

Article

Aerobic Dance Wore with EMS Suit Improves Fatness and Biomarkers of Obese Elderly Women

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Abstract: Electromyostimulation (EMS) has been shown to improve body composition, but what biomarkers it affects has not been investigated. The purpose of this study was to compare the EMS-effect of aerobic dance on fatness and biomarkers' levels in obese elderly women. Methods: Twenty-five women with obesity were randomly classified into a control group (CON; n = 12) and EMS group (EMSG; n = 13). EMS suits used in this study enabled the simultaneous activation of eight muscles with selectable intensities. Program sessions of EMS were combined with aerobic dance three times a week for 8 weeks. Although both groups received the same program, CON did not receive electrical stimuli. Results: Compared with CON, a significant effect of the EMS intervention concerning decreased fatness, as well as an increased skeletal muscle mass and basal metabolic rate, were evident. Compared with CON, aerobic dancing with an EMS suit also improved biomarkers in EMSG. Cytokines, including interleukin-6, tumor necrosis factor, C-reactive protein, resistin, and carcinoembryonic antigen were significantly changed in EMSG, whereas those of CON did not change from the baseline to the end of the experiment. These results showed significant differences between groups. Similarly, the changes caused by EMS were represented in high-density lipoprotein-cholesterol and low-density lipoprotein-cholesterol. Conclusions: The results indicate that a significant effect due to the EMS intervention was found concerning body composition and biomarkers in obese elderly women.

Keywords: aerobic dance; electromyostimulation; percent fat; cytokine

1. Introduction

The elderly population is increasing all over the world, and the incidence rate of chronic degenerative diseases is also increasing rapidly. According to a survey, 90% of seniors aged 65 and older have chronic diseases such as obesity, arthralgia, low back pain, high blood pressure, and cancer, and more than half have difficulties in their daily lives [1]. Entering an aging society is a celebration of the increased life expectancy, but to be truly celebrated, chronic diseases must be solved; otherwise, the "old and sick population" will only increase, and as a result, the elderly will be a huge burden on society. The medical community and related officials think that there is an urgent need to establish a plan to sequentially reduce chronic degenerative diseases ahead of the 'aging society'.

Among many chronic diseases, obesity causes complications, so there is an urgent need for a solution. In other words, obesity can get worse with age, and in older people, obesity is likely to cause more serious health complications. Obesity is defined as a medical condition in which excessive

amounts of body fat accumulate, resulting in detrimental health effects [2]. In women, obesity aggravates physical conditions [3], which are influenced by negative perceptions about body shape [4].

There are many methods for improving obesity, and among them, regular exercise is regarded as an important means of relieving obesity. In other words, exercise is an important factor for increasing fat metabolism in the skeletal muscle, thereby causing fat oxidation. However, it is not easy for the elderly to participate in exercise like young people due to the degeneration of joints and muscles [4,5]. In terms of the relationship between degenerative arthritis and obesity, Kellgren [6] reported that obese people have a high incidence of degenerative arthritis in the knee and the first metatarsal bone. The reason for this is because the amount of fat in the lower limbs increases the distance between both femoral joints, and the knee joint becomes the genu varum. Taken together, previous studies have suggested that the risk factors for the development of the most prevalent degenerative arthritis among chronic diseases can be summarized as aging, being female, and obesity. In patients with degenerative arthritis, rehabilitation exercise involves lower limb muscle development and weight loss through aerobic exercise [7,8]. Exercises mainly for weight loss include walking, biking, and swimming, which can reduce pain and improve physical ability. However, for elderly people who have degenerative arthritis and obesity, which promotes this disease, pursuing exercise to strengthen muscles without burdening the knee joint is recommended.

Recently, an advanced piece of equipment for improving body composition has been developed. Electromyostimulation (EMS) has the advantage of inducing muscle contractions without direct stimulation of the peripheral muscles by the central nervous system, and providing similar effects to muscle contractions. In other words, EMS allows older people to reduce body fatness without having to provide excessive loads to muscles and joints. In particular, an obese person is likely to have problems with muscles and joints, so it is difficult to provide an excessive load, but EMS can provide effectiveness without causing such a problem. It is also gentle on the joints and reduces the risk of injury due to excessive loading [9]. EMS impulses are transmitted through electrodes on the skin located close to the dermis tissue for stimulation and modulate a variety of electrical wave forms, resulting in an electrical current that can be used to stimulate innervated muscles [10,11]. In the case of muscle contractions via EMS, the motor units under the control of the larger nerves are activated and muscle fibers with high thresholds are easily mobilized, resulting in positive effects on strength [12,13]. In the past, a researcher suggested that EMS breaks down the fatty capsule that covers the muscle and also improves the blood supply to the muscles, thereby helping to gain back lost muscle tone and return it to its original size [14]. In other words, it is effective in preventing obesity by reducing fat mass through EMS. Until now, many researchers have reported that EMS has been used for the healing of pressure sores, and improving muscular endurance and strength [15-21].

