

## Original Article

# Effect of whole grain pancakes on postprandial cardiometabolic risk factors in healthy subjects: A randomized crossover trial

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**Background and Objectives:** Whole grains are rich in nutrients and have multiple health benefits, playing an important role in the diet. This study aims to investigate the acute effects of different whole grain pancakes on cardiometabolic risk factors in healthy subjects. **Methods and Study Design:** Twenty eligible healthy subjects (aged 19-24 years) were recruited, with baseline measurements including fasting blood glucose, insulin, C-reactive protein (CRP), triglycerides (TG), total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), and low-density lipoprotein cholesterol (LDL-C). Then, the subjects were randomly allocated into four groups: the steamed bun (SB) group (control group), maize pancake (MP) group, buckwheat pancake (BP) group, and oat pancake (OP) group, with a serving size of 100g. Blood samples were collected at 0h, 0.5h, 1h, 2h, and 3h, respectively. After a one-week washout period, the volunteers consumed another type of pancake in a crossover manner until each volunteer had tried all four foods. Serum cardiometabolic risk factors were analyzed using standard methods. **Results:** No significant differences in serum glucose, insulin, CRP, TG, and HDL-C concentrations were observed among the control group and three whole grain pancake groups. However, compared with the control group, the concentrations of serum TC and LDL-C in the oat pancake group were significantly reduced ( $p < 0.05$ ), which may be attributed to the presence of  $\beta$ -glucan and higher dietary fiber in oat pancakes. **Conclusions:** This study provides important dietary guidance for both pancake consumers and individuals with cardiometabolic risk factors.

**Key Words:** oats, buckwheat, whole grains, pancake, cholesterol

## INTRODUCTION

It has been suggested that whole grain and dietary fiber intake is associated with reduced risk of cardiovascular disease (CVD), obesity and gastrointestinal disorders, as well as improved lipid profiles, inflammation, blood glucose control and immune function.<sup>1</sup> The US FDA has approved a claim that diets high in whole grains, fruits, and vegetables, and low in saturated fat (<10% of total daily calories) and cholesterol, reduce the risk of coronary heart disease (CHD).<sup>2</sup> A meta-analysis by Brown et al. showed that soluble fiber intake decreases total cholesterol (TC) and low-density lipoprotein cholesterol (LDL-C) by -0.045 and -0.057 mmol/L per gram, respectively.<sup>3</sup> Soluble dietary fiber (SDF) exerts cholesterol-lowering effects influenced by fiber type, adaptation length, intake amount, and overall dietary confounders. Mechanistically, dietary fiber enhances bile acid excretion and cholesterol conversion to bile acids; colonic microbiota fermentation of SDF produces short-chain fatty acids (SCFAs), which may inhibit hepatic cholesterol synthesis.<sup>4,5</sup>

Oats confer cardiovascular benefits by improving lipid and glucose profiles, supporting their potential as an adjuvant for metabolic disorder prevention and treatment.<sup>6</sup> A double-blind randomized trial in hypercholesterolemic patients showed that 6-week oat consumption

significantly reduced serum TC and LDL-C, compared with dextrose.<sup>7</sup> For adults  $\geq 65$  years, daily intake of 5 g oat  $\beta$ -glucan lowers cholesterol and CVD risk,<sup>8</sup> while a 4-week double-blind study in healthy participants found 3 g high-molecular-weight oat  $\beta$ -glucan reduced CVD risk by 8%.<sup>9</sup> Both European Food Safety Authority (EFSA) and U.S. Food and Drug Administration (FDA) confirm that  $\geq 3$  g/day oat  $\beta$ -glucan lowers blood cholesterol and CHD risk. Mechanistically, oat  $\beta$ -glucan exerts lipid-lowering effects via regulating cholesterol/bile acid metabolism, reducing LDL-C, and producing SCFAs to modulate lipid metabolism.<sup>10-12</sup>

Buckwheat or buckwheat-enriched products offer notable health benefits. Systematic reviews and meta-analyses show buckwheat intake is associated with significantly lower fasting blood glucose, TC, and triglycerides (TG) (vs. baseline/control), but has no significant effect on

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Manuscript received 27 November 2025. Initial review completed 08 January 2025. Revision accepted 16 May 2025.

