

# Original Article

# Combination Therapy of Flaxseed and Hesperidin Enhances the Effectiveness of Lifestyle Modification in Metabolic Syndrome: A Randomized Controlled Clinical Trial

Azita Hekmatdoost<sup>1</sup>, Vahideh Behrouz<sup>2</sup>, Zahra Yari<sup>3\*</sup>

- 1- Department of Clinical Nutrition and Dietetics, Faculty of Nutrition Sciences and Food Technology, National Nutrition and Food Technology, Research Institute Shahid Beheshti University of Medical Science, Tehran, Iran.
- 2- Department of Nutrition, School of Public Health, Kerman University of Medical Sciences, Tehran, Iran.
- 3- Department of Nutrition Research, National Nutrition and Food Technology Research Institute and Faculty of Nutrition Sciences and Food Technology, Shahid Beheshti University of Medical Sciences, Tehran, Iran.

Received: July 2024 Accepted: August 2024

# **ABSTRACT**

**Background and Objectives:** Regarding the increasing prevalence of metabolic syndrome (MetS), and its association with non-communicable chronic diseases, providing preventive and therapeutic strategies is a priority. A randomized controlled study was conducted to assess the effects of combination therapy of milled brown flaxseed and hesperidin during lifestyle intervention on MetS components.

**Materials and Methods:** Forty-eight subjects were randomly assigned to receive lifestyle intervention plus combination therapy of brown flaxseed (30 g milled) and hesperidin (two 500 mg capsules) or lifestyle modification alone for 12 weeks. MetS was assessed as a primary endpoint. The trial was registered in clinicaltrial.gov:NCT03737422.

**Results:** Forty-six percent of patients in the flaxseed-hesperidin group experienced more than three MetS factors improvement from baseline to week 12, while only 17% of patients in the control group fall in this category. A comparison of blood pressure changes throughout the study indicated a greater reduction in blood pressure in the intervention group rather than in the control group (-5.66 vs. -1.56 mmHg, P = 0.049). The reduction in plasma glucose (-20.35 vs. -7.46 mg/dL, P = 0.007) and triglyceride (-45.3 vs. -8.39 mg/dL, P = 0.020) in the flaxseed-hesperidin group was significantly more than the control group after 12 weeks of intervention.

**Conclusions:** Our results indicate that co-administration of flaxseed and hesperidin as an adjunct to lifestyle modification program is more effective than lifestyle modification alone in the treatment of metabolic syndrome.

**Keywords:** Metabolic syndrome, Flaxseed, Hesperidin, Lifestyle modification

# Highlights

- Supplementing with flaxseed and hesperidin improved more than three factors of metabolic syndrome in 46% of patients.
- Supplementing with flaxseed and hesperidin caused a significant weight loss.
- Supplementing with flaxseed and hesperidin improved systolic blood pressure.
- Supplementing with flaxseed and hesperidin improved glycemic control.

# Introduction

Metabolic Syndrome (MetS) is nowadays a serious health problem all over the world (1), as it overlaps with other metabolic disorders, such as diabetes, nonalcoholic fatty liver disease and cardiovascular diseases, while increases the risk of all-cause mortality by 50% (2, 3). Although various definitions have been proposed, the diagnosis of MetS is majorly based on a cluster of abnormal features including central adiposity, glucose intolerance, hypertension, hypertriglyceridemia, and

reduction in high-density lipoprotein (HDL) cholesterol (4). This condition may eventually lead to proinflammatory and pro-oxidant states (5-7). Depending on how the syndrome is defined, its prevalence will be different. The prevalence of MetS in Iran is estimated to be above 20%, which have a growing and age-related trend (8, 9).

Lifestyle modification (LSM) including increasing exercise and optimizing dietary composition, along with consumption of functional foods have been proposed as a therapeutic and preventive strategy for MetS (10, 11). Hesperidin and flaxseed have emerged as promising phyto-therapeutic agents, which may exert potential benefits on health mainly through improving the antioxidant status (12), and providing adequate fiber and omega-3 (13-15), respectively. The therapeutic effects of these two compounds on metabolic dysregulation can be attributed to their powerful biological properties on improving lipid profiles (16-18), glucose homeostasis, and insulin resistance (19-22).

Considering lack of data on such combination therapies in patients with metabolic syndrome, we aimed to investigate whether patients following LSM program along with supplementing with 30 gmilled brown flaxseed and one gram hesperidin per day, would have better metabolic parameters compared with those who just follow the LSM program.

# **Materials and Methods**

# **Subjects**

To assess the effects of combination therapy of milled brown flaxseed and hesperidin during lifestyle modification on MetS components, 80 men and women with MetS were recruited from a nutrition and diet therapy clinic affiliated with National Nutrition and Food Technology Research Institute (NNFTRI). For study inclusion, participants were required to be aged 18 to 70 years and diagnosed with MetS based on National Cholesterol Education Program's Adult Treatment Panel III report criteria (23).

exclusion criteria The were any history of cardiovascular, pulmonary, renal, hepatic and gastrointestinal disease. Additionally, taking antiglucose-lowering medications, hypertensive, lipid modifying agents and omega-3 dietary supplement, being under treatment with steroids and nonsteroidal antiinflammatory drugs, being on calorie-restricted regimen within 3 months before the commencement of the trial, being pregnant, lactating were the other main exclusion criteria.

Of the 80 subjects screened, 57 patients including 31 females and 26 males were deemed eligible for inclusion; of which nine were reluctant to participate in the trial. Eventually, 48 patients were enrolled. A flow diagram of the trial is presented in Figure 1. Twenty four patients consumed 30 g/day milled brown flaxseed two 500 mg capsules of hesperidin along with LSM and 24 patients followed LSM program for 12 weeks.

### Study design

We conducted a randomized, open labeled, controlled study on patients with metabolic syndrome. The trial was registered with ClinicalTrials.gov, number NCT03737422.

