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Original Research

The Effect of Aerobic Training and Supplementation of Curcumin on the Glycemic Markers and Level of Myeloperoxidase Enzyme of Overweight Women

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Abstract

Background and Aims: Diabetes and cardiovascular disease are the most common diseases in the world. Diabetes has many complications and heart disease causes deaths worldwide. This study was aimed to investigate the effect of curcumin supplementation alone and with aerobic training on Body Mass Index (BMI), insulin resistance and myeloperoxidase (MPO) level in overweight women. **Material and Method:** In this randomized double-blind placebo-controlled trial, forty healthy sedentary overweight females aged 30–45 years and with a BMI 25–30 kg/m^2, were assigned to four groups of ten subjects each: "Curcumin supplementation (Cur)", "placebo (Pla)", "Cur+aerobic training (Tra)" and "Pla+Tra". The aerobic training program comprised 3 days per week and Curcumin or placebo (500 mg/day) was administered in Cur and Cur+Tra groups in 8 weeks. Before and after intervention, BMI, HOMA-IR (Homeostatic Model Assessment-Insulin Resistance) and MPO enzyme were measured. ANOVA and ANCOVA was used to assess differences between the groups. **Results:** The within-group analysis showed no significant difference in BMI, HOMA-IR and MPO in four groups ($p>0.05$). Aerobic training and curcumin supplementation had no significant effect on BMI, HOMA-IR, and MPO levels in overweight middle-aged women ($p>0.05$). **Conclusions:** These findings suggest the combination of curcumin supplementation with aerobic training has no significant effect on BMI, HOMA-IR, and MPO levels in overweight middle-aged women. Further studies with different doses and longer interventions are needed to clarify the results.

Keywords: Aerobic Training, Curcumin, Diabetes mellitus, Peroxidase.

Background and Aims

In recent decades, due to inactive and sedentary lifestyles, cardiovascular disease (CVD) has increased [1, 2]. CVD is one of the most important causes of mortality, worldwide [3]. Although CVD mortality has reduced in high-income countries, it is rising in low and lower-middle-income countries [3].

The Nurses' Health Study (NHS) cohort study found that most women gain weight through middle adulthood. The women enrolled in the study were selected as a homogeneous population, predominantly caucasian, with high socioeconomic status and having a nurse occupation [4]. Other studies showed the prevalence of obesity among women more than in men, in developing countries [57]. This

condition has an important effect on health impairment, many diseases, and mortality [4, 8, 9]. Previous studies have shown the effect of overweight and obesity on insulin resistance index [10]. Insulin resistance has been associated with metabolic problems and some diseases [5, 11, 12]. The result of one study showed that aerobic training of eight weeks reduced insulin resistance in obese women [13]. Therefore, aerobic training is a good measure to control and improve the risk factors of insulin resistance such as diabetes and CVD [13].

Also, the results of some studies have shown prolonged or short-duration high intensity exercise causes increase of radical production, oxidative stress, and damage to body cells in active skeletal muscles [14]. Exercise-induced oxidative stress increases the production of free radicals. These free radicals are highly reactive and have short half-lives therefore, are difficult to measure [15]. As a result, the oxidative stress markers are used to measure oxidative stress. These markers indicate increase in free radicals [15]. One of these markers is myeloperoxidase (MPO). MPO is a myeloid-lineage restricted enzyme that is largely expressed by neutrophils and found in the azurophilic granules. According to the results of some studies, this marker is also produced in pathological conditions from monocytes and macrophages [16]. MPO has powerful antibacterial properties and defends humans against various pathogens by phagocytosis. MPO employs H_2O_2 to produce hypochlorous acid (HClO) and other reactive parts, which destroy pathogens during infections. In contrast, many studies have shown elevated blood MPO level is associated with inflammation, CVD, neurodegenerative, autoimmune, and cancer diseases [16–19]. In addition, some studies have also shown that intense physical activity can cause degranulation of neutrophils, leading to an increased blood MPO concentration. Exercise-induced muscle damage is associated with the activation of leukocytes and onsets an inflammatory response. Some studies have shown oxidative stress following intensive training [20].