doi: 10.6133/apjcn.202604\_35(2).0001

LDL-C or high-density lipoprotein cholesterol (HDL-C).<sup>13</sup> Its cholesterol-lowering activity is mediated by enhancing fecal excretion of bile acids and neutral cholesterol, while buckwheat extract reduces intestinal cholesterol absorption (possibly via shortened intestinal transit time).<sup>14</sup> Additionally, its lipid-lowering effect is accompanied by reduced hepatic glucose-6-phosphate dehydrogenase and fatty acid dehydrogenase activities.<sup>15</sup>

Maize peptides, prepared by enzymatic hydrolysis of maize starch wet-processing by-products, are rich in leucine and glutamic acid, and can effectively improve high-sugar/high-fat induced glycolipid metabolism disorders. Additionally, multi-grain combinations exert more significant effects.<sup>16</sup> Liu Shufan et al. compared the hyperlipidemic metabolism-regulating effects of maize, buckwheat, and maize-buckwheat mixed powder, finding the mixed powder was more effective in reducing TC, TG, LDL-C, enhancing hepatic antioxidant capacity, and repairing liver injury.<sup>17</sup> However, there are few reports on acute metabolic responses to maize or maize-enriched products in healthy subjects.<sup>18</sup>

Whole-grain pancakes are a popular traditional food in China, rich in whole grains and dietary fiber. However, there is currently no research on the acute effects of whole-grain pancakes on postprandial cardiometabolic risk factors in healthy individuals. We hypothesized that different whole-grain pancakes have different acute effects on cardiometabolic risk factors. Therefore, a randomized controlled crossover trial was designed to evaluate the effects of maize, buckwheat, and oat pancakes on cardiometabolic risk factors in healthy subjects.

## METHODS

### *Study design and participants*

A randomized, crossover design was performed to evaluate the effects of three different pancakes. The study was approved by Medical Ethics Committee of Affiliated Hospital to Qingdao University, and was registered at [www.chictr.org.cn](http://www.chictr.org.cn) (ChiCTR1900025559).

Eligible subjects included generally healthy adults, aged 19–24 years, limited to a body mass index (BMI) of 18.5 kg/m<sup>2</sup>–23.5 kg/m<sup>2</sup>. Criteria for inclusion required participants be non-smokers and non-drinkers, free of the following diagnoses, including CHD, diabetes mellitus, metabolic syndrome, hyperglycemia, dyslipidemia and hyperuricemia.

Twenty six healthy subjects were recruited from the students at Linyi University. One woman was excluded at the beginning of the test due to difficulty in drawing blood, and two men were excluded due to conflicts with their class schedules. The remaining participants were excluded mainly due to personal reasons. Ultimately, ten men and ten women [(mean ± SD): age 21.4±1.2 years (range 19–24 years); BMI 21.5±1.7 kg/m<sup>2</sup> (range 18.5–23.4 kg/m<sup>2</sup>)] participated in this study.

The trial was conducted at Women and Children's Health Care Hospital of Linyi. Questionnaires including health history, diet and physical activity were obtained from each participant. All eligible participants signed an informed consent form before any study procedures began and were informed about the study protocol. In addition, all participants were required to maintain their nor-

mal dietary intake and physical activity level, and abstain from caffeine and medication in the morning prior to and during the test.

### *Randomization*

After screening the subjects' age, gender, and BMI, the researchers randomly assigned 20 subjects who met the inclusion criteria to one of the four groups: the control steamed buns group (SB), the maize (*Zea mays L.*) pancake (MP) group, the buckwheat (*Fagopyrum esculentum Moench*) pancake (BP) group, and the oat (*Avena sativa L.*) pancake (OP) group using a computer-generated random number sequence. Special staff were assigned to be responsible for providing and distributing the test foods to the participants, as well as collecting and analyzing data, during the test period.

### *Treatments*

After an 8-h overnight fast, venous blood was collected from each subject. Subsequently, the subjects were randomly assigned to one of four groups (MP group, BP group, OP group, and SB group). Each food (100 g) was consumed together with 300 mL of water. Each meal was required to be consumed within 15 min, after which blood samples were collected from each subject at four time points: 0.5 h, 1 h, 2 h, and 3 h. All venous blood samples were centrifuged at 3000×g for 10 min at 4°C. The serum was immediately stored in a -80°C refrigerator. After a one-week washout period, the subjects were crossed over to another group.