Ethical approval was granted by the Ethical Committee of Shahid Beheshti University of Medical Sciences and all procedures followed were in accordance with the Helsinki Declaration. Participation was voluntary, and all patients provided written informed consent, before beginning the study.

The participants were recruited from advertisement and primary health care centers between January 2018 and July 2018. One of the research assistants who had no other involvement in the trial randomly allocated patients to groups. Since the study was designed as open-labeled, neither patients nor researchers were blind to the interventions. All patients underwent follow-up visits at 4, 8, and 12 weeks after enrolment, at which dietary advice was repeated to patients and adequate supplements were given. Participants were explicitly counseled regarding supplement consumption and how to follow the LSM program, before initiation of the trial.

# **Supplementation**

Each patient was assigned to one of two treatment groups using a table of random numbers for twelve-week intervention of either: 1) lifestyle modification program, 2) lifestyle modification plus combination therapy of brown flaxseed (30 g milled) and hesperidin (two 500 mg capsules). Also, patients were instructed to divide supplements into two equal doses (one capsule of hesperidin and 15 g milled flaxseed), one with breakfast and the other at lunchtime. Hesperidin was 95% pure. The chemical compounds content of flaxseed is well-known. An analysis of brown flaxseed showed about 41% fat, 20% protein, 28% total dietary fiber and 3.4% mineral-rich ash (15).

Nutrition and Food Sciences Research 12 Vol 11, No 3, Jul-Sep 2024

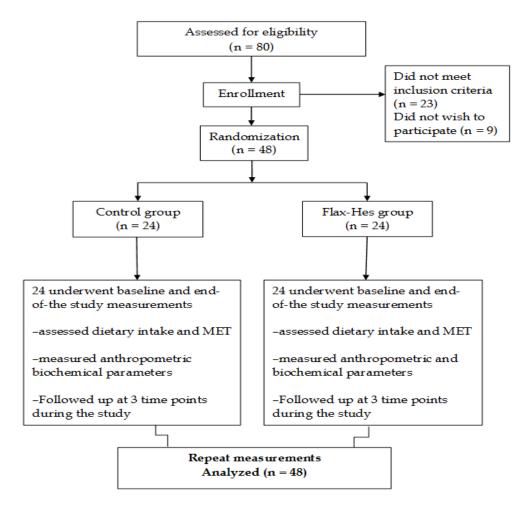


Figure 1. Flow chart depicting the study design

# Blood sampling and laboratory tests

Blood samples were collected in the morning after overnight fasting, before and after the intervention, for biochemical measurements. After centrifugation of blood at 3700 rpm for 10 min at room temperature, the separated serum was stored frozen at -80°C until the tests were run.

The levels of serum HDL-C and Triglyceride (TG) were measured by the standard enzymatic method (Pars Azmoon kit, Tehran, Iran). Fasting serum glucose was analyzed by the glucose-oxidase method (Pars Azmoon Co., Tehran, Iran) and serum insulin was determined by enzyme-linked immunosorbent assay (ELISA) (Monobind Inc., USA).

Insulin Resistance was calculatedby using this formula: Fasting Glucose (mg/dl) x fasting Insulin ( $\mu$ U/mL) / 405. Also, the following formula was used to calculate insulin sensitivity:1/ [log glucose (mg/dL) + log insulin( $\mu$ U/mL)].

#### **Anthropometrics measurements**

A clinical evaluation including medical history and clinical data sheet was performed. All participants

underwent a detailed anthropometric assessment at the baseline and after completion of the intervention. Height and weight were measured by a trained research assistant based on the standard protocol using calibrated Seca scale with attached stadiometer and recorded to the nearest 0.5 cm and 0.1 kg, respectively. For body mass index (BMI) calculation, weight in kilograms divided by the square of height in meters. Minimum waist circumference was measured with a tape to the nearest 0.5 cm so that we could properly identify subjects who had metabolic syndrome.

# Dietary assessment and physical activity

For dietary assessment, we completed three 24-hour food recall questionnaires at the at baseline and after the 12-week study intervention for each patient. Dietary intakes were then analyzed by using the nutritionist version 4 (N4) software. To assess physical activity level, a questionnaire was completed for each patient, which included a list of daily activities, the frequency and time of activities spent per day. Physical activity levels were expressed as metabolic equivalent (METs h/day).

Vol 11, No 3, Jul-Sep 2024

Nutrition and Food Sciences Research

#### **Blood pressure**

After 15 min of resting, blood pressure was measured twice on the right arm, using a standard mercury sphygmomanometer, at baseline and at 12 weeks. The average of two measurements with at least 10-min interval was recorded.

#### Data analysis

All analyses were performed using the statistical package for social sciences software (IBM SPSS Statistics for Windows, release 21.0. Armonk, NY, USA: IBM Group). Before further analysis, normal distribution of the variables was checked by applying the Shapiro-Wilk test. Comparison of changes before and after intervention in each group were analyzed by paired t-test. In addition, as appropriate, student's t-test and analysis of covariance (ANCOVA) were applied to assess differences between two groups using baseline value of the outcome, age, sex and mean changes in BMI, MET and energy as covariates. P <0.05 was considered as the significance level.

# **Results**

A total of 80 patients completed the initial screening, of which 32 were excluded due to a lack of interest and failure to meet the entrance criteria. Finally, 48 patients received the allocated intervention. All patients completed a 12-week follow-up program, with an overall intervention

adherence rate of 100%. Flow diagram of the study is presented in Figure 1.

As shown in Table 1, there was no significant difference between two groups with regard to baseline characteristics including anthropometric measurements, blood pressure and serum biochemistry tests. There were 25 men (52%), and 23 women (48%) among all participants. The mean age of participants was  $44.5 \pm 10.7$  and  $46.7 \pm 11.5$  in intervention and control group, respectively. No significant differences among the distributions of sex and age between two groups were observed.

