Effects of Short-term Individual and Mixed Dietary Supplementation of High-Level Oat -Glucan and Microcrystalline Cellulose on Growth Performance, Organ Indices, and Fecal Bacterial Community Structure in Mice (Postprint)

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Abstract

This study aimed to investigate the effects of short-term individual and combined supplementation of high-level oat -glucan and microcrystalline cellulose (MCC) in diets on growth performance, organ indices, and fecal bacterial community structure in mice. Thirty-six healthy BALB/c mice with a body weight of (17.95±0.95) g were selected and randomly divided into 4 groups according to body weight: control group (CON group), diet without oat -glucan and microcrystalline cellulose; glucan group (G group), diet containing 28% oat -glucan; MCC group (M group), diet containing 20% MCC; mixed group (GM group), diet containing 14% oat -glucan and 10% MCC. The experimental period was 21 days. The results showed: 1) The average daily gain (ADG) of mice in each group during the whole period (days 1–21) was not significantly different (P>0.05), while the average daily feed intake (ADFI) during the whole period was significantly different (P<0.05), showing that the G group 0.05), the epididymal fat pad index of mice in each fiber-added group (G group, M group, GM group) was not significantly different from that of the CON group (P>0.05), but the epididymal fat pad index of mice in both G group and M group was significantly lower than that of the GM group (P<0.05). 3) On days 4 and 7 of the experiment, the Shannon-Wiener index of fecal bacteria in G group mice was significantly lower than that in the CON group (P<0.05). PCR-denaturing gradient gel electrophoresis (DGGE) profile cluster analysis showed that on days 13 and 17 of the experiment, the fecal bacterial community structure of mice in each group was significantly different, with samples from each group clustering separately on the phylogenetic tree. In conclusion, short-term individual or combined supplementation of high-level oat -glucan and MCC in mouse diets

can reduce ADFI in mice, but does not affect ADG and spleen index in mice; combined supplementation of oat -glucan and MCC promotes epididymal fat deposition more than individual supplementation; high-level oat -glucan can reduce the diversity of fecal bacteria in mice; supplementation of both oat -glucan and MCC can alter the fecal microbiota in mice, suggesting that there may be a core bacterial community in the hindgut of mice that specifically utilizes these two types of fiber.

Full Text

Short-Term Adding High-Level Oat -Glucan, Microcrystalline Cellulose and Their Mixture in Diets Affects Growth Performance, Organ Indexes and Fecal Bacterial Community Structure in Hindgut of Mice

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Abstract: This experiment was conducted to investigate the effects of adding high-level oat -glucan, microcrystalline cellulose (MCC) as well as their mixture in diets on the growth performance, organ indexes and fecal bacterial community structure of mice over a short-term period. Thirty-six healthy BALB/c mice with body weight of (17.95±0.95) g were selected and randomly allocated to four groups according to body weight: control group (CON group) fed a diet without oat -glucan and microcrystalline cellulose; glucan group (G group) fed a diet containing 28% oat -glucan; MCC group (M group) fed a diet containing 20% MCC; and mixture group (GM group) fed a diet containing 14% oat -glucan and 10% MCC. The experiment lasted for 21 days.

The results showed as follows: 1) During the whole experimental period (days 1–21), the average daily gain (ADG) of mice showed no significant difference among groups ($P>0.05$), while the average daily feed intake (ADFI) differed significantly ($P<0.05$), following the trend G group < M group < GM group < CON group. 2) The spleen index of mice showed no significant difference among groups ($P>0.05$), and the epididymal fat pad index of mice in the three fiber-supplemented groups (G, M and GM groups) showed no significant difference compared with CON group ($P>0.05$), but the epididymal fat pad index in both G and M groups was significantly lower than that in GM group ($P<0.05$). 3) On experimental days 4 and 7, the bacterial Shannon-Wiener index of feces in G group was significantly lower than that in CON group ($P<0.05$). Cluster analysis of PCR-denaturing gradient gel electrophoresis (DGGE) profiles showed that the fecal bacterial community structure differed markedly among groups on days 13 and 17, with samples from each group clustering separately on the phyloge-

netic tree. In conclusion, short-term supplementation of high-level oat -glucan, MCC or their mixture in diets can reduce ADFI in mice without affecting ADG and spleen index. The mixture of oat -glucan and MCC promotes epididymal fat deposition more effectively than single-fiber supplementation. High-level oat -glucan reduces fecal bacterial diversity in mice. Both oat -glucan and MCC alter the fecal microflora of mice, suggesting that specific core bacterial groups may exist in the hindgut that selectively utilize these two types of dietary fiber.