The dietary nutrients of steamed buns and pancakes were summarized in Table 1. Energy (kJ/100g, GB/Z [National Guiding Technical Documents] 21922–2008), carbohydrate (g/100g, GB/Z 21922–2008), protein (g/100g, GB [Mandatory National Standards] 5009.5–2016), fat (g/100g, GB [Voluntary National Standards] 5009.6–2016), and dietary fiber (g/100g, GB 5009.88–2014) of the study food were determined. The average total dietary fiber intake was 14.4 g/d for oat pancakes, 13.8 g/d for buckwheat pancakes, 11.7 g/d for maize pancakes, and 2.3 g/d for steamed buns.

### *Anthropometric measurements*

All subjects underwent anthropometric measurements (including height, body weight, blood pressure, waist circumference, and hip circumference) conducted by researchers. Participants were kept in a quiet and comfortable environment during the entire trial period (8:00 am–11:00 am).

### *Laboratory measurements*

Serum glucose, TG, TC, HDL-C, LDL-C and C-reactive protein (CRP) were analyzed using enzyme-based colorimetric test on a Beckman AU5800 fully automatic biochemistry instrument (Beckman Coulter Commercial Enterprise Co., Ltd, Shanghai, China). Insulin was analyzed by chemiluminescence immunoassay on Cobas e 601 automatic analyzer (Roche Diagnostics Ltd, Shanghai, China).

**Table 1.** Comparison of dietary nutrients of the study foods

Variables	Steamed buns	Maize pancakes	Buckwheat pancakes	Oats pancakes
Protein (g/100 g)	8.5	8.3	13.2	11.8
Fat (g/100 g)	0.9	3.6	3.4	3.0
Carbohydrate (g/100 g)	52.2	73.4	69.3	69.4
Dietary fiber (g/100 g)	2.3	11.7	13.8	14.4
Energy (kJ/100 g)	1064	1522	1528	1491

### Statistical analysis

The sample size was determined based on randomized controlled trials using fiber cereals and maizeflakes.<sup>19</sup> The present study was designed to detect a 0.5 mmol/L difference in TG and a 1.4 mg/dL difference in serum glucose. We assumed a dropout rate of 25%, an SE of 0.4 mmol/L for TG, and an SE of 0.6 mg/dL for serum glucose, respectively. According to these assumptions, we enrolled 20 participants for the study to achieve an 80% power for a two-sided test with a significance level of 0.05.

All data were examined for normal distribution using the Shapiro-Wilk test before analysis. Baseline characteristic values were analyzed using one-way analysis of variance (ANOVA) to ensure that important covariates were equally distributed among the randomized groups. The effects of time (baseline and 4 post-treatment time points) and treatment (MP, BP, OP, and SB) on all parameters were evaluated using two-way repeated measures ANOVA. Within-group significance was assessed by repeated measures ANOVA, followed by post hoc multiple comparisons. The results in tables are presented as mean  $\pm$  SD, and those in figures as mean  $\pm$  SE. Differences were considered significant if  $p < 0.05$ . All statistical calculations were performed using SPSS version 22.0 (SPSS Inc., Chicago, IL, USA).

## RESULTS

### Characteristics of study participants

Figure 1 demonstrates a participant flow chart of the trial for the trial's crossover design. A total of twenty subjects (ten men and ten women) participated in this trial. These subjects had a mean age of  $21.4 \pm 1.2$  years and a mean BMI of  $21.5 \pm 1.7$  kg/m<sup>2</sup>. Baseline characteristics of the participants are shown in Table 2, and there were no significant differences across the treatment groups. Meanwhile, there was no significant change in the parameters related to liver and kidney functions among all participants. Changes in all parameters after the consumption of different pancakes within 3 hours are shown in Figure 2 and Figure 3.

### Postprandial cardiometabolic risk factors in participants

Compared with the baseline level, the blood glucose concentrations of the subjects significantly increased at 0.5h after consuming maize pancakes, buckwheat pancakes, and steamed buns ( $5.64 \pm 0.26$  mmol/L,  $5.39 \pm 0.21$  mmol/L, and  $5.73 \pm 0.23$  mmol/L, respectively;  $p < 0.01$ ). Half an hour after consuming oat pancakes, the blood glucose concentration was slightly elevated to  $5.33 \pm 0.17$  mmol/L (baseline:  $4.99 \pm 0.08$  mmol/L), but this difference was not statistically significant. Relative to baseline, blood glucose concentrations in the MP and OP groups were significantly reduced at 1 h and 2 h ( $p < 0.05$ ,  $p < 0.01$ ,

respectively). Although the blood glucose concentrations in the BP group and SB group were lower than the baseline level, no significant difference was observed. The insulin concentrations in all four groups significantly increased at 0.5h, 1h, and 2h ( $p < 0.001$ ,  $p < 0.001$ ,  $p < 0.05$ , respectively), and returned to the baseline level after 3h, which indicated that there was no statistical difference in insulin concentrations among the four food groups (Figure 2).

