Insoluble Dietary Fiber from Pear Pomace Can Prevent High-Fat Diet-Induced Obesity in Rats Mainly by Improving the Structure of the Gut Microbiota

Shimin Chang, Xingtian Cui, Mingzhang Guo, Yiling Tian, Wentao Xu, Kunlun Huang*, and Yuxing Zhang*

1College of Horticulture, Agricultural University of Hebei, Baoding 071001, P.R. China
2College of Agriculture, Hebei University of Engineering, Handan 056038, P.R. China
3Beijing Advanced Innovation Center for Food Nutrition and Human Health, College of Food Science & Nutritional Engineering, China Agricultural University, Beijing 100083, P.R. China
4College of Food Science and Technology, Agricultural University of Hebei, Baoding 071001, P.R. China

Supplement of dietary fibers (DF) is regarded as one of the most effective way to prevent and relieve chronic diseases caused by long-term intake of a high-fat diet in the current society. The health benefits of soluble dietary fibers (SDF) have been widely researched and applied, whereas the insoluble dietary fibers (IDF), which represent a higher proportion in plant food, were mistakenly thought to have effects only in fecal bulking. In this article, we proved the anti-obesity and glucose homeostasis improvement effects of IDF from pear pomace at first, and then the mechanisms responsible for these effects were analyzed. The preliminary study by real-time PCR and ELISA showed that this kind of IDF caused more changes in the gut microbiota compared with in satiety hormone or in hepatic metabolism. Further analysis of the gut microbiota by high-throughput amplicon sequencing showed IDF from pear pomace obviously improved the structure of the gut microbiota. Specifically, it promoted the growth of Bacteroidetes and inhibited the growth of Firmicutes. These results are coincident with previous hypothesis that the ratio of Bacteroidetes/Firmicutes is negatively related with obesity. In conclusion, our results demonstrated IDF from pear pomace could prevent high-fat diet-induced obesity in rats mainly by improving the structure of the gut microbiota.

Keywords: Insoluble dietary fiber, anti-obesity, gut microbiota, amplicon sequencing

Introduction

The long-term intake of a high-fat diet seriously affects human health and results in metabolic disorders and chronic diseases such as overweight, fatty liver, cardiovascular disease, hypertension, hypercholesteremia, insulin resistance, and leptin resistance [1]. The prevalence of these diseases is increasing in the current society and the pathogenesis is interrelated in a complex manner [2]. Supplement of dietary fiber (DF) in the diet is believed to be one of the most effective approaches to prevent the chronic diseases caused by over-intake of fat. Studies have proved that DFs can reduce the risk of hyperlipidemia, hypercholesterolemia, hyperglycemia, and colon cancer by modulating food ingestion, digestion, absorption, and metabolism [3]. The American Dietetic Association suggested that the daily diet of the public should contain more than 25 g DFs for adult women and 38 g DFs for adult men to reduce the risk of cardiovascular disease [4]. The health benefits of DF has been widely accepted and applied.

DF is generally classified into soluble dietary fiber (SDF) and insoluble dietary fiber (IDF) according to their solubility in water. Previously, it was considered that SDF contributed to the reduction of serum lipids and cholesterol...
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level, whereas the effect of IDF was mainly fecal bulking [3]. Thus, previous studies have paid more attention to the effects and mechanisms of SDF. Suggestions regarding the mechanisms responsible for these effects of SDF include the regulation of the expression of key genes involved in hepatic metabolism (such as fatty acid synthase, acetyl-CoA carboxylase, cholesterol 7 a-hydroxylase, and AMP-activated protein kinase) [5–8], alterations in satiety hormone secretion (such as ghrelin, peptide YY, and leptin) [9, 10], delayed gastric emptying, energy dilution, increased energy expenditure, and regulation of the gut microbiota [9]. Regarding IDF, fewer studies have been conducted to assess the relationship between IDF and overweight. Isken et al. [11] reported that supplementing the diet with insoluble cereal fiber resulted in significantly lower weight gain and improved insulin sensitivity. Tan et al. [12] also found the associations of IDF intake and glycemic index value with subsequent HbA1c levels. In spite of these efforts, whether and how IDF affects the gut microbiota, key lipometabolism, and satiety hormone secretion is largely unknown.

