Effect of Omega-3 PUFAs Supplementation with Lifestyle Modification on Anthropometric Indices and Vo₂ max in Overweight Women

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Abstract

Background: Despite the fact that the recommendations of counteracting obesity advocate for changing lifestyle and physical activity habits, the prevalence of obesity continues to rise. The aim of the study was to investigate the effect of omega-3 PUFAs supplementation with lifestyle modification on anthropometric indices and Vo₂max in overweight women.

Methods: Fifty overweight women aged between 20 to 45 years were recruited in this interventional study. Women randomly were divided into two experimental groups (n = 25). Group 1 received omega-3 supplement, aerobic exercise program, and a healthy diet education. Group 2 was similar to group 1, except in that patients received placebo instead of omega-3 capsules. Experimental and placebo group subjects were asked to take one supplementary capsule every day, for 8 weeks. Anthropometric indices were measured in the fourth and eighth weeks of the trial. The maximum aerobic capacity (Vo₂max) was determined using a gas analysis device. The level of significance for comparing the results before and after the trial was considered at P < 0.05.

Results: According to the data, body weight, body fat percentage, waist circumference, and abdominal skinfold thickness significantly reduced in the omega-3 treated group compared to the control group during 8 weeks after the initiation of the study (P < 0.05). In addition, supplementation of omega-3, significantly improved the VO₂max outcome compared to that of the control group (P = 0.03).

Conclusion: According to the results, it seems that omega-3 PUFAS supplementation with lifestyle modification has positive effects on anthropometric indices and Vo, max in overweight women.

Keywords: Anthropometric indices, omega-3, overweight, Vo2max

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Introduction

O besity and overweight are defined as abnormal or excessive body fat accumulation.¹ This phenomenon is strongly associated with systemic inflammation and chronic diseases such as dyslipidemia and cardiovascular disease.² Obesity is commonly measured using body mass index (BMI: weight _(kg)/height²_(m2)). In adults, obesity was defined as a BMI greater than or equal to 30.³ The prevalence of obesity and overweight is increasing throughout the developed and developing world.⁴ National Health and Nutrition Examination Survey data showed the prevalence of overweight increased to 32.3% in 2005 – 2006, in adults aged 20 – 74 years. Furthermore, the prevalence of obesity increased to 2.3 billion overweight teenagers in 2015.⁶ In Iran, the prevalence of obesity is 11.1 % in men and 25.2% in women.⁷

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Obesity is mainly caused by imbalance between energy intake and energy expenditure.⁸ Recent suggestions for reducing body fat are based on lifestyle modifications such as increasing physical activity and eating a healthy, balanced diet; however lifestyle modification is not acceptable to all people .⁹

Docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA) are long-chain omega-3 polyunsaturated fatty acids (PUFAs). EPA is a nonessential n-3 fatty acid, which can convert essential n-3 alpha-linolenic acid (ALA) to EPA and DHA in the human body. However, this conversion is not sufficient to meet the EPA and DHA demand of the body; thus, it is expected to obtain these fatty acids from dietary sources. In addition, omega-3 PUFAs has crucial benefits such as cardiovascular health, central nervous system function, anti-inflammatory role, etc. Omega-3 fatty acids are introduced as fish oil in the human diet.^{10,11} Evidences support the relation between the n-3 fatty acids (FAs) consumption, exercise, and weight loss as omega-3 PUFAs consumption could increase performance during endurance exercise.12,13 It was reported during cycling bouts (60% of VO₂max), the omega-3 PUFAs supplementation reduced plasma glucose disappearance rate and hepatic glucose production, as well as glucose metabolic clearance rate compared to the controls.14 Additionally, omega-3 PUFAs supplementation increased cytoplasmic fatty-acid-binding protein content and fat oxidation in rats.¹⁴ Moreover, a significant decrease in body fat was observed by supplementation of fish oil for 12 weeks plus exercise compared to exercise-only group.¹³ Peoples,

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et al. reported fish oil (8 g/day for 8 weeks) had significantly lowered heart rates during incremental workloads to exhaustion and whole-body oxygen consumption, without significant impact on peak oxygen consumption.¹⁵

Despite widespread recommendations from public health bodies to change lifestyle and physical activity habits, the prevalence of obesity is still high.⁹ A limited number of human studies have been performed directly examining the influence of omega-3 PUFAs supplementation on exercise performance.¹⁴ Also, scarce information exists on the aspects of omega-3 PUFAs consumption with lifestyle modification on weight loss and Vo₂max in overweight women. We hypothesized that consumption of omega-3 PUFAs with lifestyle modification may alter anthropometric indices and Vo₂max in overweight women.