Compared with the baseline, the TG concentrations significantly increased at 2h and 3h after consumption of maize pancakes and oat pancakes ( $p < 0.05$ ). Compared with the SB group, serum TC concentrations were significantly lower in the OP group at 2 h and 3 h post-consumption ( $p < 0.05$  for both time points), while serum LDL-C concentrations were significantly reduced at 3 h ( $p < 0.05$ ); no statistically significant difference in HDL-C concentrations was observed between the two groups. Furthermore, oats showed a potential regulatory tendency on serum cholesterol levels, as reflected by the consistently lower TC, HDL-C, and LDL-C concentration curves in the OP group compared to other test foods (Figure 2, Figure 3). During the entire trial period, no significant changes in CRP concentration were observed either within or between groups (Figure 2).

## DISCUSSION

The present study was the first time to compare the effects of whole grain pancakes from different raw material sources with those of commonly consumed steamed buns on blood glucose and lipid profiles in healthy individuals. The aim of this study was to investigate whether different foods (maize, oat, or buckwheat pancakes) could exert effects in reducing postprandial blood glucose, insulin, lipids, or pro-inflammatory markers in healthy subjects. The trial investigated the potential acute hypocholesterolemic, hypoglycemic, and anti-inflammatory effects of whole grains and dietary fiber in healthy participants. Although the effects of whole grains and dietary fiber have been extensively reported in participants with CVD, obesity, hypercholesterolemia, diabetes, or metabolic syndrome, these studies have generally been medium and long-term interventions.

Previous studies show that oat consumption is associated with 5% and 7% reductions in TC and LDL-C levels, respectively.<sup>20</sup> Othman concluded that the inclusion of at least 3 g/day of oat  $\beta$ -glucan may be beneficial for cardiovascular health.<sup>20</sup> Additionally, some studies have reported that consuming 3 g per day of oat or barley  $\beta$ -glucan can significantly reduce blood cholesterol levels.<sup>21</sup> Maki claimed that consumption of whole-grain oat cereal as part of a weight loss dietary program had favorable effects on fasting lipid levels and waist circumference.<sup>22</sup>