In recent years, many researchers have paid attention to the use of spices on the prevention and treatment of various diseases [21]. Curcuminoids are one of the most important anti-inflammatory agents in nature. It is found in turmeric rhizomes, of *Curcuma longa* L. [21, 22]. Yellow pigments of curcuminoides are its main active components [21]. It has many therapeutic effects in various diseases including chronic inflammatory diseases, obesity, depression, and fatigue [21]. In metabolic syndrome, curcumin can decrease leptin concentrations [23]. It is a bioactive polyphenol found in turmeric rhizomes, of *Curcuma longa* L., (Zingiberaceae), and has multiple pharmacological actions [21, 22]. Yellow pigments of curcuminoides including 3–5% of turmeric composed of curcumin, demethoxy curcumin, and bisdemethoxy curcumin are main active components [21]. There is an evidence that curcumin is an anti-inflammatory agent, so, it has many therapeutic effects in various diseases including chronic inflammatory diseases of Alzheimer's, Parkinson's, multiple sclerosis, brain injuries, CVD, types of cancer, allergies, asthma, arthritis, obesity, depression, and fatigue [21]. In metabolic syndrome, curcumin can decrease Body Mass Index (BMI), weight, waist circumference and leptin concentrations [23].

A study showed that regular consumption of curcumin with regular aerobic training improves endothelial function significantly [24]. This research has shown 150 mg curcumin supplementation, daily, with aerobic training, for eight weeks improves endothelial function in healthy postmenopausal women [24]. In a study of the effect of endurance training with curcumin intake on the hemodynamics of the central arteries of healthy postmenopausal women, it was concluded that daily intake of 150 mg curcumin in combination with aerobic training, three to six sessions per week for eight weeks, reduced BMI significantly [25]. Evaluation of the effect of curcumin on rheumatoid arthritis patients showed that daily consumption of 500 mg curcumin led to inflammatory index of C-reactive protein [26].

Most studies have investigated the effects of different training or curcumin supplementation, separately on myeloperoxidase levels. No study has found a combination of them in overweight middle-aged women. Therefore, this study aims to answer the question whether

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aerobic training and curcumin supplementation have an effect on myeloperoxidase enzyme levels and body composition in overweight middle-aged women.

Material and Method

Study design and patients

Forty healthy sedentary women (less than 30 minutes a day of moderate intensity physical activity for three days a week [27], overweight, and aged 30–45 years) participated in this study who were referred to the comprehensive health center or other health centers of Tehran University of Medical Sciences, located in Yaft-Abad district. Participants were recruited only if they met the following criteria: 24.9<BMI<29.9 Kg/m^2, no allergy to curcumin, no pregnancy, non-smoking, no tobacco and hookah consumption, no following a special diet, no underlying or infectious disease, no known history of high blood lipid without medication, cardiac failure, taking anti-inflammatory drugs, severe endocrine or metabolic disorders, hospitalization for any reason, and no intake of dietary supplements and herbal tea. All volunteers completed a self-administered questionnaire on demographic, physical activity, exercise, dietary intake, lifestyle information, and health history prior to the study. While none of the study participants were trained in physical activity or aerobic exercises of any form and type in the past 6 months.

In this method a simple randomizing model like drawing of lots was employed. In this case, the sample was categorized in control and intervention group according to the result of lots. The Fleiss equation for sample volume estimation was used in this survey in order to approximate the volume of sample; in which the power of experiment, alpha of equation and the average alteration were 0.8, 0.05 and 5, respectively. According to the conducted estimation, the sample size was calculated 8.81. To be more cautious, among the overweight volunteer women ten persons were chosen as the experimental group.

A double-blind, placebo-controlled, clinical trial was used and performed from January to March 2018. The participants randomly underwent, in one of laboratory-based trials, in four randomized orders by a list prepared with a random number generator in four groups of ten: (1) placebo supplementation (Pla), (2) curcumin supplementation (Cur), (3) curcumin supplementation besides aerobic training (Cur+Tra), and (4) placebo supplementation besides aerobic training (Pla+Tra). The Cur and Tra groups, took two curcumin capsules in a day (500 mg/day) and the Pla and Pla+Tra groups received a placebo every day, for eight weeks.