Materials and Methods

Subjects

Fifty overweight women aged between 20 to 45 years were recruited in this double-blinded randomized trial. Overweight volunteer women were recruited through advertising, and selected via preliminary interviews and measurements in the multidisciplinary obesity clinic in the Imam Khomeini Hospital. The inclusion criteria were being female between 20 and 45 years of age; BMI equal to or greater than 25 and less than 30; sedentary lifestyle (not participating in at least 30 minutes of moderate intensity exercise 3 days per week in the 3 months prior to the commencement of the study); not suffering from any known cardiovascular, pulmonary or metabolic diseases; not taking any medication affecting heart rate, blood pressure or exercise capacity; not experiencing musculoskeletal problems that would limit exercise capacity and neither being pregnant nor menopause. In case a participant refused participation or had any indication of exercise test termination during the test, she would be excluded. All protocols for experiments were approved by the institution of Ethical Committee, Tehran University of Medical Science (TUMS) and Health Services (IRCT201504267903N4).

Study Protocol

Women randomly were divided into two experimental groups. Group 1 received omega 3 supplements aerobic exercise, and diet education (n = 25). Group 2 was similar to group 1, except patients received placebo capsules instead of omega 3 capsules (n = 25). After inclusion, in the second visit, the study process such as exercise test and eight weeks of supervised exercise sessions were described. The participants completed and signed the informed consent. Each subject was educated about the 24-hour food consumption record. The 24-hour food records were analyzed using the FPI II software (Food Processor II, Nutrition System ESHA Research, Salem, Oregon 1987), which could calculate the consumed calorie, carbohydrate, protein, and fat intake based on percentage and grams. Finally, the 24-hour records at baseline, fourth and eighth weeks were compared between two groups. Low calorie diet was prescribed using the Harris Benedict Equation with 500 - 1000 kilocalorie deficit. The main groups of foods, serving sizes, and exchange list were described. The educators were expected to engage in a face-to-face training. The third visit for exercise test and anthropometric measurements with proper preparations was determined.

Anthropometric indices

On the third visit, the participants' heights, hip and waist circumferences were measured using a standard tape. Waist circumference was measured immediately above the iliac crest, according to the National Institutes of Health Guideline.¹⁶ The skin fold thickness was measured by the standard Harpenden caliper (British Indicators Ltd, UK). The abdominal skin fold thickness was measured 2 cm in the right side of the umbilicus by a raised vertical fold. Suprailiac skin fold thickness was measured at the cross marking on the anterior axillary line and the horizontal line of the superior border of the ilium, by the raised oblique fold. They were weighed on a medical scale with a precision of 100 grams. The percentage of their body fat, soft lean mass, and lean body mass was calculated using Body Impedance Analyzer (AVIS33 body composition analyzer, Jawon Medical Co. Ltd, South Korea).

Vo₂max index

Before the third visit, participants were requested to avoid high intensity physical activity, to refrain from coffee drinking through the last day, and eat a light meal 2 hours prior to the test. Determination of the maximum aerobic capacity (Vo₂max) was done by gas analysis device (Quark CPET, COSMED, Italy). After calibrating the device, the participants wore the proper mask and exercise test was directed based on Bruce protocol and continued until maximal effort (a respiratory exchange ratio (RER) ≥ 1.1 .¹⁶

Fish oil supplementation

The omega 3 capsules (NATURALab, Canada) contained 600 mg EPA plus 300 mg DHA. The size, color and shape of the placebo capsules were as same as omega 3 capsules (ZAHRAVI Pharmaceutical. CO, Iran). All subjects were asked to take one capsule per day (Omega 3 or placebo) for 8 weeks.

Statistical Analysis

Data were analyzed by two-way analysis of variance (ANOVA) for repeated measurement using SPSS 16.0 for Windows (SPSS Inc. Chicago, IL, USA) and is presented as mean \pm SD. For treatments showing a main effect by ANOVA and linear regression, means were compared using the post hoc Bonferroni test. Mean difference in 8 weeks measurement between two groups adjusted for the baseline values. Differences between treatments were considered to be significant at P < 0.05.