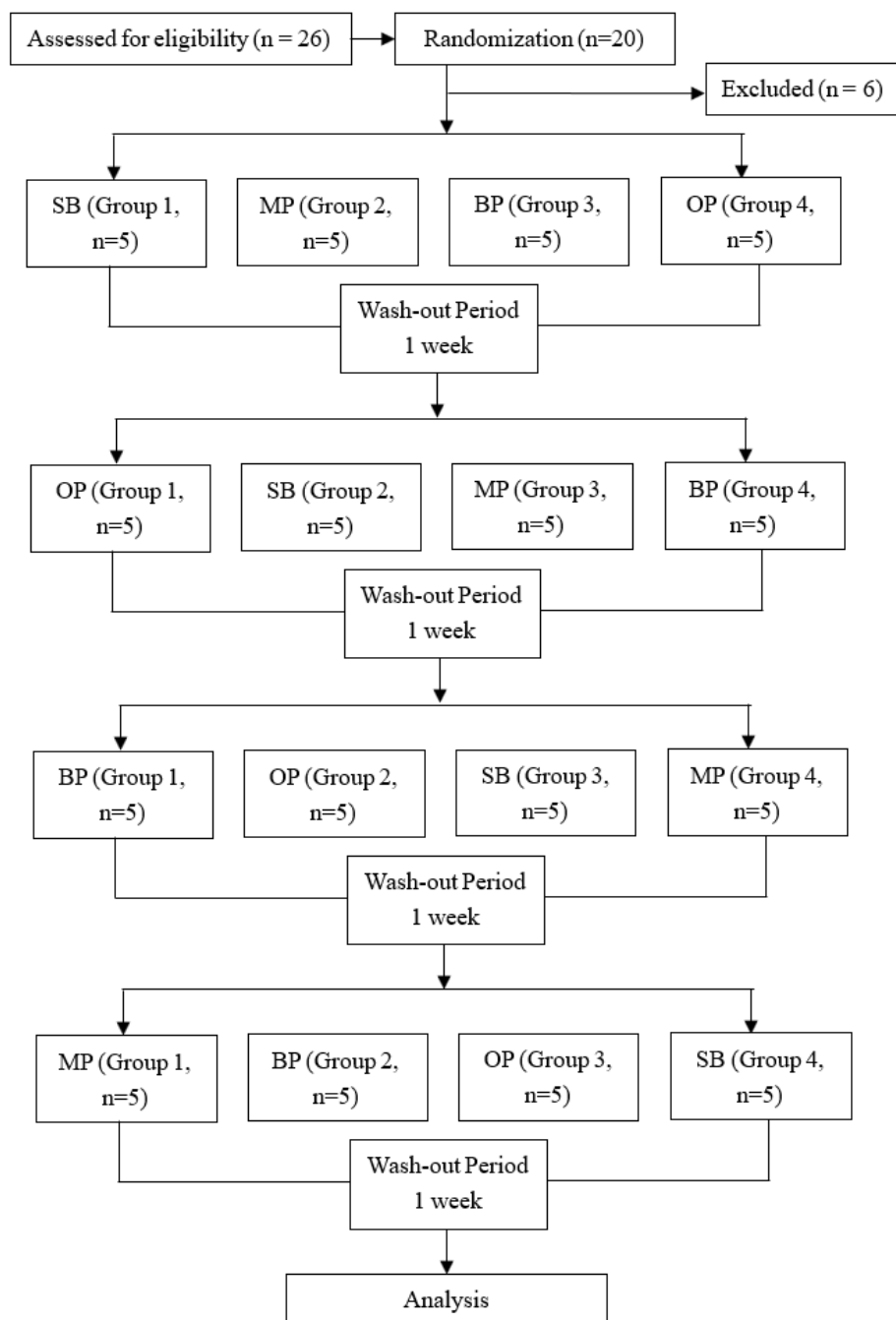


Figure 1. Flow chart of the screening process for the selection of eligible participants.

The mechanism is proposed to be that a meal containing such fiber increases the viscosity of gastric and intestinal contents, which may reduce the absorption or reabsorption of cholesterol and bile acids, thus lowering blood cholesterol concentration.<sup>23</sup> Similarly, Sun and colleagues observed that oat and tartary buckwheat-based food could reduce the concentrations of plasma lipids by inhibiting cholesterol absorption in the liver and promoting the excretion of fecal lipids and bile acids. Moreover, these foods increased the levels of SCFAs and effectively modulated the gut microbiota, thereby exerting hypocholesterolemic effects.<sup>24</sup> One animal study confirmed that whole barley prevented hypercholesterolemia in germ-free and humanized mice fed a high-fat diet. The molecular mechanism may involve inhibiting cholesterol synthesis by downregulating the expression of 3-hydroxy-3-

methylglutaryl coenzyme A reductase and sterol regulatory element-binding protein-1c, and simultaneously reducing cholesterol accumulation due to the upregulation of cholesterol 7 $\alpha$ -hydroxylase.<sup>25</sup> They speculated that the components of barley bran, including dietary fiber and phenolic compounds, play a major role.<sup>25</sup> The results of these studies are consistent with our findings, in which TC and LDL-C concentrations were significantly decreased at 2 h and 3 h after consumption of oat pancakes compared with the control group. Tosh reviewed 34 publications and summarized that intact cooked or fermented grains containing at least 3 g of  $\beta$ -glucan per meal are sufficient to significantly lower glycaemic response.<sup>26</sup> Despite oat  $\beta$ -glucan's established glucose-lowering potential, the OP group showed insignificant effects likely due to suboptimal  $\beta$ -glucan dose, structural degradation

**Table 2.** Baseline characteristics of participants

Characteristic	Value
Age (year)	21.4 ± 1.2
BMI (kg/m <sup>2</sup> )	21.5 ± 1.7
Body weight (kg)	60.5 ± 9.8
TG (mmol/L)	0.8 ± 0.3
TC (mmol/L)	3.9 ± 0.6
HDL-C (mmol/L)	1.3 ± 0.3
LDL-C (mmol/L)	2.1 ± 0.4
SBP (mmHg)	123.8 ± 12.5
DBP (mmHg)	73.8 ± 8.3
WC (cm)	74.0 ± 7.6
HC (cm)	94.9 ± 5.3
Glucose (mmol/L)	5.2 ± 0.3
CRP (mg/L)	2.2 ± 0.7
Insulin (uU/mL)	8.3 ± 2.7

BMI, body mass index; TG, triglycerides; TC, total cholesterol; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; SBP, systolic blood pressure; DBP, diastolic blood pressure; WC, waist circumference; HC, hip circumference; CRP, C reactive protein.