This study was approved by the Ethical Committee at the *Azad University - Tehran Branch* and registered in the Iranian Registry of Clinical Trials (NO: *IR.SSRI.REC.1397.209*). Written informed consent was obtained from all participants at the beginning of the study.

Laboratory, anthropometric and clinical data collection

Height and weight were measured using portable measuring rod and electronic digital scales (Beurer BG55 Digital Scale, RU), accurate to 0.1 kg, respectively. Anthropometric indexes were measured by one observer. Subjects were in light clothing and no shoes and height was measured in the standing position. BMI was calculated as body weight (kg) divided by squared height (m^2). Then, subjects were categorized as underweight, normal weight, overweight, or obese according to the international classification used by the WHO [28].

Venous blood samples were taken from the left vein in the sitting position in the early morning after overnight fasting. For measuring serum blood markers, samples were centrifuged. The glucose oxidase method (Using the Spanish Biosystems glucose measurement kit) Fasting blood insulin levels were estimated by Enzyme-Linked Fluorescence Assay (ELFA) method (TOSOH kit, Japan) in all the study participants. The insulin resistance index was calculated by the following formulas: HOMA-IR=Fast insulin (μU/ml) ×Fast blood glucose (mmol/L)/22.5. Plasma MPO concentration was determined by immunoassay using a commercial kit

(Immunology Consultants, Laboratory, Inc., Newberg, USA). Intra-assay and assay-inter values were 2.8% and 6.5%, respectively. The separation and recovery limit of this assay was 0.121 ng/ml and 86.7%, respectively.

Turmeric powder was provided by the reputable brand of Golha Company (Iran) in supervision of Analysis center of the institute of medicinal plants. 5000 g of the powder was poured into 5000 ml 80°C alcohol. The Percolation method was used for the extraction of alcohol. After 24 hours, the contents were smoothed using a Buchner funnel and ordinary filter paper. The extraction efficiency was 10%. The powder was dispersed in a suitable solvent (80% ethanol) to prepare the drug form. After expanding, breadcrumbs were used as expanders and fillers to achieve proper fluidity. Based on previous clinical studies, an effective dose of 250 mg of the ethanolic extract is estimated to be 80%. Total curcuminoids have been estimated at 75–85% in previous studies [29]. Therefore, 250 mg capsules (00 short stays manufactured by Iran Gelatin Company) were prepared with a semi-automatic encapsulating machine. The Pla and Pla+Tra groups received two placebo capsules per day, which were similar in shape and color and made of an equivalent dose of starch. The study duration was eight weeks and all subjects were instructed not to alter their dietary habits during this period.

Participants in the Cur+Tra and Pla+Tra groups underwent aerobic training (three days per week for eight weeks) in a gym. The aerobic capacity of the subjects was examined by a 6-minute Rock-Port test. The maximum heart rate for each person was estimated as equal to 208–0.7×(age) [30]. In the training session, first, the subjects did warm up stretching exercises for 6 minutes, then were running continuously on the treadmill, at a speed of 7 km/h, by heart rate 50–80% of maximum, for 20 minutes. Their heart rate was monitored by a polarimeter (Polar: Finland, within a 5% error rate of a heartbeat) during running, to keep it in the specified intensity. This intensity was individualized. Every person had certain intensity. The Youngers run with higher intensity and the older ones run with lower intensity. In each of the next sessions, 90 seconds were added to the running time. The training intensity was controlled using a belt heart rate sensor (polar beat). Subjects in the Cur and Pla groups were instructed not to change their level of physical activity.

Statistical analysis

The mean±SD were used to describe the results. The Kolmogorov-Smirnov test was used to determine the distribution of data. Covariance test was used to determine the effectiveness of independent variables on the dependent variables. 95% confidence level was considered and p-value was considered significant at 0.05 level. Statistical analysis was performed using SPSS 22 software (SPSS Inc., USA).

Results

The mean±SD of age, weight, height, BMI, HOMA-IR, and MPO of subjects in the pre-test are presents in Table 1, and Pre-test and post-test values of the studied variables in the subjects have been shown in table 2. The Kolmogorov-Smirnov test was used to check for normality of distribution of all variables. The distribution of these variables did not differ significantly from normal.

