# The effect of a 10-week high-intensity interval training and ginger consumption on inflammatory indices contributing to atherosclerosis in overweight women

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**Background:** Most of the cardiovascular diseases can be prevented by doing regular physical exercises and using herbal supplements. The present study is aimed at assessing ginger supplement and high-intensity interval training (HIIT) on inflammatory indices contributing to atherosclerosis in overweight women. **Materials and Methods:** The present study is a randomized, experimental, and controlled one in which thirty healthy overweight women aged 20–30 years were randomly divided into three equal groups, namely, ginger, ginger + HIIT, and placebo + HIIT. The training groups performed high-intensity interval exercises (i.e. 40-m maximal shuttle run) for ten consecutive weeks. The supplement groups daily took 3 g of ginger pills and the third group took placebo. **Results:** Paired t-test revealed a significant decrease in the density of type 1 monocytes chemo tactic protein (MCP-1) in HIIT + ginger (P = 0.026) and HIIT + placebo (P = 0.001) groups. Besides, maximum aerobic capacity in the two training groups significantly increased P = 0.002 and P = 0.000, respectively. In spite of this, analysis of variance showed no significant differences in three groups regarding the three indices such as intercellular adhesion molecule-1 (ICAM-1) (P = 0.093), MCP-1(P = 0.075), and serum interleukin-10 (IL-10) (P = 0.164). **Conclusion:** A 10-week intensive interval exercise, by itself or together with ginger supplement, improved MCP-1 and maximum oxygen consumption in overweight women, without any significant effect on soluble ICAM-1 and IL-10. These findings indicate the relative and efficient role of HIIT in overweight women without the necessity to combine with ginger as an antioxidant/anti-inflammatory supplement.

**Key words:** Ginger, high-intensity interval exercise, intercellular adhesion molecule-1, interleukin-10, monocyte chemotactic protein-1, overweight

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#### INTRODUCTION

Inflammatory and anti-inflammatory conditions contributing to atherosclerosis are assessable through studying circulatory biological indices including interleukins (ILs), attracting molecules, and chemokines. Intercellular adhesion molecules-1 (ICAM-1), monocytes chemo tactic protein-1 (MCP-1), and IL-10 are among independent and significant inflammatory and

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anti-inflammatory indices which, directly or indirectly, contribute to atherosclerosis process. [1] Different approaches including exercise training and antioxidant supplementation have been used to reduce inflammatory cytokines. [2,3] Among physical activity programs, high-intensity interval training (HIIT) has gained utmost importance and popularity in 2014. HIIT includes short-term intensive activities (6 s−4 min) with high intensity (≥90% of maximum aerobic capacity) and active or passive short-term recovery intervals. [4] The influence of HIIT on inflammatory markers shows contradictory

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results in literature, while some indicate attenuation of inflammation and<sup>[5,6]</sup> others show no change.<sup>[3,5]</sup> It has been shown that 2 weeks of HIIT improved MCP-1 and oxygen consumption to a maximum level, without any change in serum IL-10 and ICAM-1 in obese and overweight men.<sup>[5]</sup> It has been shown that the main ginger elements such as gingerols (gingerol-6, -8, and -10) and shogaols (shogaol-6 and -8) reduced MCP-1 and ICAM-1 mRNA in different cells by the inhibition of nuclear factor-κB (NF-κB) activation in response to inflammatory stimuli<sup>[7]</sup> while there are many reasons supporting the effectiveness of ginger on inflammatory indices; a few studies have reported contradictory results.[8,9] Although there are some evidences regarding the simultaneous implementation of ginger intake and physical activity on inflammatory indices, the outcomes are not in agreement.[10-12] Atashak et al. observed no change in levels of lipid profile after 10-week of progressive resistance training, together with ginger supplement.[13] In contrast, Ayaz and Roshan demonstrated reduced plasmatic levels of IL-6 and C-reactive protein following physical exercise together with ginger supplement.[14]

However, the effect of ginger which has a potent anti-inflammatory effect has not been evaluated in HIIT, exclusively on ICAM-1, MCP-1, and IL-10 markers. Therefore, the present study aimed at answering the question whether or not high HIIT solely or in combination with ginger supplement decrease chronic inflammation.