Furthermore, data on diet indicated no significant differences in total energy, macronutrients or micronutrients intake between groups. Although the results of the paired t-test revealed a significant decrease in energy intake and macronutrients, except for protein, throughout the study in both groups; but this difference between the two groups was not significant.

A paired t-test was used to compare the changes in each group during the 12-week intervention period. All of the measurements in flax-hes group were significantly improved, except for HDL-C. In control group, in addition to HDL-C, systolic blood pressure and serum triglyceride did not show significant changes (Table 2).

**Table 1.** Baseline characteristics at enrollment <sup>1</sup>

characteristics	Total	Flax-Hes group	Control group	P value
characteristics	(n = 48)	(n = 24)	(n = 24)	
Age (y)	$45.63 \pm 11.00$	$44.50 \pm 10.66$	$46.75 \pm 11.46$	0.485
Sex (M/F)	25/23	12/12	13/11	1.000
Metabolic characteristics				
Height (cm)	$166.96 \pm 10.54$	$167.38 \pm 9.03$	$166.54 \pm 12.04$	0.787
Weight (kg)	$91.24 \pm 14.96$	$92.08 \pm 16.06$	$90.40 \pm 14.08$	0.700
WC (cm)	$104.71 \pm 7.50$	$105.25 \pm 6.43$	$104.17 \pm 8.55$	0.622
BMI (kg/m2)	$33.09 \pm 5.42$	$33.27 \pm 5.43$	$32.92 \pm 5.51$	0.826
Blood pressure (mmHg)				
Systolic	$128.73 \pm 14.94$	$129.71 \pm 14.77$	$127.80 \pm 15.35$	0.660
Diastolic	$85.24 \pm 12.93$	$86.21 \pm 14.66$	$84.32 \pm 11.26$	0.615
MET (h/d)	$31.93 \pm 4.45$	$32.65 \pm 2.97$	$31.22 \pm 5.54$	0.270
Energy (kcal)	$2473.56 \pm 487.94$	$2535.15 \pm 540.39$	$2411.98 \pm 431.96$	0.388
Serum biochemistry tests				
FBS (mg/dL)	$113.05 \pm 19.61$	$117.08 \pm 21.16$	$107.96 \pm 16.62$	0.123
Insulin (mU/L)	$13.18 \pm 7.17$	$12.58 \pm 6.85$	$13.94 \pm 7.67$	0.545
HOMA-IR	$3.69 \pm 2.34$	$3.78 \pm 2.57$	$3.57 \pm 2.10$	0.773
QUICKI	$0.33 \pm 0.03$	$0.33 \pm 0.03$	$0.32 \pm 0.02$	0.902
Triglyceride (mg/dL)	$162.02 \pm 71.27$	$159.83 \pm 79.09$	$164.78 \pm 61.98$	0.824
HDL-C (mg/dL)	$35.51 \pm 9.69$	$36.15 \pm 9.82$	$34.68 \pm 9.73$	0.629

<sup>&</sup>lt;sup>1</sup> WC, waist circumference; BMI, body mass index; MET, metabolic equivalent of tasks; FBS, fasting blood sugar; HDL-C, high density lipoprotein:

Nutrition and Food Sciences Research 14 Vol 11, No 3, Jul-Sep 2024

<sup>&</sup>lt;sup>2</sup> Mean ± SD (all such values)

Table 2. Mean changes (95% CI) from baseline in metabolic characteristics by treatment group

	Flax-Hes group (n= 24)	Control group $(n = 24)$	P value <sup>1</sup>
Weight (kg)	- 5.10 (-6.45, -3.76)	- 3.10 (-4.65, -1.55)	0.048
P value <sup>2</sup>	<0.001	0.001	
WC (cm)	-6.75	-3.57	0.306
P value <sup>2</sup>	(-10.53, -2.97) 0.001	(-6.43, -0.73) 0.017	
Systolic blood pressure	-5.66 (-10.53, -1.29)	-1.56 (-3.57, 0.45)	0.049
P value <sup>2</sup>	0.013	0.123	
Diastolic blood pressure	-3.83 (-8.43, 0.76)	-2.92 (-5.65, -0.18)	0.456
P value <sup>2</sup>	0.048	0.037	
FBS	-20.35 (-28.07, -12.63)	-7.46 (-12.55, -2.37)	0.007
P value <sup>2</sup>	<0.001	0.007	
Insulin	-3.43 (-4.86, -1.99)	-2.46 (-4.21, -0.72)	0.373
P value <sup>2</sup>	< 0.001	0.008	
HOMA-IR	-1.59 (-2.37, -0.83)	-0.73 (-1.22, -0.24)	0.055
P value <sup>2</sup>	<0.001	0.006	
QUICKI	0.03 (0.01, 0.04)	0.01 (0.004, 0.014)	0.023
P value <sup>2</sup>	0.001	0.001	
Triglyceride	-45.30 (-64.16, -26.43)	-8.39 (-43.40, 26.62)	0.020
P value <sup>2</sup>	<0.001	0.620	
HDL-C	2.73 (-1.13, 6.59)	1.30 (-1.36, 3.96)	0.535
P value <sup>2</sup>	0.155	0.317	

<sup>&</sup>lt;sup>1</sup> Based on an ANCOVA model that regressed changes from baseline on treatment group, baseline value of the outcome, age, sex and mean changes in BMI, MET and energy.

The difference of changes between two groups during the interventions were analyzed using ANCOVA adjusted for baseline value of the outcome, age, sex and mean changes in BMI, MET and energy. Mean changes of metabolic characteristics of participants from baseline are presented in Table 2. Anthropometric data comparison between the two groups showed a significant reduction in weight (P= 0.048). With regard to waist circumference, although in the intervention group, Waist circumference (WC) reduction was about twice that of the control group (-6.75 cm vs -3.57 cm), but this difference was not statistically significant.