Baseline characteristics of the participants in different groups

The results of ANOVA test on BMI, HOMA-IR, and MPO of subjects in pre-test are given in Table 3. As seen, the results are not significant for any of the variables ($P>0.05$) in baseline between groups.

The effects of aerobic training, curcumin, and their interaction on variables

The results of analysis of covariance to determine the effect of aerobic training and curcumin supplementation on MPO enzyme levels, HOMA-IR, and BMI in overweight middle-aged women are presented in the table 4.

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Table 1: The central indices and distribution of individuals in the pre-test[a]

Variables	Groups			
	Cur	Pla	Cur+Tra	Pla+Tra
Age, y	38.9±5.40	40.6±3.71	35.8±3.22	38.2±5.67
Height, m	159.09±4.64	162.3±4.91	162.44±4.36	159.58±6.98
Weight, kg	67.31±6.54	71.31±5.73	74.45±6.63	70.37±9.23
BMI, kg/m^2	26.96±1.90	27.21±1.43	28.18±1.70	27.52±1.70
HOMA-IR	2.00±1.22	1.72±0.91	1.78±0.64	2.12±0.82
MPO, ng/ml	9.33±2.73	8.78±3.13	9.86±3.81	9.15±3.6

Abbreviations: BMI, body mass index; MPO, myeloperoxidase; Cur, Curcumin; Pla, Placebo; Cur+Tra, Curcumin+Training; Pla+Tra, Placebo+Training; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance;
[a] Values are expressed as mean±SD;

Table 2: Pre-test and post-test values of the studied variables in the subjects[a]

Variables	Groups							
	Cur		Pla		Cur+Tra		Pla+Tra	
	Pre-test	Post-test	Pre-test	Post-test	Pre-test	Post-test	Pre-test	Post-test
BMI, kg/m^2	26.96±1.9	26.51±1.8	27.21±1.4	26.80±1.5	28.18±1.7	27.94±1.5	27.52±1.7	27.06±1.8
HOMA-IR	2.00±1.2	1.70±0.6	1.72±0.9	2.04±1.1	1.78±0.6	1.62±0.5	2.12±0.8	1.73±0.4
MPO, ng/ml	9.33±2.7	10.16±2.7	8.78±3.1	8.75±3.2	9.86±3.8	11.09±4.1	9.15±3.6	10.24±3.8

Abbreviations: BMI, body mass index; MPO, myeloperoxidase; Cur, Curcumin; Pla, Placebo; Cur+Tra, Curcumin+Training; Pla+Tra, Placebo+Training; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance;
[a] Values are expressed as mean±SD;

Table 3: ANOVA test results on BMI, HOMA-IR, and MPO of subjects in pre-test

variable		sum of squares	Df	average of squares	F	P Value[a]
BMI, kg/m^2	Inter-group	8.397	3	2.799	1.073	0.373
	Within-group	93.916	36	2.609		
	Total	102.313	39			
HOMA-IR	Inter-group	1.71	3	0.57	1.454	0.244
	Within-group	5.53	36	5.53		
	Total	7.24	39			
MPO, ng/ml	Inter-group	6.058	3	2.019	0.180	0.909
	Within-group	403.846	36	11.218		
	Total	409.904	39			

Abbreviations: BMI, body mass index; MPO, myeloperoxidase; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance;
[a] P values were considered significant, statistically, less than 0.05.

Table 4: ANCOVA results on the effect of intervention on studied variables in subjects

variable	factor	sum of squares	Df	average of squares	F	Sig.[a]	Effect Size (μ)
BMI, kg/m²	Pre-test	4.11	1	4.11	1.53	0.220	0.407
	group	15.48	3	5.16	1.92	0.134	0.057
HOMA-IR	Pre-test	5.53	1	5.53	14.08	0.001	0.287
	group	1.71	3	1.71	1.454	0.244	0.111
MPO, ng/ml	Pre-test	2.07	1	2.07	0.194	0.661	0.337
	group	44.61	3	14.87	1.38	0.254	0.187

Abbreviations: BMI, body mass index; MPO, myeloperoxidase; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance;
[a]P values were considered significant, statistically, less than 0.05

