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Effects of Artemisia Selengensis Dietary Fiber on the Regulation of Blood Glucose and Lipids and Intestinal Flora in Diabetic Mice

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ABSTRACT

The aim of this study was to investigate the regulation effect of soluble dietary fiber from Artemisia selengensis on blood glucose and lipid in diabetic mice, and its effect on intestinal flora. SPF grade C57BL/6 mice were selected, blank group was fed basic diet, and other mice were fed high-sugar and high-fat diet for 8 weeks, and then intraperitoneally injected streptozotocin (STZ) solution to establish a diabetic mouse model. After successful modeling, the mice were randomly divided into model control group and Artemisia selengensis fiber group. The blank group was fed the basic diet, the model control group was fed the high-sugar and high-fat diet, and the Artemisia selengensis cellulose group was fed the high-sugar and high-fat diet containing 5.0% Artemisia selengensis cellulose for 4 consecutive weeks. After the feeding, blood glucose and lipid related indexes and intestinal flora composition were detected. The results showed that Artemisia selengensis cellulose could reduce fasting blood glucose, alleviate insulin resistance and improve dyslipidemia in diabetic mice. In addition, Artemisia selengensis cellulose can down-regulate the relative abundance of Firmicutes, Rikenellaceae and Lachnospiraceae, up-regulate the relative abundance of Bacteroidotas and decrease the F/B value. Artemisia selengensis cellulose can effectively improve the blood glucose and lipid levels of diabetic mice, and the mechanism may be achieved by regulating the composition of intestinal flora.

KEYWORDS

Artemisia selengensis; Soluble dietary fiber; Diabetes mellitus; Blood Sugar; Blood lipid; Intestinal flora

1. INTRODUCTION

Uncontrolled blood glucose levels are a major feature of Type 2 diabetes (T2D), especially post-meal elevated blood glucose, which is caused by low responsiveness to insulin, i.e., insulin resistance [1]. Patients with diabetes tend to have some complications, such as chronic kidney disease and cardiovascular disease, which are accompanied by a great economic burden [2, 3]. There are abundant microorganisms in the human gut. Under normal circumstances, the combined action of these microorganisms keeps the intestinal microecology in a balanced state, and the imbalance of the intestinal microecology will lead to the occurrence of some metabolic diseases, including T2D [4]. At present, healthy eating habits and scientific and reasonable recuperation have become one of the important ways to control blood sugar level. Among the most studied dietary patterns, people are increasingly interested in exploring the therapeutic effect of dietary fiber [5]. Dietary fiber is a kind of carbohydrate in plant foods, such as whole grains, vegetables, fruits and legumes. Adding dietary fiber to foods can not only enhance the nutritional value of foods, but also improve obesity, cardiovascular diseases and intestinal diseases [6]. Dietary fiber is an essential nutrient source for specific bacteria and helps maintain the function, richness, and stability of the gut microbiome. These complex carbohydrates help prevent obesity and inhibit the development of T2D by producing bacterial metabolites such as short-chain fatty acids [7]. Artemisia selengensis is an emerging source of soluble dietary fiber. In this study, by establishing a mouse diabetes model, we verified the efficacy of Artemisia selengensis cellulose in improving blood glucose and lipids, and explored the possible mechanism of Artemisia selengensis cellulose in improving the disorder of glucose and lipid metabolism in mice from the aspects of intestinal flora composition and SCFAs, in order to provide ideas for the development of related functional foods.

2. MATERIALS AND METHODS

2.1. Animal Experimen

Forty SPF grade 6-week-old male C57BL/6J mice (20±2) g were purchased from Henan Skobes Biotechnology Co., LTD. After 7 days of adaptive feeding, 6 mice were randomly selected as the control group (Con) and fed with basic diet, and the remaining mice were used to establish the model and fed with high-fat diet for 8 weeks. After 8 weeks, all mice were fasted for 12 hours, and STZ solution was dissolved in citrate buffer solution under the condition of ice bath, and STZ solution was injected intraperitoneally through 0.22μm Millipore filter membrane. The dose of STZ solution was 50mg/kgBW, and the injection volume was 0.1mL/10gBW. The mice were injected intraperitoneally for 5 days. One week later, the tail tip blood was collected to measure the fasting blood glucose of the mice, and the fasting blood glucose concentration was not less than 11.1mmol/L, which was regarded as successful modeling. Mice in the model group were randomly divided into two groups: model control group (MC) and diet fiber group (DF).

2.2. Fasting blood glucose test

The mice were fasted for 12 hours after the last feeding, and fasting blood glucose (FBG) was measured. About 1 mm of the tail tip was cut off with clean scissors, the first drop of blood was discarded, and the second drop of blood was placed on the blood glucose test paper to read the FBG value of the mice.