Comparison of blood pressure changes throughout the study between the two groups indicated a greater reduction in blood pressure in the intervention group, although this difference was only for systolic blood pressure (P = 0.049). Regarding the glucose homeostasis factors, only significant differences were observed in the values of fasting blood sugar (FBS) and quantitative insulin-sensitivity check index (QUICKI). Also, we found

approximately near to the significant level differences in HOMA-IR (P = 0.055). However, the changes in insulin levels were fairly similar in both groups (Table 2).

Based on the results presented in Table 2, the reduction in TG was significantly more in flax-hes group than the control group (-45.3 *vs.* -8.39mg/dL) after 12 weeks of intervention. Although HDL levels increased within both groups, there was no significant difference between the two groups.

After hes-flax consumption, we observed a significant decrease in the number of people with metabolic syndrome. Analysis on MetS factors improvement in response to intervention is presented in Table 3. These results indicate that 46% of patients in the flax-hes group experienced more than three MetS factors improvement from baseline to week 12, while only 17% of patients of the control group fall in this category. As a whole, there was a statistically significant difference between the two groups in terms of changes in MetS from the beginning to the end of the study.

15

<sup>&</sup>lt;sup>2</sup> Paired t-test

**Table 3.** Frequency distribution of MetS factors improvement

	Flax-hes group	Control	$P^*$
No change	3	10	0.023
One factor	4	6	0.477
Two factors	6	4	0.477
≥ Three factors	11	4	0.029

<sup>\*</sup> P values based on Pearson chi squared test

#### Adverse events and compliance

Patients were instructed to return any unused supplements at every follow up visit. None of the patients had missed more than 10% of the supplements. Also, no complication or serious side effects were observed during the study among the participants that could be attributed to intervention.

#### **Discussion**

We found that including daily intake of 30g of whole flaxseed and one gram of hesperidin can modulate metabolic risk factors. Our results indicate that metabolic syndrome was significantly improved over 12 weeks by adding flaxseed and hesperidin to a lifestyle modification program. The combination of hesperidin, as a flavonoid, and flaxseed, as a functional food, can exhibit a great potency in preventing and controlling MetS through their combined anti-inflammatory and anti-oxidant properties as well as high fiber content. Reducing the risk of MetS has been stem from increasing insulin sensitivity, improving dyslipidemia and lowering blood pressure succeeding consumption of flaxseed and hesperidin.

In the present study, although the concentration of HDL did not change, adecline in the concentration of triglyceride was observed in response to hes-flax supplementation. Lack of effectiveness of a lifestyle modification program can be due to a short duration of intervention and lack of complete adherence to the program. These findings are consistent with preceding studies. Administration of 500 mg of purified hesperidin to patients with hypertriglyceridemia markedly reduce serum triglyceride levels over a 24-week period (24), while 800 mg of hesperidin failed to affect plasma HDLcholesterol after 4 weeks (25). Though in another study, oral administration of 500 mg hesperidin for 3 weeks resulted in a significant increase in HDL, while failed to reduce triglyceride levels (26). One of the proposed mechanisms of the hesperidin effects on lipid profile modulation is through increasing the transcription of the PPAR-α gene (27). In order to achieve significant clinical outcomes, flavonoids should be sufficiently consumed. Moreover, the patient population and length of study are other imperative determinants. In fact, lipid-lowering effects of purified hesperidin should be interpreted with caution as this may not be achievable with usual dietary intake (27).

Flaxseed, in turn, has beneficial effects on lipid profile, which are due to its components specially fiber and omega-3 fatty acids. A recent animal study revealed that adding whole grain flaxseed to the standard rat diet significantly ameliorated the serum levels of TG and HDL-C (28). Also, it was reported in a study conducted on rabbits that supplementation with flaxseed lignans, although could not alter serum triglyceride levels, increased serum HDL significantly (29). High content of flaxseed lignan can reduce triglyceride levels through suppressing the sterol regulatory element-binding proteins (SREBP) mRNA (30). It has been revealed that the hypocholesterolemic effect of flaxseed is more pronounced than wheat in hypercholesterolemic adults, though it led to a slight decrease in HDL-C concentration (31). What seems to have arisen from this study is that fiber alone cannot fully account for all the positive effects of flaxseed in improving the lipid profile. At least part of these beneficial effects is due to omega-3 fatty acids, which has been mentioned in previous studies (32, 33). However, the findings of some studies are inconsistent with these results (34), possibly because the dose or duration of the intervention was insufficient. Since it has been reported that at least consumption of 30 grams flaxseed is required to convert ALA to EPA in young, healthy adults (35).

One of the most remarkable findings of the present study was the improvement of glycemic parameters, including fasting plasma glucose, insulin, HOMA-IR and QUICKI after 12 weeks treatment with combination of hesperidin and flaxseed, which is in line with the findings of most previous studies (36-38). Although no study has evaluated the effects of combination of these supplements, there are several studies that have shown the beneficial effects of each of them alone on glycemic homeostasis. A study by Cunnane et al. reported the hypoglycemic effects of both whole flaxseed and isolated flaxseed fiber (18). Improvements in either HOMA-IR or QUICKI following flaxseed consumption was described for the first time in 2013 (31). The prominent point of these studies is that flaxseed supplementation can only affect glycemic parameters in interventions duration of at least 12 weeks. The study conducted by Barre and colleagues (39) is in line with this finding, in which 3 months supplementation of flaxseed lignanin patients with type 2 diabetes led to plasma glucose reduction. This was also confirmed in

Nutrition and Food Sciences Research Vol 11, No 3, Jul-Sep 2024

another study in which flaxseed consumption in patients with MetS could result in blood glucose control and insulin resistance decline after 12 weeks (40). Also, a recently published meta-analysis in this area also indicated this, but with this difference that only whole flaxseed, but not flaxseed oil and lignan extract, has significant effects on controlling glycemic parameters (41). A possible explanation for the necessity of long-term intervention (≥12 weeks) is that improving glucose control and insulin sensitivity require an increase in EPA and DHA concentrations (42, 43) and ALA conversion to these two fatty acids is time-consuming (44). Also, the function of the gut microflora is enhanced gradually by flaxseed fiber consumption, which in turn led to improved blood glucose control and insulin function (45, 46). As well, the higher initial concentration of plasma glucose (>100 mg/dL), the more pronounced changes can be achieved via the flaxseed supplement (41).

