediated reduction of cytochrome c as previously described method (Flohe & Ötting, 1984. One unit of SOD activity in the liver and blood was defined as the amount of enzyme required to give 50% inhibition in the typical calibration curve obtained with standard SOD and was expressed as U/mg protein (ml serum).

The GSH-Px activity was determined in the liver and blood by the method reported by Paglia and Valentine (1975), respectively. In brief, tissue homogenates were centrifuged at 600×*g* for 10 min at 4 °C to remove crude fractions. Then, supernatants were centrifuged at 10 000×*g* for 20 min. One unit of enzyme activity has been defined as *n* moles of NADPH consumed/min/mg protein based on an extinction coefficient of 6.66 mM/cm.

Table 1
Diet composition (g/kg dry matter basis).

Ingredients (g/kg)	Basic diet	High fat
Casein	140	140
Wheat bran	160	160
Sucrose	95	95
Lard	40	160
tert-Butylhydroquinone	0.007	0.007
Minerals mixture	34	34
Vitamin mixture	11	11
L-Cystine	1.8	1.8
Corn starch	435.58	315.58
Choline bitartrate	2.5	2.5
Cellulose	80.12	80.12
Total	1000.07	1000.07

Rats of control group consumed 18.8 ± 0.92 g chows per day, approximately providing 59.7 ± 3.5 kcal daily.
Rats of HF group and oligosaccharides-treated groups (I, II and III) consumed 19.1 ± 0.88 g chows per day, approximately providing 132.5 ± 6.4 kcal daily.
HF: high fat.

CAT activity was assayed by the method described by Trombino et al. (2009). The enzyme-catalysed decomposition of H_2O_2 was measured. In brief, 0.5 ml aliquot of cold CAT sample and a blank consisting of 0.5 ml distilled water was taken in test tubes and the enzymatic reactions was initiated by adding 5 ml of cold 6 mM H_2O_2 and mixed thoroughly. After exactly 3 min the reaction was stopped by rapidly adding 1 ml 3 M H_2SO_4 and mixed thoroughly. Then 7 ml of 0.01 M $KMnO_4$ reagent was added, mixed thoroughly and the reading was taken at 480 nm within 30–60 s.

Serum glucose were analysed by the enzymatic reaction method using a commercially available kit by DiaSys Diagnostic Systems (Holzheim, Germany).

The concentration of TBARS was measured by a modification of the method of Yagi (1984) and calculated as malondialdehyde (MDA) equivalents using a commercial kit (Oxi-Tek, Zeptometrix Corporation, Buffalo, NY, USA).

Liver index was calculated by the formula: liver index = (liver weight/body weight) × 100. Atherosclerosis index (AI) was calculated by the formula: atherosclerosis index = (serum total cholesterol − HDL-c)/HDL-c.

2.4. Statistical methods

Analysis was performed according to the intention to treat principle. All data are expressed as mean ± SEM. Statistical analyses were performed with SPSS version 11.5 (SPSS Institute, Chicago, IL, USA). Student *t* test was used to assess the statistical significance of the continuous variables. A value of $P < 0.05$ was used as a criterion for statistical significance.

3. Results

3.1. Effect of SBOS on rats' body weight

As shown in Table 2, body weight in the HF group was significantly higher than that in the control group ($P < 0.05$). There were no significant differences between the HF group and SBOS-treated group ($P > 0.05$).

3.2. Effect of SBOS on rats' serum TC, TG, LDL-c, HDL-c levels and AI

Table 3 showed that effect of SBOS on rats' serum TC, TG, LDL-c, HDL-c levels and AI. Serum TC, TG, LDL-c levels, AI in the HF group were significantly higher, whereas HDL-c level was significantly