# **MATERIALS AND METHODS**

# Study population

This randomized placebo-controlled study was conducted at Birjand University. The study protocol was approved by the Medical Ethics Committee of Birjand University of Medical Sciences. The study protocol of the current study is registered with the Iranian Registry of Clinical Trials (www. irct.ir; IRCT2016051027836N1). We included thirty female students aged between 20 and 30 years with overweight, according to Morgan table. All the participants were randomly assigned to three equal study groups (n = 10), namely HIIT + ginger, HIIT + placebo, and ginger, using a computer-based random digit generator using the

admission numbers. The female students were excluded from the study if they had cardiovascular diseases (CVDs), hypertension, diabetes, smoking, pregnancy, and regular physical activity 6 months before the research plan. Demographic data of the participants, determining their health statuses and acquiring written consent form, were obtained before starting the program.

# Intervention

The exercise protocol was taken from 40-m maximal shuttle run, which is a valid test for the assessment of anaerobic function. During this activity, each participant totally ran and returned a 20 m route at her maximum speed in 30 s.<sup>[15]</sup> The exercise protocol was performed three times per week for 10 weeks as shown in Table 1. The exercise was conducted at 90% maximum heart rate (age - 220) intensity which was controlled by a Telemetry (Polar, Finland). According to specific doses in previous studies,<sup>[16]</sup> participants in ginger and exercise + ginger groups daily received 3000 mg ginger tablets (produced by Iran Dineh Co., Tehran, Iran), whereas the exercise + placebo group received Nokhodchi flour tablets (produced by Iran Dineh Co., Tehran, Iran) in the same manner, for 10 weeks, 7 days/week, 30 min before each meal.

# Study protocol

Baecke habitual physical activity questionnaire was used<sup>[17]</sup> initially. A week before primary measurement of variables, the participants weights were assessed by laboratory scale with an accuracy of <100 g (TCM, China) in a status without shoes and minimal clothes. Their heights were assessed by meter with an accuracy of <0.5 cm. Skin folds were measured using Caliper (EIYOKEN Type, Yagami, PAT 376843, Japan) according to Pollock three sites methodology, then percent body fat (PBF) was calculated from appropriate formula.<sup>[18]</sup> Each participant performed Bruce maximum test using a treadmill (T 150 Cos10199 model, h-p-cosmos Company). Maximum oxygen consumption (VO<sub>2 max</sub>) was calculated using an extended form of the formula;<sup>[19]</sup> then, we have

 $VO_{2}$  = (4.38 × total exercised time) – 3.9

The participants were asked to maintain their habitual diet during the protocol and refrain from consuming any

Table 1: High-intensity interval exercise protocol							
Weeks	Activity time (s)	Active relaxation time (s)	Intervals	Protocol length of time (min)	Total activity time (main activity, warming up, and cooling down) (min)		
40 m shuttle run protocol							
The first and the second	30	30	4	4	24		
The third and the fourth	30	30	5	5	25		
The fifth and the sixth	30	30	6	6	26		
The seventh and the eighth	30	30	7	7	27		
The ninth and the tenth	30	30	8	8	28		

antioxidant supplements, extra ginger, varying ordinary diet and ginger dose. Their 3 consecutive days diet, including a 1-day holiday and 2 working days in the 1st and last week of exercise program, was controlled through a questionnaire. Results showed no significant difference (HIIT + ginger:  $1221\pm40.93$  kcal,  $141.19\pm5.22$  g carbohydrate,  $56.68\pm0.90$  g fat, and  $51.29\pm0.63$  g protein; HIIT + placebo:  $1209\pm40.93$  cal,  $132.60\pm5.22$  g carbohydrate,  $55.37\pm0.90$  g fat, and  $52.38\pm0.63$  g protein; ginger:  $1275\pm40.93$  cal,  $134.72\pm5.22$  g carbohydrate,  $56.89\pm0.90$  g fat, and  $52.47\pm0.63$  g protein; energy P=0.24, carbohydrate P=0.91, fat P=0.67, protein P=0.83), between the three groups regarding macronutrient and calorie intake.