Based on the results in Table 4, it was revealed that pre-test did not have any significant effect on BMI changes in overweight middle-aged women. That is, after eliminating the pre-test effect, there was no significant difference between the mean BMI in the groups. Accordingly, it was found that after eliminating the pre-test effect or the same covariance factor, there was no difference in mean BMI. Therefore, aerobic training and curcumin supplementation and the combination of these two interventions have no effect on overweight middle-aged women's BMI. Also, by removing the effect of insulin resistance values in the pre-test as the covariate variable, the main effect of group variables on the post-test scores was not significant (F=1.454, P=0.244, μ=0.111). In other words, there was no significant relationship between aerobic training and curcumin supplementation in subjects and their mean HOMA-IR (P>0.05) between groups. The results of covariance analysis showed that pre-test had no significant effect on MPO enzyme level changes in samples. Accordingly, it was found that after eliminating the pre-test effect or the same covariance agent, there was no difference between the mean levels of MPO enzyme (F=0.194, P=0.661, μ=0.337). Thus, curcumin supplementation, alone, does not affect the level of MPO in overweight middle-aged women.

Discussion

In the current study, the influence of aerobic training and curcumin supplementation on myeloperoxidase enzyme levels, BMI, and HOMA-IR in overweight middle-aged women was investigated. Overall, the results of this study showed that aerobic training and curcumin supplementation had no significant effect on myeloperoxidase enzyme levels, BMI, and HOMA-IR in overweight middle-aged women.

According to the findings of this study, aerobic training and supplementation of curcumin had no significant effect on myeloperoxidase in overweight middle-aged women. The results of this hypothesis are in line with the research of Dastani et al. [31], and Duzova et al [32]. In a study of the effect of a selected stationary training course on plasma myeloperoxidase in inactive women by Dastani et al. it was shown that intra-group changes in serum myeloperoxidase levels were not significant in any of the experimental and control groups. Also, intergroup changes were not statistically significant. Duzova et al. investigated two methods of continuous moderate training (60 minutes) and intense running (20 minutes) on treadmill in rats for five weeks. Results of the study did not report a significant change in plasma and muscle MPO in rats in any of the groups [32], which is consistent with the results of the present study. In contrast, Morozov et al., when performed very intensive training in rats with a program including repetitive swimming by adding rats, concluded that very intensive training increased the MPO level of muscle immediately and 24 h after training [33]. It seems that these results are inconsistent with the results of the present

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study, and it may be due to the type of training and the duration of activity in this study. Also, the results of this study showed that 8 weeks of curcumin supplementation and aerobic exercise decreased the insulin resistance index and body mass index after the intervention but this decrease was not significant (P>0.05). A study by Maithilikarpagaselvi et al. in mice showed that curcumin consumption after 10 weeks significantly decreased the insulin resistance index [34]. This result is similar to results of Sh ao et al.[35] and Teich et al. [36] studies. In Shao study, the duration of curcumin intake was 28 weeks. The studies of Bai [37] and Azza [38] have shown similar findings. But, none of the studies reviewed showed inconsistent results with the above findings.

On the other hand, various studies have shown the effect of aerobic training on insulin resistance index. A study by Nezamdoust et al. on type 2 diabetic patients shows that 12 weeks of aerobic training for three sessions per week has a significant relation with insulin resistance index.

This study was a short-term trial that it is unclear whether curcumin supplementation will have significant effects on BMI, HOMA-IR, or MPO in the longer term or not. This is a limitation of this study. Also, the impact of different levels of doses was not investigated in the current research.

It is recommended to investigate the effect of strength training (or interval training) and curcumin supplementation on myeloperoxidase enzyme levels, BMI, and HOMA-IR in overweight middle-aged women.

Conclusions

The results of this study conducted showed that independent variables (aerobic training and curcumin supplementation) had no significant effect on the dependent variables (MPO enzyme and BMI). The results of this study were compared with previous research. The findings of this study appear to be inconsistent with previous studies. Some similar studies showed that these independent variables had a significant effect and others showed no significant effect. However, different reasons can be considered such as age, animal subjects to humans, intensity of training, amount of supplementation and others. Therefore, it is suggested that these two variables should not be used to control MPO and BMI.

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Conflict of Interest

The author declares no conflict of interest.

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