Keywords: oat -glucan; microcrystalline cellulose; BALB/c mice; growth performance; organ indexes; fecal microflora

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Dietary fiber (DF) refers to carbohydrates with ten or more monomeric units that cannot be hydrolyzed by endogenous enzymes in the small intestine of humans or animals. Based on water solubility, DF can be classified into soluble dietary fiber (SDF) and insoluble dietary fiber (IDF) [**?**]. Epidemiological studies have found that low DF intake is associated with obesity, colorectal cancer, diabetes and certain cardiovascular diseases [**?**]. The physiological functions of DF are related to its physicochemical properties, where its bulking characteristics and particle size significantly affect colonic function in monogastric animals. Moreover, the fermentation pattern of DF in the host hindgut directly influences the types and proportions of short-chain fatty acids (SCFAs), thereby affecting energy metabolism and intestinal immune function [**?**].

The gut of monogastric animals, particularly the colon, harbors a vast and complex microbial community influenced by genetic background, sex, age, immune system, intestinal environment (pH, etc.) and diet [**?**]. DF serves as one of the primary utilizable substrates for colonic bacteria [**?**]. Numerous studies have demonstrated that DF can increase the abundance of certain probiotics (e.g., *Bifidobacterium* and *Lactobacillus*) in the animal gut [**?**], though the effects of different DF types on hindgut microbiota structure in monogastric animals remain unclear.

Therefore, this study utilized BALB/c mice as a model to investigate the short-term effects of supplementing high-level typical SDF (oat -glucan) or typical IDF [microcrystalline cellulose (MCC)] alone or in combination on hindgut bacterial community structure using molecular fingerprinting techniques. Additionally, we examined whether different DF types differentially affected growth performance and organ indexes, laying a foundation for further research on the mechanisms through which these two DF types influence host energy metabolism and intestinal health.

1.1 Test Materials

Oat -glucan was purchased from Shaanxi Ciyuan Biotechnology Co., Ltd., extracted from oats as a light yellow powder with 70% purity; the remaining 30% consisted mainly of oat bran and protein. MCC was purchased from Qufu Tianli Pharmaceutical Excipients Co., Ltd., as a white powder with purity 99%. Male BALB/c mice were purchased from Chengdu Dossy Experimental Animals Co., Ltd.

1.2 Experimental Design

Thirty-six healthy male BALB/c mice at 6 weeks of age with consistent body condition (17.95±0.95) g were randomly divided into four groups according to body weight: control group (CON group) fed a diet without additional fiber; glucan group (G group) fed a diet supplemented with 28% -glucan; MCC group (M group) fed a diet supplemented with 20% MCC; and mixture group (GM group) fed a diet supplemented with 14% oat -glucan + 10% MCC, with nine mice per group. Using casein, corn starch, sucrose, soybean oil and palm oil as basal ingredients, experimental diets were formulated according to AIN93 standards following the principle of equal energy and equal nitrogen (Table 1). The exogenous fiber content in the fiber-supplemented groups (G, M and GM groups) was 19.6%–19.8%. In addition to considering protein and carbohydrate levels across groups, we also accounted for total energy balance. Due to the large fiber dosage (~20%), bentonite clay was added to the control group to reduce its total energy and make the gross energy as similar as possible across the four groups. Furthermore, the measured energy-to-protein ratios showed no significant differences among the four groups (P>0.05). Mice were housed individually with ad libitum access to feed and water, and the experimental period lasted 21 days.

1.3.1 Body Weight and Feed Intake Measurement

Mice were weighed in the morning on an empty stomach on days 1, 4, 7, 10, 13, 17 and 21 of the experiment to calculate average daily gain (ADG) for each stage and the whole period. Daily feed intake of each mouse was recorded to calculate average daily feed intake (ADFI) for each stage and the entire period.

1.3.2 Organ Index Measurement

At the end of the experiment (day 21), mice were weighed and then euthanized to collect the spleen and epididymal fat pad, which were weighed immediately. Organ indexes were calculated using the following formula: Organ index (mg/g) = organ weight (mg) / live body weight (g).