Moreover, calorie intake from the last meal, before testing, was balanced to 500–600 cal for all participants. At the end of the program, two cases from each group were eliminated due to irregular physical activity, taking hormonal tablets, traveling, and pregnancy. Finally, only 24 individuals remained [Figure 1].

# Sample collection and biochemical determination

Venus blood samples (10 cc) were withdrawn from all the participants 24 h before the 1st and 48 h after the last training session, whereas the participants were in their follicular phase of menstruation and fasting for 12 h. Then, serum was stored at -80°C for the following analysis. We used 96-well enzyme-linked immunosorbent assay kits to measure the levels of ICAM 1 (Ka Za Bayoi, China, sensitivity < 15.6 pg/ml, intraassay CV < 8%), MCP 1 (Ka Za Bayoi, China, sensitivity < 30.184 pg/ml, intraassay CV < 8%), and IL 10 (Diaclone SAS, France, sensitivity < 4.9 pg/ml, intraassay CV

< 3.5%) in the serum. The assay was performed according to the manufacturers' instructions.

#### Statistical analysis

Data were analyzed using Statistical Package for Social Sciences (SPSS Inc., Chicago, IL, USA) software version 16.0 and were expressed as mean ± standard deviation or median (25th, 75th) percentiles. Differences between the baseline means of the three groups in all variables were analyzed using one-way analysis of variance (ANOVA). After determination of normality and homogeneity of variances by Kolmogorov-Smirnov test and the Levene's test, respectively, the between-group differences were examined using either a one-way ANOVA for normally distributed data, or Kruskal-Wallis test for nonnormal distributed data (MCP-1, IL-10, and VO<sub>2</sub>). Then, to locate differences, Tukey's post hoc test was used (P < 0.05) in the next step. Within-group differences were determined using either paired t-test for normally distributed data and Wilcoxon signed-rank test for nonnormal distributed data (MCP-1, IL-10, and  $VO_{2}$ ). A two-sided P < 0.05 was considered statistically significant.

#### RESULTS

Mean demographic characteristics of three groups are summarized in Table 2. There was no significant difference between three groups regarding the baseline characteristics [Tables 1 and 2]. The Kruskal–Wallis test showed  $VO_{2_{max}}$  changes (pre- to post-test) were significantly different between groups (P < 0.001). Tukey's post hoc test revealed

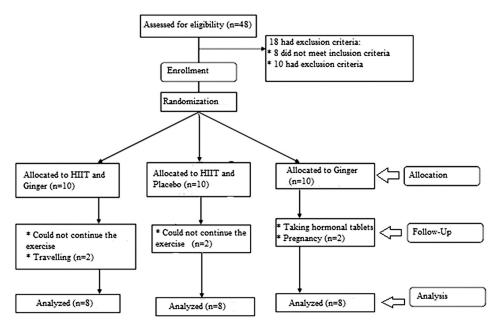


Figure 1: CONSORT flow diagram of the study

Table 2: Baseline comparison of demographic characteristics

Variables	Exercise +	Exercise +	Ginger	P
	ginger (n=8)	placebo (n=8)	( <i>n</i> =8)	(ANOVA)
Age (year)	21.88±3.40	22.38±3.24	21.63±1.77	0.87
Height (cm)	161±0.07	160.05±0.09	159±0.06	0.94
Weight (kg)	74.19±11.53	72.24±6.86	64.91±3.60	0.07
BMI (kg/m²)	28.68±2.60	28.42±2.40	26.06±1.69	0.06