According to the Nurses' Health Studies (NHS) I and II, higher urinary excretion of hesperetin is associated with a lower middle- and long-term risk of type 2 diabetes (OR: 0.68) (47). Improving insulin sensitivity and reducing insulin resistance has been stated repeatedly after treatment with hesperidin (26, 36, 48). Increased skeletal muscle glucose uptake has been shown following the positive regulation of insulin signaling by hesperidin. That is why intake of orange juice containing hesperidin following a high-fat and high-carbohydrate meal, prevents increases in the expression of suppressor of cytokine signaling-3, a negative modulator of insulin signaling (49).

Our results demonstrated that patients supplementing with the combination of flaxseed and hesperidin experienced lowered systolic blood pressure after 12 weeks. The anti-hypertensive effect of flaxseed and hesperidin has been shown in previous studies. In the same way, a study conducted in 2015 found that higher urinary polyphenol excretion was allied with lower systolic and diastolic blood pressure, which was attributed to nitric oxide production (50, 51). A meta-analysis of the effects of flaxseed on blood pressure showed that supplementation of various products of the flaxseed can exert beneficial effects on systolic and diastolic blood pressure control (52). This study stated that flaxseed can cause a decrease of 2.85/2.39 mmHg in blood pressure. Since patients with MetS are at high risk for cardiovascular disease, this finding is very valuable. Similarly, Heart Outcome Evaluation study proposed that a 3.3/1.4 mmHg reduction in blood pressure is associated with a 22% decline of relative risk of cardiovascular mortality (53). It is supposed that flaxseed lignan can possess an inhibitory action on angiotensin-converting enzyme (ACE) which results in blood pressure reduction (54). Also, ALA in flaxseed suppresses the soluble

epoxide hydrolase activity, which reduces blood pressure through inhibiting the production of inflammatory oxylipins and following vasoconstriction, inflammation and hypertension (55). Although the blood pressure lowering effects of flaxseed have been attributed to fiber and ALA, the exact mechanisms involved in this effect are still not fully understood. Although bioavailability of ALA in whole flaxseed is lower than that of flaxseed oil, whole grain flaxseed has the advantage of providing all bioactive components and fiber with well tolerability (56, 57).

Our study showed significant reduction in weight and waist circumference after 12 weeks supplementation with the combination of flaxseed and hesperidin, although the difference of WC was not significant between two groups, compared to baseline. Studies regarding the effect of hesperidin on anthropometric measures are limited. Supplementation with citrus juice containing vitamin C and hesperidin have no significant effect on weight and WC (48). Therefore, it appears that the weight loss observed in present study is possibly due to the flaxseed, which is in line with previous studies (31, 58). Recent review and meta-analysis study also confirms the positive effects of flaxseedon weight loss, especially if the intervention durations of ≥12 weeks (59). ALA can exert its anti-obesity effects via subsequent converting into long chain poly unsaturated fatty acids in the body (60-62). Furthermore, flaxseed lignans under the gut microflora produced enterolactone, the main component of mammalian lignans, which has been negatively correlated with obesity (63, 64).

The present study does however have its strengths; to the best of our knowledge it is the first interventional study investigating the effect of flaxseed and hesperidin combination as an adjunct to lifestyle modification for the treatment of metabolic syndrome. Although the exact clinical implication of these findings is yet undefined, but could be proposed as a preventive strategy. It needs further studies to explore full potential of flaxseed and hesperidin and confirm these findings.

In conclusion, incorporating 30g of brown flaxseed and one-gram hesperidin into a healthy diet along with increasing physical activity seems reasonable and valuable to reduce risk of MetS through ameliorating insulin resistance, lowering blood pressure, improving glycemic control and modifying lipid profile (15, 65, 66).

# **Acknowledgement**

Authors have no acknowledgments to declare.

#### **Financial disclosure**

The authors declare that they have no competing interests.

Vol 11, No 3, Jul-Sep 2024

Nutrition and Food Sciences Research

# Funding/Support

**Authors' contributions.** Conceptualization, ZY and AH; Formal analysis, ZY; Methodology, ZY and VB; Project administration, ZY and AH; Writing – original draft, VB and ZY; Writing – review & editing, ZY and AH. All authors read and approved.