2.3. Measurement of Blood Lipid

After the intervention, all mice were fasted for 12 hours, and the blood of mice was removed by eyeball blood sampling in enzyme-free tubes. After standing at room temperature for 2 hours, the blood was centrifuged at 4 ºC and 5000 rpm for 10 minutes, and the upper serum was collected. The contents of TC, TG, LDL-C and HDL-C were detected by the kits, and the detection methods were carried out according to the instructions of each index kit.

2.4. Compositions of Intestinal Flora

After the intervention, each mouse was placed in a clean cage under sterile conditions, and about 200 mg fecal samples of each mouse were collected in a sterile centrifuge tube and stored at -80 ºC. Genomic DNA was extracted from fecal samples using kits from Zymo Research Corp (Irvine, CA, USA). The V3-V4 hypervariable regions of the bacterial 16S rRNA gene were amplified and sequenced on the Illumina NovaSeq platform. The DADA2 plugin in QIIME2 software was used to filter, denoise, merge, and remove chimeric sequences from all raw sample sequences to define operational taxonomic units (OTUs). Species composition, OTU variance between cases, alpha diversity, and beta diversity were assessed for each sample, based on the OTUs’ absolute abundance and annotations.

2.5. Data Statistics and Analysis

Data were analyzed and plotted by R software. The experimental data were expressed as mean ± standard deviation, and ordinary one-way analysis of variance was used between groups. $P<0.05$ indicates a significant difference, $P<0.01$ indicates a very significant difference, $P<0.001$ was considered to be highly significant.

3. RESULTS

3.1. Effect of Artemisia Selengensis Cellulose on Fasting Blood Glucose in Diabetic Mice

The results showed that chenopodium cellulose could effectively inhibit FBG levels in diabetic mice. As shown in Fig. 1, at the end of the intervention, the FBG values of the MC group were significantly different from those of the Con group ($P<0.001$). The FBG values of the DF group were significantly lower than those of the MC group ($P<0.001$), a significant reduction of 41.6%.

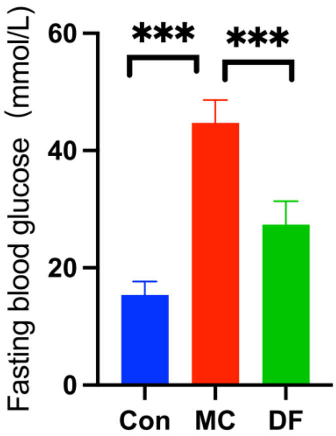

Figure 1. Effect of Artemisia selengensis cellulose on glucose tolerance in diabetic mice

3.2. Effects of Artemisia Selengensis Cellulose on Lipid Levels in Diabetic Mice

At the end of the dietary intervention, the eyeball blood was collected from the mice, and the four indicators of blood lipids in the serum of the mice in each group were measured, which were HDL-C, LDL-C, TG and TC. The results are shown in Fig. 2. Compared with Con group, the content of HDL-C in MC group was significantly decreased. Compared with the MC group, the content of HDL-C in the DF intervention group increased by 37.1%.

Compared with the Con group, the levels of LDL-C, TG and TC in the MC group were significantly increased. After dietary intervention, compared with MC group, the contents of LDL-C in DF group decreased by 27.3%, the contents of TG in DF group decreased by 42.0%, and the contents of TC in DF group decreased by 36.6%. These results indicate that Artemisia selengensis cellulose intervention can improve the abnormal blood lipid levels in diabetic mice.

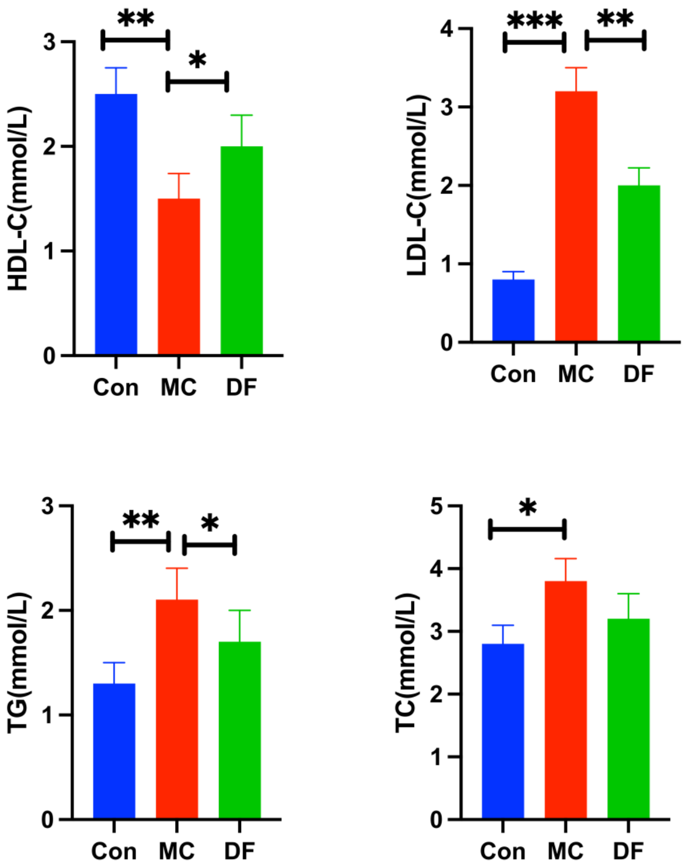

Figure 2. Effect of Artemisia selengensis cellulose on blood lipids in diabetic mice