Results

Fifty overweight female subjects were recruited in this study. Mean age of the participants was 36.88 (5.99) in the control group and 36.34 (6.04) in the omega 3 group. Six of the participants did not participate in the exercise sessions and follow up measurements. Finally, a total of 44 subjects were analyzed (Figure 1). Impacts of omega-3 PUFAs supplementation with lifestyle modification on anthropometric indices are presented in Table 1. Also, the effect of omega-3 PUFAs supplementation on Vo₂max is shown in Table 1. The summary of the 24-hour food records over time in control and omega-3 treated groups is provided in Table 2.

Anthropometric outcomes

According to the data in Table 1, body weight significantly diminished in omega-3 treated groups compared to control group at 8 weeks after the initiation of the study (P = 0.001). Also, BMI

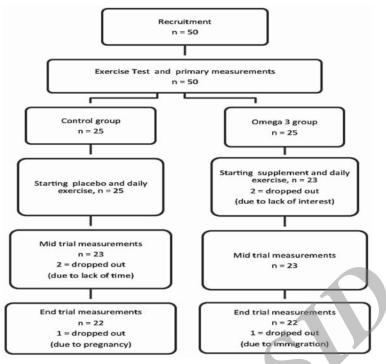


Figure 1. Flow chart of how participants progressed through the study, and how many contributors completed each stage.

| Variables | Baseline | Week 8 | <i>P</i> -value ¹ | Mean difference (95% CI | |
|------------------------------------|----------------------------|----------------------------|------------------------------|-------------------------|-------------------|
| Weight, kg | | | | 0.001 | 1.54 (0.67,2.41) |
| Placebo group ^{£*} | 71.63 (5.57) ^a | 70.91 (5.91) ^a | 70.83 (6.29) ^a | | |
| Omega 3 treated group | 71.43 (6.88) ^a | 70.09 (6.76) ^a | 69.10 (6.59) ^b | | |
| BMI, kg/m ² | | | | 0.001 | 0.59 (0.25,0.94) |
| Placebo group ^{£*} | 27.59 (1.26) ^a | 27.31(1.38) ^a | 27.28(1.56) ^a | | |
| Omega 3 treated group | 27.90(1.45) ^a | 27.38(1.44) ^a | 27.01(1.43) ^b | | |
| Body fat, % | | | | 0.009 | 0.91 (0.26,1.56) |
| Placebo group ^{£*} | 35.50(1.72) ^b | 35.32(2.08) ^b | 35.17 (2.14) ^b | | |
| Omega 3 treated group | 36.54 (2.26) ^a | 36.01 (2.18) ^b | 35.30 (2.32) ^b | | |
| Lean body mass, % | | | | 0.201 | 0.47 (-0.01,0.96) |
| Placebo group ^{£*} | 46.15 (3.11) | 45.82 (3.18) | 45.87 (3.40) | | |
| Omega 3 treated group | 45.25 (3.80) | 44.73 (3.53) | 44.52 (3.52) | | |
| Soft lean mass, % | | | | 0.576 | 0.29 (-0.22,0.79) |
| Placebo group ^{£*} | 42.10 (2.82) | 41.72 (2.85) | 41.76 (3.06) | | |
| Omega 3 treated group | 41.22 (3.43) | 40.78 (3.17) | 40.62 (3.20) | | |
| Waist circumference, cm | | | | 0.001 | 2.34 (0.99,3.69) |
| Placebo group ^{£*} | 95.23 (5.70) | 93.95 (6.35) | 93.84 (6.44) | | |
| Omega 3 treated group | 95.59 (6.77) | 93.66 (6.21) | 91.84 (6.18) | | |
| Hip circumference, cm | | | · | 0.006 | 1.70 (0.52,2.87) |
| Placebo group ^{£*} | 101.88 (4.35) ^a | 100.94 (4.35) ^a | 100.56 (5.06) ^a | | |
| Omega 3 treated group | 102.13 (6.09) ^a | 100.45 (5.78) ^a | 99.09 (5.42) ^b | | |
| Abdominal skinfold thickness, mm | | | | 0.001 | 1.15 (-0.68,2.37) |
| Placebo group ^{£*} | 30.61 (6.82) ^a | 29.63 (6.60) ^a | 29.53 (6.74) ^a | | |
| Omega 3 treated group | 32.03 (8.00) ^a | 30.52 (7.59) ^a | 29.68 (7.38) ^b | | |
| Supra-iliac skinfold thickness, mm | | | | 0.603 | 0.01 (-1.14,1.16) |
| Placebo group ^{£*} | 25.96 (8.45) | 24.84 (8.24) | 24.35 (8.07) | | |
| Omega 3 treated group | 27.51 (8.12) | 26.67 (7.44) | 25.70 (6.77) | | |
| VO ₂ .max | | | | 0.003 | -0.97(-2.08,0.15) |
| Placebo group £* | 30.94 (3.10) ^b | - | 31.78 (3.26) ^b | | |
| Omega 3 treated group | 30.29 (3.82) ^b | - | 32.74 (3.67) ^a | | |