1.3.3 Fecal Bacterial Diversity Measurement

Fresh fecal samples were collected aseptically from mice on days 4, 7, 10, 13, 17 and 21. Fecal metagenomic DNA was extracted using the QIAamp DNA Stool Mini Kit (Germany) following the manufacturer' s instructions. For each group, fecal DNA samples from three randomly selected mice were used as templates to amplify the V6–V8 variable region of bacterial 16S rDNA using primers 968f-GC [**?**] and 1401r [**?**]. PCR amplification conditions were: 94°C for 5 min, followed by 35 cycles of 94°C for 30 s, 56°C for 20 s, and 68°C for 40 s, with a final extension at 68°C for 7 min. PCR products were identified by 1.0% agarose gel electrophoresis. Denaturing gradient gel electrophoresis (DGGE) was performed using the Bio-Rad Dcode system with a denaturant concentration gradient of 45%–60%. Electrophoresis was conducted at 80 V and 60°C for 12 h in 1×TAE buffer, followed by silver nitrate staining and imaging using the UVP gel imaging system.

1.4 Statistical Analysis

The Quantity One 4.6.2 software was used for band counting and analysis of PCR-DGGE profiles, and the Shannon-Wiener index was calculated. Data on growth performance and organ indexes were analyzed by one-way ANOVA using SPSS 21.0 software, with Duncan' s multiple comparison test used for intergroup comparisons. Data are expressed as means ± standard deviation. $P<0.05$ was considered statistically significant, and $P<0.01$ was considered highly significant.

2.1 Changes in Body Weight, Weight Gain and Feed Intake of Mice

Body weight of mice in all groups increased over the feeding period (Table 2). At the end of the experiment, body weight increased by 16.77%, 17.46%, 17.41% and 16.74% in G, M, GM and CON groups, respectively, with no significant differences among groups ($P>0.05$). On day 7, body weight in G group was significantly lower than in other groups ($P<0.05$).

ADG of mice in stages 3 (days 7–9) and 4 (days 10–12) was significantly affected by group ($P<0.05$) (Table 3). In stage 3, ADG showed no significant difference between fiber-supplemented groups and CON group ($P>0.05$), but ADG in G group was significantly higher than in M and GM groups ($P<0.05$). In stage 4, ADG in G group was significantly higher than in M and CON groups ($P<0.05$) but showed no significant difference from GM group ($P>0.05$).

Except for stage 4, ADFI in all stages was significantly or highly significantly affected by group ($P<0.05$ or $P<0.01$) (Table 4), with the overall trend being CON group highest, G group lowest, and M group slightly higher than GM group.

2.2 Differences in Organ Indexes of Mice

As shown in Table 5 , spleen index and epididymal fat pad index showed no significant difference between CON group and fiber-supplemented groups (P>0.05). However, epididymal fat pad index in GM group was significantly higher than in G and M groups (P<0.05).

2.3 Differences in Hindgut Bacterial Community Structure of Mice

Bacterial diversity in feces differed among groups at various time points (Table 6). On day 4, the Shannon-Wiener index in G group was significantly lower than in CON group (P<0.05). On day 7, the Shannon-Wiener index in G group was significantly lower than in all other groups (P<0.05). On day 10, the Shannon-Wiener index in fiber-supplemented groups showed no significant difference from CON group (P>0.05), but the index in M group was significantly higher than in G and GM groups (P<0.05).

Cluster analysis of PCR-DGGE profiles revealed that samples from all groups clustered randomly at the start of the experiment (Figure 1 [Figure 1: see original paper]-a). On days 7, 10 and 13, samples from the same group clustered within the same clade (Figure 1-b, Figure 1-c and Figure 1-d). On day 7, CON, G and M groups each had two samples clustering together with similarities of 71%, 52% and 60%, respectively, while GM group had three samples clustering together with 60% similarity. On day 10, G and M groups each had three samples clustering together with similarities of 63% and 72%, respectively, while CON and GM groups each had two samples clustering together with similarities of 71% and 61%, respectively. On day 13, each group had two samples clustering together with similarities of 70% (G group), 75% (M group), 74% (GM group) and 78% (CON group). On day 17, overall clustering was random, though G and GM groups each still had two samples clustering together with similarities of 63% and 68%, respectively (Figure 1-e). On day 21, only G group had two samples clustering together with 57% similarity, while other groups showed random clustering (Figure 1-f).

The cluster analysis was generated using the UPMGA method according to the PCR-DGGE profile, and the phylogenetic trees of six sampling time points were shown in figure a to figure f. Symbols d4, d7, d10, d13, d17 and d21 represent fecal samples obtained at days 4, 7, 10, 13, 17 and 21, respectively. CON indicates control group without extra fiber supplementation; G indicates oat-derived -glucan supplemented group; M indicates MCC supplemented group; GM indicates the mixture of oat-derived -glucan and MCC supplemented group.