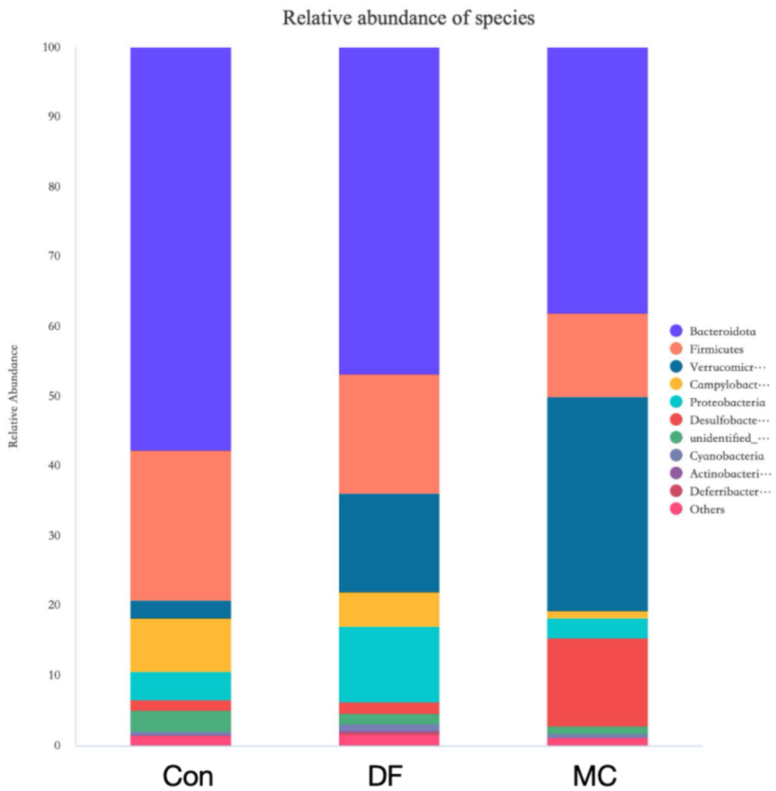

Figure 3. Effect of Artemisia selengensis cellulose on gut microbiota in diabetic mice

3.3. Effects of Dietary Fiber on Intestinal Flora

As shown in Fig.3, the relative abundance at the phylum level of mouse fecal microbiota showed that Bacteroidota and Firmicutes were the dominant phylum in each group. Compared with the Con group, the relative abundance of Firmicutes was significantly changed, and the relative abundance of Bacteroidotas was decreased in the MC group. The top 10 bacteria in relative abundance of species at the family level, among which Muribaculaceae, Bacteroidaceae, Rikenellaceae and Lachnospiraceae are the bacteria with relatively high relative abundance. Compared with the Con group, the relative abundance of Muribaculaceae in the MC group decreased by 22.24%, and after dietary intervention, the relative abundance of Muribaculaceae in the DF group increased by 6.04%. Compared with the Con group, the relative abundance of Rikenellaceae in the MC group increased by 10.72%, and after dietary intervention, the relative abundance of Rikenellaceae in the DF group decreased by 10.41%. Compared with the Con group, the relative abundance of Lachnospiraceae in the MC group increased by 6.63%, and after dietary intervention, the relative abundance of Lachnospiraceae in the DF group decreased by 6.74%. These results suggest that, for normal mice, the intestinal flora of diabetic mice is disordered, and the intervention of artemisia quinoa cellulose can promote the intestinal flora of mice to be close to normal mice to a certain extent. In conclusion, Artemisia selengensis cellulose may regulate the gut microbiota of diabetic mice by increasing the abundance of Bacteroidota and reducing the F/B value.

4. SUMMARY

This study found that Artemisia selengensis cellulose could effectively regulate blood glucose level, reduce serum insulin level, reduce insulin resistance and improve dyslipidemia in diabetic mice. Promote the secretion of acetic acid, propionic acid, butyric acid, valeric acid, isobutyric acid, isovaleric acid; At the phylum level, the treatment increased the relative abundance of Bacteroidota, decreased the relative abundance of Firmicutes, and decreased the F/B value. At the family level, the relative abundance of Muribaculaceae was up-regulated, and the relative abundance of Rikenellaceae and Lachnospiraceae was down-regulated, and tended to be normal.

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