Data were presented as mean (standard deviation) for continuous measures

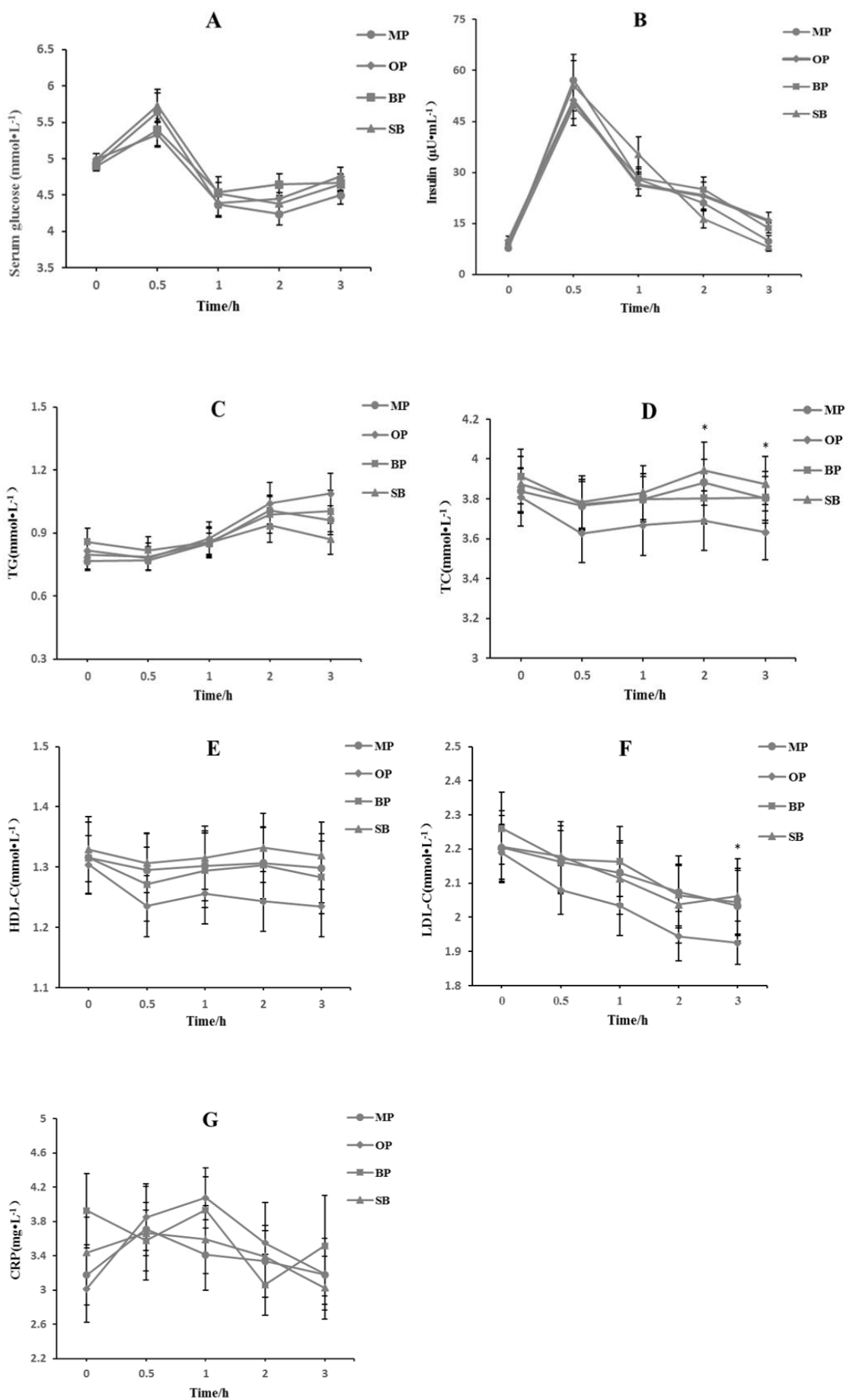
during pancake processing, and mismatched measurement timings that may have missed peak responses. Additionally, the single-meal intervention's limitations and lack of synergistic components could have further attenuated the glucose-regulating efficacy observed in previous studies.