The values are expressed as mean (SD). ¹The *P*-value for Group × Time interaction; BMI: body mass index; ${}^{e}P < 0.05$ for statistical difference from baseline to week 3 within the group; ${}^{*}P < 0.05$ for statistical difference from baseline to week 6 within the group; ${}^{*}P < 0.05$ for statistical difference from baseline to week 6 within the group.

| Table 2. Summary | of 24-hour food | l records over | time in control | and omega | 3 treated groups |
|------------------|-----------------|----------------|-----------------|-----------|------------------|
|------------------|-----------------|----------------|-----------------|-----------|------------------|

| | Assessment time points | | | | | |
|-----------------------------|------------------------|-------------------|-------------------|------------------------------|--|--|
| Variable | Baseline | Week 4 | Week 8 | <i>P</i> -value ¹ | | |
| Total energy intake, kcal | | | | 0.15 | | |
| Placebo group ^{£*} | 1750.64 (204.953) | 1728.86 (238.580) | 1716.77 (110.449) | | | |
| Omega 3 treated group | 1751.59 (289.871) | 1656.23 (140.059) | 1737.23 (254.854) | | | |
| Protein, gr | | | | 0.29 | | |
| Placebo group ^{£*} | 56.72 (17.34) | 59.49 (19.64) | 60.73 (12.00) | | | |
| Omega 3 treated group | 60.60 (20.77) | 60.45 (15.33) | 64.76 (20.69) | | | |
| Energy from protein, % | | | | 0.15 | | |
| Placebo group ^{£*} | 12.73 (3.58) | 14.00 (3.79) | 14.14 (2.88) | | | |
| Omega 3 treated group | 13.68 (4.26) | 15.09 (7.19) | 14.68 (4.19) | | | |
| Carbohydrate, gr | | | | 0.47 | | |
| Placebo group ^{£*} | 239.67 (38.76) | 241.57 (36.36) | 237.06 (44.44) | | | |
| Omega 3 treated group | 229.86 (52.53) | 214.77 (45.76) | 222.47 (46.66) | | | |
| Energy from carbohydrate, % | | | | 0.69 | | |
| Placebo group ^{£*} | 54.41 (9.09) | 55.32 (7.45) | 54.41 (8.89) | | | |
| Omega 3 treated group | 51.55 (9.64) | 51.00 (7.13) | 50.36 (7.63) | | | |
| Fat, gr | | | | 0.12 | | |
| Placebo group ^{£*} | 65.69 (19.93) | 60.12 (17.55) | 60.62 (15.31) | | | |
| Omega 3 treated group | 68.87 (20.80) | 64.67 (15.16) | 67.95 (16.62) | | | |
| Energy from fat, % | | | | 0.68 | | |
| Placebo group £* | 32.77 (1.70) | 31.09 (9.20) | 33.00 (10.43) | | | |
| Omega 3 treated group | 34.73 (8.24) | 34.77 (7.00) | 34.73 (6.84) | | | |

The values are expressed as mean (SD). ¹The *P*-value for Group × Time interaction; ${}^{t}P < 0.05$ for statistical difference from baseline to week 4 within the group; ${}^{*}P < 0.05$ for statistical difference from baseline to week 8 within the group. ${}^{*}P < 0.05$ for statistical difference from baseline to week 8 within the group.

significantly diminished in the group consuming omega-3 compared to that in the control group (P = 0.001). As seen, body fat percentage diminished significantly in the group receiving omega-3 in comparison to that in the control group at different assessment time points (4 and 8 weeks) (P = 0.009). The same procedure was observed in abdominal circumference and abdominal skinfold thickness in omega 3-treated group compared to the placebo group, where the abdominal circumference and abdominal skinfold thickness diminished in omega 3-treated group (P < 0.05).