3.1 Effects of Different Dietary Fiber Types on Body Weight and Feed Intake of Mice

Current reports on the effects of SDF or IDF on body weight in rats and mice have yielded inconsistent results. Studies have shown that feeding mice -glucan of different molecular weights for six weeks significantly reduced body weight [**?**]. Dongowski et al. [**?**] reported that adding barley rich in -glucan to rat diets increased weight gain. Conversely, other research found that adding 10% cellulose to rat diets had no significant effect on body weight [**?**]. Our results indicate that short-term supplementation of high-level oat -glucan and MCC alone or in combination had no significant effect on final body weight or overall ADG in mice.

Compared with CON group, ADFI was lower in all fiber-supplemented groups, possibly due to reduced diet palatability caused by high-level dietary fiber. ADFI in mice fed diets containing oat -glucan was significantly lower than in other groups, consistent with previous findings [**?**]. This may be attributed to two mechanisms: first, high-viscosity -glucan slows gastric emptying rate and intestinal transit [**?**]; second, -glucan can be fermented by microorganisms in the hindgut to produce SCFAs [**?**]. Both factors can stimulate secretion of anorexic hormones such as peptide YY (PYY) and glucagon-like peptide-1 (GLP-1), inducing satiety [**?**] and suppressing appetite to reduce feed intake.

Throughout the experimental period, ADFI in G group remained consistently low, and body weight on day 7 was significantly lower than in other groups, though final body weight showed no significant difference. Similarly, Isken et al. [**?**] found that feeding high-fat diet-induced obese mice diets containing 10% SDF (guar gum) and IDF (cereal fiber) for 45 weeks resulted in significantly higher body weight in the SDF group, significantly lower fecal energy, and significantly higher colonic SCFA content. Interestingly, ADG in G group was the highest during both stage 3 and stage 4. SCFAs produced by hindgut microbial fermentation of fiber in monogastric animals are considered an important energy source for the host [**?**]. We speculate that oat -glucan was fermented in the mouse hindgut to produce abundant SCFAs, which were absorbed by intestinal epithelium and may have served as an energy supplement. This could explain why G group mice, despite consistently low feed intake, showed faster weight gain and ultimately achieved similar body weight to other groups, though the specific mechanism requires further verification.

Under our experimental conditions, ADFI in both M and GM groups was also significantly lower than in CON group, yet final body weight showed no significant difference. This may be because a small portion of IDF can be fermented by hindgut bacteria to produce SCFAs, indirectly providing energy. Additionally, IDF can increase digesta transit rate, and this "sweeping"mechanism may reduce adhesion of harmful bacteria, benefit intestinal health, and facilitate nutrient absorption, though the specific mechanism warrants further investigation.

3.2 Effects of Different Dietary Fiber Types on Organ Indexes of Mice

DF has immune-promoting effects on animals [?]. The spleen, as a secondary lymphoid organ, is closely related to humoral and cellular immunity, and spleen index can serve as a preliminary indicator of immune status [?]. However, no significant differences in spleen index were observed among groups in this study, possibly because DF primarily affects the immune system in the intestinal tract [?]. Therefore, future research should focus on exploring DF' s effects on the intestinal immune system and its underlying mechanisms.

Reports indicate that oat SDF can inhibit body fat deposition [?], and adding 7.5%–30.0% oat bran to high-fat diets significantly reduces epididymal fat content in rats [?]. Interestingly, our study found that while single or combined supplementation of high-level oat -glucan and MCC had no significant effect on epididymal fat pad index, the mixture of both fiber types resulted in significantly higher epididymal fat pad index compared with single-fiber groups. This suggests that oat -glucan and MCC may have complex interactive effects on fat deposition in mice, though the specific mechanism requires further investigation.

3.3 Effects of Different Dietary Fiber Types on Fecal Bacterial Community Structure of Mice

Studies have shown that gut microbiota is extremely sensitive to dietary changes, with bacterial community structure responding within a short period [?]. Zhou Mengyi [?] used PCR-DGGE to compare cecal bacterial community structure in mice fed diets containing cellulose or curdlan for 28 days, finding that the two groups clustered into distinct clades, demonstrating clear diet specificity, which is consistent with our results.