Data as mean±SD. BMI=Body mass index; ANOVA=Analysis of variance; SD=Standard deviation

increased  $VO_{2_{max}}$  in both "exercise + ginger" (P < 0.000) and "exercise + placebo" (P < 0.000) groups, in comparison to "ginger" group. While no significant between-group differences in MCP-1, ICAM-1, and IL-10 changes were determined (P = 0.178, P = 0.07, and P = 0.14; respectively), and the results are summarized in Table 3. On the other hand, the results of Wilcoxon signed-rank test showed significant increase in  $VO_{2_{max}}$ ; P = 0.012 and P = 0.012, and a significant decrease in MCP-1 concentration; P = 0.025 and P = 0.012 in "exercise + ginger" and "exercise + placebo," respectively, after 10 weeks of intervention [Table 3].

# **DISCUSSION**

Although popular suggestions regarding health improvement recommend moderate-intensity physical activity on most preferably all days, today experts are more concerned with high-intensity interval exercises, either in healthy participants or cardiovascular patients. [20,21] ICAM-1, MCP-1, and IL-10 act as primary markers in the first stages of progression, adhesion, and attraction of monocytes to intima and involve in systemic inflammation.[1] In the present study, we found that although 10 weeks of HIIT, by itself or in combination with ginger supplement, resulted in improved MCP-1 and VO<sub>2</sub> in overweight women, it did not affect significantly serum-soluble ICAM (sICAM)-1 and IL-10. Regarding the effectiveness of intensive interval exercise and ginger consumption, our findings were consistent with other studies that showed a significant reduction in circulating MCP-1 in 12 overweight men following 2 weeks of HIIT on ergometer with 89.5% VO<sub>2</sub> and lack of a significant change in serum IL-10 and ICAM-1.[5] In addition, Bartlett et al. showed that 10 weeks of HIIT on ergometer reduced serum MCP-1 level in inactive 20–60 years participants. [6] Reduction in MCP-1 might be as a result of decrease in PBF. In contrast, Ahmadizad et al. did not observe any change in young men's inflammatory indices after 6 weeks of HIIT.[3]

The short length of training in the mentioned study may be the relevant cause of discrepancy.

Very few studies have dealt with the combined effect of exercises and ginger consumption on inflammatory indices. Black *et al.* and Mashhadi *et al.* did not find a combined effect of ginger and physical activity on inflammatory indices, <sup>[11,12]</sup> in contrast to the studies of Atashak *et al.* and Ayaz and Roshan that mentioned decreased values of inflammatory markers following physical exercises together with ginger consumption. <sup>[13,14]</sup>

Inflammatory indices such as ICAM-1, MCP-1, and IL-10 play a significant role in CVDs.[1] IL-10 exerts its anti-inflammatory effect on ICAM-1 through influence on NF-κB factor activation, and as a result, the ICAM-1 gene expression decreases.[22] In the current study, a significant decrease of IL-10 in the exercise + placebo group was associated with the significant increase of ICAM-1 in the same group. Perhaps one can claim that in "exercise + ginger" and "ginger" groups, consumption of ginger (gingerol and shogaol) for 10 weeks has had decisive anti-inflammatory effects on nuclear transcription factor<sup>[23]</sup> resulting in insignificant reduction of ICAM-1 expression. A few studies, however, have reported different results.[24,25] In these studies, participants were suffering from different disorders and pretest ICAM-1 values were two to three times more than present participants. While the participants in the current study were healthy and probably, the effects of exercise and ginger have little been observed.

It has been said that the anti-inflammatory properties of IL-10 relate to its capacity to reduce the production of pro-inflammatory cytokines (such as IL-1  $\beta$ , tumor necrosis factor- $\alpha$ , and MCP-1) from monocytes through decreasing NF- $\kappa$ B activity and increasing interleukin -1 receptor antagonist production. However, in the current study, decrease in MCP-1 was not parallel to IL-10 increase; perhaps this mechanism was not so effective.