# References

- Miranda PJ, DeFronzo RA, Califf RM, Guyton JR. Metabolic syndrome: definition, pathophysiology, and mechanisms. American heart journal. 2005;149:33-45.
- Mottillo S, Filion KB, Genest J, Joseph L, Pilote L, Poirier P, Rinfret S, Schiffrin EL, Eisenberg MJ. The metabolic syndrome and cardiovascular risk: a systematic review and meta-analysis. Journal of the American College of Cardiology. 2010;56:1113-32.
- Tarantino G, Finelli C. What about non-alcoholic fatty liver disease as a new criterion to define metabolic syndrome? World journal of gastroenterology: WJG. 2013;19:3375.
- 4. Alberti K, Eckel RH, Grundy SM, Zimmet PZ, Cleeman JI, Donato KA, Fruchart J-C, James WPT, Loria CM, Smith Jr SC. Harmonizing the metabolic syndrome: a joint interim statement of the international diabetes federation task force on epidemiology and prevention; national heart, lung, and blood institute; American heart association; world heart federation; international atherosclerosis society; and international association for the study of obesity. Circulation. 2009;120:1640-5.
- 5. Cornier M-A, Dabelea D, Hernandez TL, Lindstrom RC, Steig AJ, Stob NR, Van Pelt RE, Wang H, Eckel RH. The metabolic syndrome. Endocrine reviews. 2008;29:777-822.
- Ford ES. Intake and circulating concentrations of antioxidants in metabolic syndrome. Current atherosclerosis reports. 2006;8:448-52.
- Lee K-U. Oxidative stress markers in Korean subjects with insulin resistance syndrome. Diabetes research and clinical practice. 2001;54:S29-S33.
- Gharipour M, Sarrafzadegan N, Sadeghi M, Khosravi A, Hoseini M, Khosravi-Boroujeni H, Khaledifar A. The metabolic syndrome and associated lifestyle factors among the Iranian population. Advanced biomedical research. 2015;4.
- Mazloomzadeh S, Rashidi Khazaghi Z, Mousavinasab N. The Prevalence of Metabolic Syndrome in Iran: A Systematic Review and Meta-analysis. Iranian journal of public health. 2018 Apr;47:473-80.
- Brown L, Poudyal H, Panchal S. Functional foods as potential therapeutic options for metabolic syndrome. Obesity reviews. 2015;16:914-41.
- 11. de la Iglesia R, Loria-Kohen V, Zulet M, Martinez J, Reglero G, Ramirez de Molina A. Dietary strategies implicated in the prevention and treatment of metabolic syndrome. International journal of molecular sciences. 2016;17:1877.
- 12. Cavia- Saiz M, Busto MD, Pilar- Izquierdo MC, Ortega N, Perez- Mateos M, Muñiz P. Antioxidant properties, radical scavenging activity and biomolecule protection capacity of flavonoid naringenin and its glycoside naringin: a comparative study. Journal of the Science of Food and Agriculture. 2010;90:1238-44.

- Poudyal H, Panchal SK, Diwan V, Brown L. Omega-3 fatty acids and metabolic syndrome: effects and emerging mechanisms of action. Progress in lipid research. 2011;50:372-87.
- Oomah BD. Flaxseed as a functional food source. Journal of the Science of Food and Agriculture. 2001;81:889-94.
- 15. Morris DH. Flax: A health and nutrition primer: Flax Council of Canada; 2007.
- 16. E Mulvihill E, W Huff M. Citrus flavonoids and the prevention of atherosclerosis. Cardiovascular & Haematological Disorders-Drug Targets (Formerly Current Drug Targets-Cardiovascular & Hematological Disorders). 2012;12:84-91.
- Bloedon LT, Szapary PO. Flaxseed and cardiovascular risk. Nutrition reviews. 2004;62:18-27.
- 18. Cunnane SC, Ganguli S, Menard C, Liede AC, Hamadeh MJ, Chen Z-Y, Wolever TM, Jenkins DJ. High α-linolenic acid flaxseed (Linum usitatissimum): some nutritional properties in humans. British Journal of Nutrition. 1993;69:443-53.
- 19. Munir KM, Chandrasekaran S, Gao F, Quon MJ. Mechanisms for food polyphenols to ameliorate insulin resistance and endothelial dysfunction: therapeutic implications diabetes and its cardiovascular for complications. Journal of American Physiology-Endocrinology and Metabolism. 2013;305:E679.
- Jung UJ, Lee M-K, Jeong K-S, Choi M-S. The hypoglycemic effects of hesperidin and naringin are partly mediated by hepatic glucose-regulating enzymes in C57BL/KsJ-db/db mice. The Journal of nutrition. 2004;134:2499-503.
- Rhee Y, Brunt A. Flaxseed supplementation improved insulin resistance in obese glucose intolerant people: a randomized crossover design. Nutrition journal. 2011;10:44.
- Hutchins AM, Brown BD, Cunnane SC, Domitrovich SG, Adams ER, Bobowiec CE. Daily flaxseed consumption improves glycemic control in obese men and women with pre-diabetes: a randomized study. Nutrition research. 2013;33:367-75.
- 23. Expert Panel on Detection E. Executive summary of the third report of the National Cholesterol Education Program (NCEP) expert panel on detection, evaluation, and treatment of high blood cholesterol in adults (Adult Treatment Panel III). Jama. 2001;285:2486.
- 24. Miwa Y, Mitsuzumi H, Sunayama T, Yamada M, Okada K, Kubota M, Chaen H, Mishima Y, Kibata M. Glucosyl hesperidin lowers serum triglyceride level in hypertriglyceridemic subjects through the improvement of very low-density lipoprotein metabolic abnormality. Journal of nutritional science and vitaminology. 2005;51:460-70.
- 25. Demonty I, Lin Y, Zebregs YE, Vermeer MA, van der Knaap HC, Jäkel M, Trautwein EA. The Citrus Flavonoids Hesperidin and Naringin Do Not Affect Serum Cholesterol in Moderately Hypercholesterolemic Men and Women–3. The Journal of nutrition. 2010;140:1615-20.
- 26. Rizza S, Muniyappa R, Iantorno M, Kim J-a, Chen H, Pullikotil P, Senese N, Tesauro M, Lauro D, Cardillo C. Citrus polyphenol hesperidin stimulates production of nitric oxide in endothelial cells while improving endothelial function and reducing inflammatory markers in patients with metabolic syndrome. The Journal of Clinical Endocrinology & Metabolism. 2011;96:E782-E92.