Our study found that fecal bacterial diversity remained consistently low throughout the experimental period in mice fed diets containing high-level oat -glucan, which contradicts findings in rats by Snart et al. [?]. This discrepancy may be due to the higher -glucan supplementation level (20%) used in our study, suggesting that the effect of dietary fiber on hindgut bacterial diversity in monogastric animals depends not only on fiber type but also on supplementation level. However, PCR-DGGE technology typically detects only bacterial groups representing 1% of the total bacterial population [?]. Although no significant differences in fecal bacterial diversity were observed among groups in the later experimental stages, cluster analysis results implied the existence of core bacterial groups in the mouse hindgut that specifically utilize SDF or IDF, warranting further investigation.

In conclusion: Under our experimental conditions, short-term single or combined supplementation of oat -glucan and MCC reduced ADFI in mice without significantly affecting ADG. Single supplementation of oat -glucan or microcrystalline cellulose had no significant effect on spleen index or epididymal fat

pad index, but combined feeding of both fibers promoted fat deposition in the epididymal fat pad. Different dietary fiber types altered fecal microbiota structure in mice, and the reasons for differential effects of oat -glucan and MCC on fecal microbiota structure may differ. Our results suggest that specific bacterial groups capable of degrading SDF or IDF exist in the mouse hindgut.

References

[1] Codex Alimentarius Commission. Report of the 30th session of the codex committee on nutrition and foods for special dietary uses[R]. Cape Town: Codex Alimentarius Commission, 2009.

[2] GUTKOSKI L C, DE ALMEIDA BONAMIGO J M, DE FREITAS TEIXEIRA D M, et al. Development of oat based cereal bars with high dietary fiber content[J]. Food Science and Technology (Campinas), 2007, 27(2): 355–363.

[3] SIERRA M, GARCÍA J J, FERNÁNDEZ N, et al. Therapeutic effects of psyllium in type 2 diabetic patients[J]. European Journal of Clinical Nutrition, 2002, 56(9): 830–842.

[4] LATTIMER J M, HAUB M D. Effects of dietary fiber and its components on metabolic health[J]. Nutrients, 2010, 2(12): 1266–1289.

[5] BRESTOFF J R, ARTIS D. Commensal bacteria at the interface of host metabolism and the immune system[J]. Nature Immunology, 2013, 14(7): 676–684.

[6] MASLOWSKI K M, MACKAY C R. Diet, gut microbiota and immune responses[J]. Nature Immunology, 2011, 12(1): 5–9.

[7] SCOTT K P, GRATZ S W, SHERIDAN P O, et al. The influence of diet on the gut microbiota[J]. Pharmacological Research, 2013, 69(1): 52–60.

[8] HAMAKER B R, TUNCIL Y E. A perspective on the complexity of dietary fiber structures and their potential effect on gut microbiota[J]. Journal of Molecular Biology, 2014, 426(23): 3838–3850.

[9] LEE Y K, SALMINEN S. Handbook of probiotics and prebiotics[M]. 2nd ed. New York: John Wiley & Sons, 2009.

[10] LANE D J. 16S/23S rRNA sequencing[J]. Nucleic Acid Techniques in Bacterial Systematics, 1991: 125–175.

[11] NÜBEL U, ENGELEN B, FELSKE A, et al. Sequence heterogeneities of genes encoding 16S rRNAs in *Paenibacillus polymyxa* detected by temperature gradient electrophoresis[J]. Journal of Bacteriology, 1996, 178(19): 5636–5643.

[12] BAE I Y, LEE S, KIM S M, et al. Effect of partially hydrolyzed oat -glucan on the weight gain and lipid profile of mice[J]. Food Hydrocolloids, 2009, 23(7): 2016–2021.

[13] DONGOWSKI G, HUTH M, GEBHARDT E, et al. Dietary fiber-rich barley products beneficially affect the intestinal tract of rats[J]. The Journal of Nutrition, 2002, 132(12): 3704–3714.

[14] LIN Liping, LI Juesheng, WU Huiyun, et al. Effects of different components of dietary fiber on lipid metabolism in rats[J]. Acta Nutrimenta Sinica, 1993, 15(2): 137–141.

[15] ZHANG Peipei, FAN Mingtao, HU Xinzhong, et al. Effects of oat whole flour and oat -glucan on growth and blood biochemical indices in rats[J]. Journal of the Chinese Cereals and Oils Association, 2010, 25(9): 27–31.

[16] EL KHOURY D, CUDA C, LUHOVYY B L, et al. Beta glucan: health benefits in obesity and metabolic syndrome[J]. Journal of Nutrition and Metabolism, 2012, 2012: 851362.