He and colleagues reported that higher consumption of whole oats and oat bran, but not oat or barley  $\beta$ -glucan extracts, is closely associated with lower HbA1c, fasting glucose, and fasting insulin levels in subjects with T2D, hyperlipidaemia, and overweight, especially in patients with T2D. However, the effects of different food forms vary: extracted  $\beta$ -glucan does not exert the same effects as whole oats, whereas whole oats exhibit beneficial effects on HbA1c, fasting glucose, and insulin sensitivity.<sup>27</sup> The pancakes used in the present study were made from whole oats, maize, or buckwheat. In this study, no significant statistical differences in HbA1c, fasting glucose, or insulin levels were found between the experimental group and the control group. This may be because the effects of whole grains are not primarily evident in acute short-term periods, but instead rely on longer-term intervention to become fully manifested.

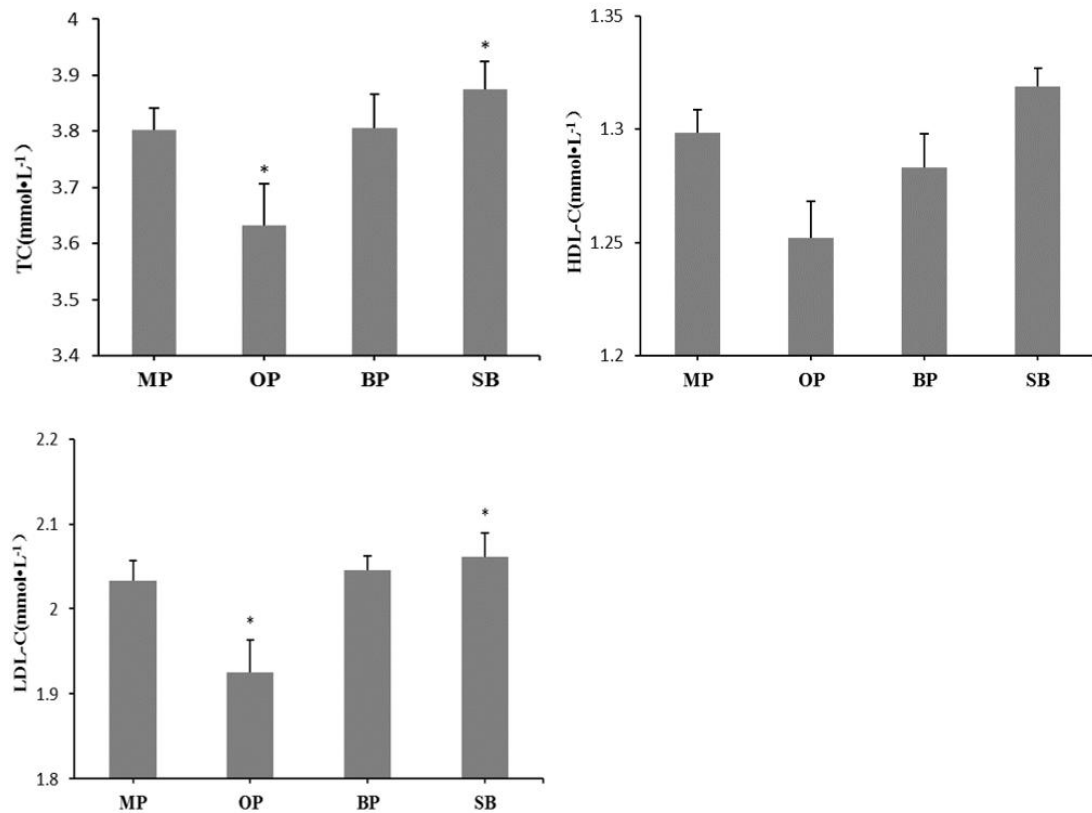
Buckwheat is considered a low-glycemic index (GI) food, and studies have shown that low-GI diets significantly improve lipid profiles in medium- and longer-term interventions, particularly in reducing TC and LDL-C levels.<sup>28, 29</sup> Studies have reported that buckwheat or buckwheat products may help prevent hyperlipidemia, which may in turn help delay the development of CVD. The cholesterol-lowering effect is primarily mediated by mechanisms such as increasing fecal sterol excretion, reducing the digestibility of buckwheat protein,<sup>30</sup> or down-regulating circulating cardiovascular risk factors and markers of oxidative stress, including TC, LDL-C, blood glucose, insulin, and the homeostasis model assessment (HOMA) index.<sup>31</sup> It has been reported that tartary buckwheat flour has cholesterol-lowering effects, and its cholesterol-lowering activity is associated with the inhibition of cholesterol absorption.<sup>32</sup> Furthermore, Mišan and colleagues found that intake of buckwheat porridge