VO₂max outcome

According to the data, supplementation of omega 3, significantly improved VO₂max outcome compared to the control group (P = 0.003) (Table 1). As seen, VO₂max outcome increased in the control group, but the difference was not significant. However, over time VO₂max outcomes improved significantly in the omega 3 treated groups (P < 0.05).

Results of 24-hour food record

As seen in Table 2, there was no significant difference in the 24-hour food records over time in the omega 3 group compared to control group (P > 0.05).

Adherence to exercise

As seen in Table 3, there was no significant difference for days and time spent on exercise per week between control and omega 3 treated groups during the eight weeks of the study (P > 0.05).

Discussion

Several investigations have been done to determine the effect of omega-3 PUFAs on body function, but mechanisms of action of

omega-3 PUFAs are complex, but still incompletely understood.¹⁷ To the best of our knowledge, there are no studies describing the effect of omega-3 PUFAs supplementation with lifestyle modification on anthropometric indices and Vo₂max in overweight women. According to results, body weight, body fat percentage, as well as abdominal circumference and abdominal skinfold thickness diminished in omega 3 treated groups at 8 weeks after the initiation of the study.

A large number of experimental and epidemiological studies have been conducted to examine the effect of consuming fish oil on health.¹³ It is reported that consumption of n-3 is associated with favorable alterations in body composition. Animal studies revealed consumption of n-3 and n-6 fatty acids reduced adiposity and increased lean tissue growth.¹⁸ To date, several experiments have been done to investigate the effect of differing dietary fatty acid compositions on body composition in human. Some human trials reported reductions in fat mass with n-3 consumption compared to other oils.13 Omega-3 were preferentially metabolized by the body after ingestion than other fatty acids.¹⁹ In a study, Thorsdottir, et al. reported omega 3 supplementation for 8 weeks diminished waist circumference in overweight young adults.²⁰ Recently, in a study on animal models, it is reported n-3 fatty acids promote size reduction of visceral adipose depots, without altering body weight and composition, in male Wistar rats fed a highfat diet.²¹ Several mechanisms were reported for possible action of omega-3 FA on weight loss. The mechanisms by which long chain omega-3 PUFA assists the reduction of body fat and/or body weight are still being explored. It is suggested omega-3 PUFA modulates lipid metabolism, promoting lipolysis, enhancing hepatic fatty acid oxidation, and inhibiting fatty acid synthesis.1 N-3 FA increase hepatic activities of carnitine palmitoyltransferase-II. Therefore, it seems via this mechanism n-3 FA increases fatty

| Weeks | Groups | Days in week (±SD) | P-value | Time in week (min) (±SD) | P-value | |
|-------|-----------------------|--------------------|---------|--------------------------|---------|--|
| 1 | Placebo group | 3.86 (1.03) | - 0.34 | 152.05 (61.73) | 0.11 | |
| 1 | Omega 3 treated group | 4.14 (0.83) | 0.34 | 183.64 (58.59) | 0.11 | |
| 2 | Placebo group | 3.68 (1.21) | 0.70 | 155.91 (82.38) | 0.58 | |
| 2 | Omega 3 treated group | 3.81 (1.14) | 0.70 | 172.5 (89.51) | 0.38 | |
| 3 | Placebo group | 3.50 (1.30) | 0.30 | 13.77 (71.9) | 0.15 | |
| 3 | Omega 3 treated group | 3.89 (.099) | 0.30 | 174.55 (79.95) | 0.15 | |
| 4 | Placebo group | 3.36 (1.17) | - 0.46 | 131.36 (58.59) | 0.32 | |
| 4 | Omega 3 treated group | 3.59 (0.59) | 0.40 | 147.95 (56.56) | | |
| 5 | Placebo group | 3.64 (1.00) | 0.61 | 137.95 (58.59) | 0.39 | |
| 5 | Omega 3 treated group | 3.77 (0.75) | 0.01 | 161.59 (64.72) | 0.39 | |
| 6 | Placebo group | 3.59 (0.09) | 0.73 | 137.95 (47.8) | 0.26 | |
| 0 | Omega 3 treated group | 3.68 (0.83) | - 0.75 | 152.50 (57.93) | 0.36 | |
| 7 | Placebo group | 3.00 (0.91) | 0.28 | 135.45 (49.97) | 0.13 | |
| 7 | Omega 3 treated group | 3.77 (0.81) | - 0.38 | 162.27 (64.76) | 0.15 | |
| 0 | Placebo group | 3.32 (0.89) | 0.74 | 121.82 (47.62) | 0.72 | |
| 8 | Omega 3 treated group | 3.31 (0.89) | 0.74 | 126.82 (48.44) | 0.73 | |