[17] SCHROEDER N, MARQUART L F, GALLAHER D D. The role of viscosity and fermentability of dietary fibers on satiety- and adiposity-related hormones in rats[J]. Nutrients, 2013, 5(6): 2093–2113.

[18] LIN H V, FRASSETTO A, KOWALIK E J, Jr, et al. Butyrate and propionate protect against diet-induced obesity and regulate gut hormones via free fatty acid receptor 3-independent mechanisms[J]. PLoS One, 2012, 7(4): e35240.

[19] OVERDUIN J, SCHOTERMAN M H C, CALAME W, et al. Dietary galacto-oligosaccharides and calcium: effects on energy intake, fat-pad weight and satiety-related, gastrointestinal hormones in rats[J]. British Journal of Nutrition, 2013, 109(7): 1338–1348.

[20] ZHOU J, MARTIN R J, TULLEY R T, et al. Dietary resistant starch upregulates total GLP-1 and PYY in a sustained day-long manner through fermentation in rodents[J]. American Journal of Physiology-Endocrinology and Metabolism, 2008, 295(5): E1160–E1166.

[21] ADAM C L, WILLIAMS P A, DALBY M J, et al. Different types of soluble fermentable dietary fibre decrease food intake, body weight gain and adiposity in young adult male rats[J]. Nutrition & Metabolism, 2014, 11(1): 36.

[22] ISKEN F, KLAUS S, OSTERHOFF M, et al. Effects of long-term soluble vs. insoluble dietary fiber intake on high-fat diet-induced obesity in C57BL/6J mice[J]. The Journal of Nutritional Biochemistry, 2010, 21(4): 278–284.

[23] SCHLEY P D, FIELD C J. The immune-enhancing effects of dietary fibres and prebiotics[J]. British Journal of Nutrition, 2002, 87(Suppl. 2): S221–S230.

[24] ANGUITA M, CANIBE N, PÉREZ J F, et al. Influence of the amount of dietary fiber on the available energy from hindgut fermentation in growing pigs: use of cannulated pigs and *in vitro* fermentation[J]. Journal of Animal Science, 2006, 84(10): 2766–2778.

[25] MCNEIL N I. The contribution of the large intestine to energy supplies in man[J]. The American Journal of Clinical Nutrition, 1984, 39(2): 338–342.

[26] WANG Dajun, WANG Qi, WANG Ningping, et al. Effects of total alkaloids of *Cynanchum chinense* on humoral immune function in mice[J]. Journal of Ningxia Medical University, 2009, 31(2): 161-162, 170.

[27] YAMADA K, TOKUNAGA Y, IKEDA A, et al. Effect of dietary fiber on the lipid metabolism and immune function of aged Sprague-Dawley rats[J]. Bioscience, Biotechnology, and Biochemistry, 2003, 67(2): 429-433.

[28] LIM B O, YAMADA K, NONAKA M, et al. Dietary fibers modulate indices of intestinal immune function in rats[J]. The Journal of Nutrition, 1997, 127(5): 663-667.

[29] SHEN Ruiling, CHEN Ming, DONG Jilin. Study on obesity prevention in high-fat fed mice by oat soluble dietary fiber[J]. Cereals & Oils, 2012, 25(2): 10-12.

[30] PENG C H, CHANG H C, YANG M Y, et al. Oat attenuate non-alcoholic fatty liver and obesity via inhibiting lipogenesis in high fat-fed rat[J]. Journal of Functional Foods, 2013, 5(1): 53-61.

[31] RAMIREZ-FARIAS C, SLEZAK K, FULLER Z, et al. Effect of inulin on the human gut microbiota: stimulation of *Bifidobacterium adolescentis* and *Faecalibacterium prausnitzii*[J]. British Journal of Nutrition, 2008, 101(4): 541-550.

[32] ZHOU Mengyi. Study on the regulatory effects of curdlan on intestinal physiology and pathology[D]. PhD thesis. Nanjing: Nanjing University of Science and Technology, 2014.

[33] SNART J, BIBILONI R, GRAYSON T, et al. Supplementation of the diet with high-viscosity beta-glucan results in enrichment for lactobacilli in the rat cecum[J]. Applied and Environmental Microbiology, 2006, 72(3): 1925-1931.

[34] BEN OMAR N, AMPE F. Microbial community dynamics during production of the Mexican fermented maize dough pozol[J]. Applied and Environmental Microbiology, 2000, 66(9): 3664-3673.

Note: Figure translations are in progress. See original paper for figures.