can improve lipid profiles and reduce inflammation, indicating that consuming buckwheat porridge is an effective therapeutic approach to improve lipid profiles and inflammation in patients with metabolic syndrome.<sup>33</sup> Meanwhile, a study demonstrated that serum LDL-C and TG concentrations decreased significantly in animals fed diets containing 20% ground buckwheat products (buckwheat flour, meal, and bran) and rye-buckwheat bread containing these products. This indicates that long-term consumption of cereal products rich in ground buckwheat can improve lipid metabolism in the body.<sup>34</sup> Another animal study also reached a similar conclusion, namely that tartary buckwheat can reduce TG and LDL-C levels in mice on a high-fat diet.<sup>35</sup> Most studies to date have reported the effects of various buckwheat extracts or individual molecular components in cell experiments or animal models, with relatively few studies conducted on humans.<sup>36</sup> Despite buckwheat's documented lipid-modulating potential in medium/longer-term interventions and animal models, its insignificant acute effects on cholesterol and blood lipids in the present study may stem from the short-term intervention design and the use of healthy subjects with baseline normal lipid profiles, which limited the detectability of effects. Additionally, the discrepancy could be attributed to the consumption of whole buckwheat products in acute doses, as opposed to high-concentration extracts or prolonged exposure employed in most prior studies.<sup>36</sup>

Maize bran can reduce the glucose concentration in diabetic patients, according to one study that investigated the effects of soluble maize fiber (in the form of a drink) on TC concentration and postprandial glycemic response.<sup>37</sup> Meanwhile, an animal study indicated that the use of maize bran is more effective than that of whole wheat bran for lowering the blood glucose concentration of patients with hyperglycemia.<sup>38</sup> A blue maize extract can increase HDL-C levels and decrease serum TG and TC concentrations in Wistar rats, which may be a promising nutraceutical for the treatment of metabolic syndrome.<sup>39</sup> In this study, the effects of maize pancakes on



**Figure 2.** Changes of serum glucose (A), insulin (B), TG (C), TC (D), HDL-C (E), LDL-C (F) and CRP (G) within 3h in response to different pancakes and steamed buns. \*Between-group significance, OP group compared with SB group ( $p < 0.05$ ). All values are shown as the mean  $\pm$  SE.



**Figure 3.** Changes of serum TC, HDL-C and LDL-C at 3h in response to different pancakes and steamed buns. \*Between-group significance, OP group compared with SB group ( $p < 0.05$ ). All values are shown as the mean  $\pm$  SE.

blood lipids and blood glucose were not significant, possibly due to differences in intake amount, intervention duration, or maize cultivars.

Several limitations of this study should be addressed. One limitation is that there were some differences in the participants' diets on the day before the test. Some participants failed to maintain a light diet; for example, some may have eaten barbecue or hot pot on the day before the study began. In addition, for some participants, dinner time was too late, which could affect lipid or CRP concentrations.

### Conclusion

In conclusion, individuals at cardiometabolic risk need to be more cautious in their dietary choices. The results of this study showed that consumption of oat pancakes significantly improved TC and LDL-C concentrations in healthy individuals. This study is of great significance to such consumers and individuals with cardiometabolic risk factors. Larger-sample randomized controlled trials could deepen insights into the specific effects and mechanisms of whole-grain products on glucose and lipid profiles in healthy individuals. The findings will further advance the adoption of whole-grain diets (especially whole oat products) for more effective prevention of postprandial cardiometabolic risk.

### ACKNOWLEDGEMENTS

We are grateful to all the participants in our study for their support. Also, we would like to express our gratitude to Professor Duo Li for his guidance on the design of the research project.

### CONFLICT OF INTEREST AND FUNDING DISCLOSURES

The authors declare no conflict of interest.

This study was supported by the Qingdao Postdoctoral Application Research Project (RZ1900010484) and Optimization of Chondroitin Sulfate Preparation Process and Its Purification (X2025104521015). The study funders had no role in the study design, data collection, analysis, or preparation of the manuscript.

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