Of course, some scholars hold that MCP-1 as potentially independent diagnostic index regarding CVDs.[26,27] The other aspect of MCP-1 to carry out its pro-inflammatory role is the fatty tissue, which has been reported. [5] MCP-1 is secreted from intestinal fatty tissue and attracts an increasing number of macrophages to the fatty tissue, thus, organizes the inflammatory process.<sup>[5]</sup> MCP-1 reduction in the "exercise + ginger group" in the present study might be due to insignificant decrease of waist: Hip girth ratio and significant decrease of body fat. Regarding the relationship between aerobic fitness and improvement of inflammatory indices to lower CVDs, in the current study, significant improvement in  $VO_{2_{max}}$ , probably related to the improvement of MCP-1 in the training groups. Frequent touching of legs with the ground and producing stress-fracture in legs because of brisk running is associated with muscular injuries and inflammation. Present exercise protocol was a form of intense-shuttle running, which is a reason for the insignificant reduction in ICAM-1 value.

Table 3: Comparison of inflammatory indices of the participants in the three studied groups after a 10-week exercise and taking ginger supplement

Variables	Groups	Measurement time		Within	Scores difference	Between group (P)
		Pretest	Posttest	group ( <i>P</i> )		
sICAM-1 (ng/ml)	Exercise + ginger (n=8)	9028.22±2452.08	7287.86±1156.44	0.14	1740.36±3030.80	0.093
	Exercise + placebo (n=8)	7578.41±540.31	8785.15±643.91	0.015*	1206.73±1067.12	
	Ginger (n=8)	7018.11±1531.66 <i>P</i> =0.07	6727.82±2246.18	0.79	-2902.94±3045.20	
MCP-1# (ng/ml)	Exercise + ginger (n=8)	86.65 (52.25, 135.63)	49.56 (25.05, 66.60)	0.025*	-42.43 (-99.82, -18.82)	0.178
	Exercise + placebo (n=8)	57.51 (35.51, 64.65)	27.80 (23.99, 43,83)	0.012*	-28.13 (-30.17, -11,68)	
	Ginger (n=8)	54.34 (43.88, 90.21) <i>P</i> =0.058	49.56 (40.00, 66.30)	0.20	-12.55 (-34.07, 17.03)	
IL-10# (ng/ml)	Exercise + ginger (n=8)	3.76 (2.46, 4.53)	2.11 (1.51, 3.89)	0.20	-0.78 (-3.29, 1.21)	0.14
	Exercise + placebo (n=8)	3.76 (2.55, 8.24)	3.02 (1.72, 5.34)	0.036*	-1.64 (-2.80, -0.39)	
	Ginger ( <i>n</i> =8)	2.16 (1.81, 2.42) <i>P</i> =0.06	2.46 (0.98, 3.37)	0.77	0.08 (-1.30, 1.51)	
$Vo_{2_{max}}^{ \#}$ (ml/kg/	Exercise + ginger (n=8)	34.64 (29.73, 39.30)	47.47 (44.31, 49.18)	0.012*	11.36 (6.73, 19.27)	0.001†
min)	Exercise + placebo (n=8)	33.38 (31.94, 36.80)	48.85 (41.12, 49.75)	0.012*	12.48 (8.91, 19.12)	
	Ginger ( <i>n</i> =8)	39.62 (37.98, 40.42) <i>P</i> =0.057	39.43 (37.69, 40.73)	0.86	0.34 (-1.38, 0.80)	
Body fat (%)	Exercise + ginger (n=8)	52.89±4.55	46.21±4.50	0.005*	6.67±4.76	0.02†
	Exercise + placebo (n=8)	55.50±1.38	41.30±3.01	0.19	4.20±8.29	
	Ginger (n=8)	41.61±5.90 <i>P</i> =0.11	44.25±1.87	0.19	2.63±5.17	
Waist:hip girth ratio	Exercise + ginger	0.81±0.05	0.80±0.05	0.30	0.006±0.01	0.29
	Exercise + placebo	0.78±0.02	0.78±0.02	0.5	0.003±0.01	
	Ginger	0.78±0.03 <i>P</i> =0.33	0.77±0.04	0.18	0.006±0.01	