Nutrition and Food Sciences Research Vol 11, No 3, Jul-Sep 2024

- Assini JM, Mulvihill EE, Huff MW. Citrus flavonoids and lipid metabolism. Current opinion in lipidology. 2013 Feb;24:34-40.
- 28. Naik HS, Srilatha C, Sujatha K, Sreedevi B, Prasad T. Supplementation of whole grain flaxseeds (Linum usitatissimum) along with high cholesterol diet and its effect on hyperlipidemia and initiated atherosclerosis in Wistar albino male rats. Veterinary world. 2018 Nov;11:1433-9.
- Prasad K. Hypocholesterolemic and antiatherosclerotic effect of flax lignan complex isolated from flaxseed. Atherosclerosis. 2005 Apr;179:269-75.
- Fukumitsu S, Aida K, Ueno N, Ozawa S, Takahashi Y, Kobori M. Flaxseed lignan attenuates high-fat diet-induced fat accumulation and induces adiponectin expression in mice. British Journal of Nutrition. 2008;100:669-76.
- Bloedon LT, Balikai S, Chittams J, Cunnane SC, Berlin JA, Rader DJ, Szapary PO. Flaxseed and cardiovascular risk factors: results from a double blind, randomized, controlled clinical trial. Journal of the American College of Nutrition. 2008;27:65-74.
- 32. Mirmasoumi G, Fazilati M, Foroozanfard F, Vahedpoor Z, Mahmoodi S, Taghizadeh M, Esfeh NK, Mohseni M, Karbassizadeh H, Asemi Z. The Effects of Flaxseed Oil Omega-3 Fatty Acids Supplementation on Metabolic Status of Patients with Polycystic Ovary Syndrome: A Randomized, Double-Blind, Placebo-Controlled Trial. Experimental and clinical endocrinology & diabetes: official journal, German Society of Endocrinology [and] German Diabetes Association. 2018 Apr;126:222-8.
- 33. Mirfatahi M, Tabibi H, Nasrollahi A, Hedayati M. Effects of Flaxseed Oil on Serum Lipids and Lipoproteins in Hemodialysis Patients: a Randomized Controlled Trial. Iranian journal of kidney diseases. 2016 Nov;10:405-12.
- 34. Kontogianni MD, Vlassopoulos A, Gatzieva A, Farmaki AE, Katsiougiannis S, Panagiotakos DB, Kalogeropoulos N, Skopouli FN. Flaxseed oil does not affect inflammatory markers and lipid profile compared to olive oil, in young, healthy, normal weight adults. Metabolism: clinical and experimental. 2013 May;62:686-93.
- 35. Edel AL, Patenaude AF, Richard MN, Dibrov E, Austria JA, Aukema HM, Pierce GN, Aliani M. The effect of flaxseed dose on circulating concentrations of alpha-linolenic acid and secoisolariciresinol diglucoside derived enterolignans in young, healthy adults. European journal of nutrition. 2016 Mar;55:651-63.
- 36. Rizza S, Muniyappa R, Iantorno M, Kim JA, Chen H, Pullikotil P, Senese N, Tesauro M, Lauro D, et al. Citrus polyphenol hesperidin stimulates production of nitric oxide in endothelial cells while improving endothelial function and reducing inflammatory markers in patients with metabolic syndrome. The Journal of clinical endocrinology and metabolism. 2011 May;96:E782-92.
- 37. Akiyama S, Katsumata S, Suzuki K, Ishimi Y, Wu J, Uehara M. Dietary hesperidin exerts hypoglycemic and hypolipidemic effects in streptozotocin-induced marginal type 1 diabetic rats. Journal of clinical biochemistry and nutrition. 2010 Jan;46:87-92.
- 38. Li C, Schluesener H. Health-promoting effects of the citrus flavanone hesperidin. Critical reviews in food science and nutrition. 2017 Feb 11;57:613-31.
- 39. Barre D, Mizier-Barre K, Stelmach E, Hobson J, Griscti O, Rudiuk A, Muthuthevar D. Flaxseed lignan complex

- administration in older human type 2 diabetics manages central obesity and prothrombosis—an invitation to further investigation into polypharmacy reduction. Journal of nutrition and metabolism. 2012;2012.
- 40. Thakur G, Mitra A, Pal K, Rousseau D. Effect of flaxseed gum on reduction of blood glucose and cholesterol in type 2 diabetic patients. International journal of food sciences and nutrition. 2009;60:126-36.
- 41. Mohammadi-Sartang M, Sohrabi Z, Barati-Boldaji R, Raeisi-Dehkordi H, Mazloom Z. Flaxseed supplementation on glucose control and insulin sensitivity: a systematic review and meta-analysis of 25 randomized, placebo-controlled trials. Nutr Rev. 2018 Feb 1;76:125-39.
- 42. Albert BB, Derraik JG, Brennan CM, Biggs JB, Smith GC, Garg ML, Cameron-Smith D, Hofman PL, Cutfield WS. Higher omega-3 index is associated with increased insulin sensitivity and more favourable metabolic profile in middleaged overweight men. Scientific reports. 2014;4:6697.
- 43. González-Périz A, Horrillo R, Ferré N, Gronert K, Dong B, Morán-Salvador E, Titos E, Martínez-Clemente M, López-Parra M, Arroyo V. Obesity-induced insulin resistance and hepatic steatosis are alleviated by ω-3 fatty acids: a role for resolvins and protectins. The FASEB journal. 2009;23:1946-57.
- 44. Wien M, Rajaram S, Oda K, Sabaté J. Decreasing the Linoleic Acid to α- Linolenic Acid Diet Ratio Increases Eicosapentaenoic Acid in Erythrocytes in Adults. Lipids. 2010;45:683-92.
- Kuo S-M. The interplay between fiber and the intestinal microbiome in the inflammatory response. Advances in Nutrition. 2013;4:16-28.
- 46. Karlsson FH, Tremaroli V, Nookaew I, Bergström G, Behre CJ, Fagerberg B, Nielsen J, Bäckhed F. Gut metagenome in European women with normal, impaired and diabetic glucose control. Nature. 2013;498:99.
- 47. Sun Q, Wedick NM, Tworoger SS, Pan A, Townsend MK, Cassidy A, Franke AA, Rimm EB, Hu FB, van Dam RM. Urinary Excretion of Select Dietary Polyphenol Metabolites Is Associated with a Lower Risk of Type 2 Diabetes in Proximate but Not Remote Follow-Up in a Prospective Investigation in 2 Cohorts of US Women–3. The Journal of nutrition. 2015;145:1280-8.
- 48. Mulero J, Bernabé J, Cerdá B, García-Viguera C, Moreno DA, Albaladejo MD, Avilés F, Parra S, Abellán J, Zafrilla P. Variations on cardiovascular risk factors in metabolic syndrome after consume of a citrus-based juice. Clinical nutrition. 2012;31:372-7.
- 49. Ghanim H, Sia CL, Upadhyay M, Korzeniewski K, Viswanathan P, Abuaysheh S, Mohanty P, Dandona P. Orange juice neutralizes the proinflammatory effect of a high-fat, high-carbohydrate meal and prevents endotoxin increase and Toll-like receptor expression—. The American journal of clinical nutrition. 2010;91:940-9.
- 50. Medina- Remón A, Casas R, Tressserra- Rimbau A, Ros E, Martínez- González MA, Fitó M, Corella D, Salas- Salvadó J, Lamuela- Raventos RM, Estruch R. Polyphenol intake from a Mediterranean diet decreases inflammatory biomarkers related to atherosclerosis: a substudy of the PREDIMED trial. British journal of clinical pharmacology. 2017;83:114-28.
- Medina-Remón A, Tresserra-Rimbau A, Pons A, Tur JA, Martorell M, Ros E, Buil-Cosiales P, Sacanella E, Covas MI,