Table 3. Frequency and time of exercise during the 8 weeks in control and omega 3 treated groups

acid oxidation.²¹ On the other hand, n-3 FA stimulates mitochondrial and peroxisomal fatty acid oxidation in liver and muscle, as well as inhibition of hepatic lipogenesis and VLDL formation. Also, DHA inhibits cyclooxygenase, the key enzyme involved in the synthesis of these compounds.²² All these mechanisms could contribute to the possible explanation for the greater reduction in weight experienced by the women in this study. However, in this study, we were not able to determine fatty acid oxidation in overweight women.¹

According to the data, supplementation of omega 3 improved VO₂max outcome compared to that of the control group. As with the research on body composition, there are limited reports on direct effects of n-3 supplementation on exercise performance. It is reported fish oil supplementation significantly attenuated RBC deformability under hypoxic conditions.²³ Furthermore, Peoples, et al. studied the role of fish oil supplementation on oxygen consumption during exercise.¹⁵ They reported, those individuals who received fish oil had significantly lowered heart rates during incremental workloads up to exhaustion, lowered steady-state submaximal exercise heart rates, and whole-body oxygen consumption. Therefore, fish oil could enhance oxygen delivery to contracting muscle and maximum oxygen uptake (VO,max), thus improving endurance performance.24 In this regard, controversial reports exist where our finding was consistent with some^{25,26} but not all reports.²⁷ As mentioned before, a number of studies suggest that n-3 derivatives upsurge the deformability of RBCs, which might promote oxygen and nutrient delivery to exercising muscles and thereby increasing performance.13

Based on outcomes of 24-hour food record from Table 2, there was no significant difference in 24-hour food records over time among different groups. Therefore, both groups received uniform diet and calories and this may minimize experimental error.

Despite a wealth of research on the health-related benefits of n-3

acids, studies investigating the effects of combining n-3 supplementation and exercise are limited. As observed from our results, exercise had no additive role on body weight reduction in omega 3-treated group compared to control group because exercise adherence in both groups was similar (Table 3). Previously it was shown, that administration of 3.6 g/day conjugated linoleic acids (CLA) for six weeks plus exercise are effective in improving endurance performance and body composition.28 With respect to exercise, one potential mechanism whereby n-3 supplementation may enhance benefits is via increased lipolysis and β -oxidation. The n-3 acids are able to bind and activate the peroxisome proliferator-activated receptor (PPAR) isoforms, including PPAR-a, PPAR- γ and PPAR- δ . PPARs are members of the nuclear receptor superfamily. The n-3 acids have affinity to the PPAR- α , which exhibit high oxidative rates of fatty acids. PPAR-α plays an integral role in the expression of several genes for lipid transport and oxidation, including hepatic such as carnitine acyltransferase in hepatic and skeletal muscle peroxisomal acyl-CoA oxidase. Increased PPAR- α activity should enable a greater reliance on fat for fuel during exercise. Additionally, n-3 acids might indirectly affect lipid oxidation via suppressing the generation of acetyl-CoA carboxylase.13 Another mechanism, whereby n-3 acids may confer a positive effect on exercise, is via improving fatty acid delivery to exercising muscles by an increased blood flow.13

Finally, the authors recommend merit researches needed to identify direct cellular and molecular signaling pathways of omega 3 with lifestyle modification on anthropometric indices and Vo_2max in overweight women.

Conflict of interest

Authors declare that they have no conflict of interest.

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