\*P<0.05 significant difference compared to pretest. †P<0.05 significant difference between three groups. Within Group P: Paired t-test or Wilcoxon signed-rank test. Between Group P: One-way ANOVA or Kruskal-Wallis test. \*Variables marked with were presented as medians (25\*\*, 75\*\*) percentiles and all others were presented as means±SD. sICAM-1=Soluble Intercellular adhesion molecule-1; MCP-1=Monocyte chemotactic protein-1; IL-10=Interleukin-10; Vo<sub>2max</sub> =Maximal oxygen consumption; ANOVA=Analysis of variance; SD=Standard deviation

An example of this phenomenon is seen in the study by Akimoto *et al.*, which assessed the effect of different exercises on sICAM-1 in healthy men. Lack of change in serum ICAM-1 levels after an ergometer exercise, while increased values following endurance running have been observed. They suggested that exercises associated with muscular injuries and inflammation increased the plasma concentration of sICAM-1.<sup>[28]</sup> However, it is certain that quality and intensity of exercising stimulus, individuals' genetic flexibility, various exercise protocols, and other factors of lifestyle influence the interpretation of the obtained results.<sup>[29]</sup>

The significant increase observed in training groups,  $VO_{2_{max}}$  compared to the "ginger group" is a reason supporting the

beneficial effects of HIIT on exercise performance and recovering from CVDs. Butcher reported the effectiveness of such exercises on  $\mathrm{VO}_2$  of the old and youth through exercising for 2–15 weeks. [4] Improved stroke volume relates to increased heart muscle contraction capacity during maximum pressure and increase in mitochondria biogenesis which are the main possible mechanisms of  $\mathrm{VO}_2$  advance as a result of HIIT protocols. [4] The main characteristic of interval exercises, which makes it more valuable than continuous ones, is their more energy outcome of peripheral muscles without interfering with cardiopulmonary increasing capacity. The recovery intervals between intensive activity intervals, while reforming phosphocreatine and myoglobin stores influences the blood lactate concentration through delectating it. [7] Therefore, interval

exercises compared with continuous ones with the same volume can impose maximum pressure on peripheral muscles and oxygen delivery organs without employing anaerobic metabolism and lactic acid concentration. Increased  $\mathrm{VO}_{2_{\mathrm{max}}}$  is of value because weak  $\mathrm{VO}_{2_{\mathrm{max}}}$  predisposes individuals to cardiovascular mortality. $^{[30]}$ 

# **CONCLUSIONS**

A 10-week intensive interval exercise, by itself or together with ginger supplement, improved MCP-1 and  $\mathrm{VO}_{2_{\mathrm{max}}}$  in overweight women; without any significant effect on sICAM-1 and IL-10. These findings indicate the relative and efficient role of HIIT in overweight women without the necessity to combine with ginger as an antioxidant/anti-inflammatory supplement. Since the present study lacked a "placebo group" and the participants' diets were not accurately controlled during the research, further investigations are required.

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#### Conflicts of interest

The authors have no conflicts of interest.

# **AUTHORS' CONTRIBUTION**

SN contributed in the conception of the work, conducting the study, revising the draft, approving the final version of the manuscript, and agreed for all aspects of the work data and final approval of the version to be published. MEA and TK contributed in the conception of the work, conducting the study, revising the draft, approving the final version of the manuscript, and agreed for all aspects of the work data. SHA contributed in biochemical analyzing, approval of the final version of the manuscript, and agreed for all aspects of the work data. MM contributed in the conception of the work, consulting and conducting the study and agreed for all aspects of the work data.

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