- Corella D. Effects of total dietary polyphenols on plasma nitric oxide and blood pressure in a high cardiovascular risk cohort. The PREDIMED randomized trial. Nutrition, Metabolism and Cardiovascular Diseases. 2015;25:60-7.
- 52. Ursoniu S, Sahebkar A, Andrica F, Serban C, Banach M. Effects of flaxseed supplements on blood pressure: A systematic review and meta-analysis of controlled clinical trial. Clinical nutrition (Edinburgh, Scotland). 2016 Jun;35:615-25.
- 53. Sleight P, Yusuf S, Pogue J, Tsuyuki R, Diaz R, Probstfield J, Investigators HOPES. Blood-pressure reduction and cardiovascular risk in HOPE study. The Lancet. 2001;358:2130-1.
- 54. Prasad K. Secoisolariciresinol diglucoside (SDG) isolated from flaxseed, an alternative to ACE inhibitors in the treatment of hypertension. The International journal of angiology: official publication of the International College of Angiology, Inc. 2013;22:235.
- Caligiuri SP, Edel AL, Aliani M, Pierce GN. Flaxseed for hypertension: implications for blood pressure regulation. Current hypertension reports. 2014;16:499.
- 56. Austria JA, Richard MN, Chahine MN, Edel AL, Malcolmson LJ, Dupasquier CM, Pierce GN. Bioavailability of alpha-linolenic acid in subjects after ingestion of three different forms of flaxseed. Journal of the American College of Nutrition. 2008;27:214-21.
- 57. Kuijsten A, Arts IC, van't Veer P, Hollman PC. The relative bioavailability of enterolignans in humans is enhanced by milling and crushing of flaxseed. The Journal of nutrition. 2005;135:2812-6.
- 58. Yari Z, Rahimlou M, Eslamparast T, Ebrahimi-Daryani N, Poustchi H, Hekmatdoost A. Flaxseed supplementation in non-alcoholic fatty liver disease: a pilot randomized, open labeled, controlled study. International journal of food sciences and nutrition. 2016;67:461-9.

- 59. Mohammadi- Sartang M, Mazloom Z, Raeisi- Dehkordi H, Barati- Boldaji R, Bellissimo N, Totosy de Zepetnek J. The effect of flaxseed supplementation on body weight and body composition: a systematic review and meta- analysis of 45 randomized placebo- controlled trials. Obesity Reviews. 2017;18:1096-107.
- 60. Garaulet M, Hernández-Morante J, Tébar F, Zamora S. Anthropometric indexes for visceral fat estimation in overweight/obese women attending to age and menopausal status. Journal of physiology and biochemistry. 2006;62:245-52
- 61. Buckley JD, Howe P. Anti- obesity effects of long- chain omega- 3 polyunsaturated fatty acids. Obesity reviews. 2009;10:648-59.
- 62. Buckley JD, Howe PR. Long-chain omega-3 polyunsaturated fatty acids may be beneficial for reducing obesity—a review. Nutrients. 2010;2:1212-30.
- 63. Setchell K, Adlercreutz H. Mammalian Lignans and Phytooestrogens Recent Studies on their Formation, Metabolism and Biological Role in Health and. Role of the gut flora in toxicity and cancer. 1988;315.
- 64. Morisset A-S, Lemieux S, Veilleux A, Bergeron J, Weisnagel SJ, Tchernof A. Impact of a lignan-rich diet on adiposity and insulin sensitivity in post-menopausal women. British journal of nutrition. 2009;102:195-200.
- 65. Alam MA, Subhan N, Rahman MM, Uddin SJ, Reza HM, Sarker SD. Effect of citrus flavonoids, naringin and naringenin, on metabolic syndrome and their mechanisms of action. Advances in Nutrition. 2014;5:404-17.
- 66. Andriantsitohaina R, Auger C, Chataigneau T, Étienne-Selloum N, Li H, Martínez MC, Schini-Kerth VB, Laher I. Molecular mechanisms of the cardiovascular protective effects of polyphenols. British Journal of Nutrition. 2012;108:1532-49.

Nutrition and Food Sciences Research 20 Vol 11, No 3, Jul-Sep 2024