For SDF, many studies indicated its supplement could alter the composition of the microbiota, thus preventing obesity [6, 9, 13]. Compared with SDF, IDF is less possible to be absorbed, thus it remains in the intestinal tract and has interaction with the gut microbiota for a longer time. The gut microbiota is highly influenced by the host diet and the fermentability of ingested fibers [14]. Recently, many studies reported that obesity was related to disorder of the gut microbial community. Generally, people who possess less Bacteroidetes and more Firmicutes in their gut microbiota are more likely to be obese [15]. With the development of high-throughput sequencing and metagenomics, increasing evidence has indicated the role of the gut microbiota in the etiology of host chronic metabolic diseases such as type II diabetes [16] and symptomatic atherosclerosis [17]. As IDF could hardly be absorbed, the gut microbiota could be a most possible intermediary between IDF and human health.

Pear pomace from fruits is a good source of IDF [18–20], and thus the reasonable use of pomace could be of great benefit to human health and the protection of the environment. Pears are popular fruits worldwide, particularly in juices and soft drinks. Thousands of tons of pear seeds, pulp, and peels are generated as agricultural byproducts and are typically discarded and wasted. Natural IDF extracted from pear pomace was added to high- and normal-fat diets in the present study to evaluate the functional effects of the extracted IDF on high-fat diet-challenged rats. The purpose of the present study was to evaluate the functional effects of the IDF extracted from pear pomace on rats fed a high-fat diet. The gut microbiota, key lipometabolism gene expression, and satiety hormone secretion in these rats were determined by real-time PCR and ELISA. After finding the potential effect of IDF on the gut microbiota, the change in pattern of the gut microbiota was further analyzed by high-throughput amplicon sequencing. Our results suggested IDF from pear pomace could prevent high-fat diet-induced obesity in rats mainly by improving the structure of the gut microbiota.

Materials and Methods

Animal Model

Eight-week-old, specific pathogen-free male Sprague-Dawley rats were purchased from Vital River Laboratories (China; license number SCXK (Beijing) 2012-0001). These rats were housed at a constant room temperature (22 ± 2°C) and humidity (55% ± 10%) and maintained on a 12-h light/dark cycle in an experimental animal room in the Supervision, Inspection and Testing Center for Genetically Modified Organisms of the Ministry of Agriculture (Beijing, China; license number SYXK (Beijing) 2010-0036). Food and water were provided ad libitum throughout the test. All animal experiments were approved by the Animal Experimental Welfare and Ethical Inspection Committee (No. 100034) of the Supervision, Inspection and Testing Center for Genetically Modified Organisms of the Ministry of Agriculture (Beijing, China).

Pear Pomace Samples and Diets

The alkali-catalyzed hydrolysis method was applied to extract the IDF from the pear pomace. The pear pomace was prepared by removing the juice from the fruit and grinding the fruit to a granularity of 60 mesh using a high-speed disintegrator followed by the addition of ethyl acetate. After 3 h, the pear pomace was washed with water and dried with hot air at 55°C overnight. Sodium hydroxide was added at 20 times the volume of the pomace, and the mixture was then centrifuged at 4,000 rpm for 15 min. The collected matter was then deposited and washed with water. The IDF was recovered from the residue after the deposit was washed with 76% ethanol, 95% ethanol, and acetone at 4 times the volume of the pomace and dried with hot air at 55°C overnight. The IDF content in the final pear pomace extract was approximately 40%, and the other 60% of the extract was nitrogen-free extract, which is also a common composition in normal rat chow, providing energy only. The content of IDF was determined according to GB 5009.88-2014.

Before the test, the animals were acclimatized to the environment for 7 days. The rats were then randomly divided into four groups (8 rats per group) according to average body weight, and the groups were fed with a normal chow diet (NCD), a high dietary fiber diet (HD) containing 8% fiber supplement in total, a high-fat