Although EMS has been reported to improve body composition, strength, and performance, there is a lack of research among obese elderly people. Moreover, few studies have been conducted on changes in cytokines related to obesity via EMS. While the degree of visual improvement in obesity is also important, the changes of blood biomarkers and lipids in the human body are more important. In particular, the aging process, when combined with obesity, contributes to increased cytokines and tumor factors generated by various organs, which may lead to health impairments or shorten one's lifespan.

Therefore, the aim of this study was to investigate the changes in body composition and cytokines when EMS was used during the course of providing elderly people with aerobic dance. The reason why we chose a variety of biomarkers in this study was because we wondered whether the aerobic dance and EMS program could affect inflammatory substances, blood lipid factors, and tumor factors induced in human tissues. This study examined a group of patients with obesity in a randomized controlled trial and assessed the physiological effects of the EMS intervention.

2. Materials and Methods

2.1. Study Design and Participants

This is a prospective, randomized, and controlled study, which takes EMS as the independent variable, and body composition and cytokines as the dependent variables. This study took place from October 4 to December 4, 2018. The first assessment was conducted from October 4 to 5, 2018 and the last assessment was conducted from December 3 to 4, 2018. The program period for the aerobic dance with an EMS suit was from 8 October to 30 November, 2018. Prior to the study, participants received detailed explanations regarding study procedures and were then asked to complete a questionnaire. The inclusion criteria required that patients were obese in terms of the percent of fat and had not exercised regularly for over 6 months. Additionally, patients were also included if they had not received treatment or medication for weight loss or anything known to affect body composition and cytokines, and if they did not have any internal metallic materials. Exclusion criteria consisted of having a history of impairment of a major organ system or a psychological disorder.

All patients knew they had to wear their EMS suits when aerobic dancing, but they did not know if electrical stimulation was provided. In other words, they were assigned using random number tables and assigned identification numbers upon recruitment. In order to prevent communication between the electromyostimulation group (EMSG) and control group (CON), the patients were classified according to their community areas, and EMSG was sent to the center in the morning and CON in the afternoon. At the beginning of the measurement, only EMSG realized that a current was coming from their suits. After excluding seven patients out of thirty-two eligible participants, the remaining thirty patients belonged to one of two groups. Of the 15 patients in the CON who were allocated to the non-EMS group, one did not receive assessment and two were lost in the follow-up phase. Therefore, 12 patients in the CON were analyzed in our study. Furthermore, of the 15 patients in the EMSG, one did not receive assessment and another was lost in the follow-up phase. Therefore, 13 patients of the EMSG were analyzed in our study. Participant characteristics, which indicated homogeneity, are presented in Table 1.

Table 1. Physical characteristics of the subjects

Variables (unit)	CON (n = 12)	EMSG (n = 13)	Z*	p
Age (y)	71.75 ± 2.73	70.38 ± 2.93	-0.908	0.376
Height (cm)	151.49 ± 2.50	151.51 ± 4.02	-0.328	0.769
Weight (kg)	63.75 ± 3.82	63.05 ± 8.78	-0.272	0.810
Percent fat (%)	38.33 ± 4.31	39.21 ± 1.76	-0.082	0.936

All data represents the mean ± standard deviation. EMS and CON mean the electromyostimulation group and control group, respectively. Symbol * was analyzed by the Mann–Whitney U test.

2.2. Research Ethics

This study was conducted in accordance with the Declaration of Helsinki and was approved by the ethics committee at Sahmyook University (Ref. No: 2-1040781-AB-N-01-2017083HR). Written informed consent was obtained before enrollment. First, all of the patients arrived at Songdo hospital to sign an informed consent form and completed a self-reported questionnaire about their health status. After this procedure, all subjects participated in the experiment conducted by an expert.

2.3. Anthropometric Measurements

To measure body composition, all patients were weighed while wearing light clothes and no shoes. The bioelectrical impedance analysis was employed using the BMS 330 and InBody 320 Body Composition Analyzer (Biospace Co., Ltd., Korea), respectively. This analyzer is a segmental impedance device measuring voltage drops in the upper and lower body. Eight tactile electrodes located in palms, fingers, front soles, and rear soles were placed on the surfaces of the hands and feet. The precision of the repeated measurements expressed as the coefficient of variation was, on average, 0.6% for the percentage of fat mass [22]. This analyzer is a segmental impedance device in which the

electrodes are made of stainless steel interfaces. The subjects stood upright by placing their bare feet on the foot electrodes and gripping the hand electrodes. The analysis of body composition was measured before dinner and after voiding [23].

2.4. Biomarker Measurements

Blood samples were taken after fasting for 10 hours or longer before assessment and were collected using BD vacutainer tubes (Becton Dickinson, NJ, USA) at 8 am the following day. After the subjects were stabilized for 10-15 minutes, 5 mL of blood was collected from the antecubital vein of the subjects with a disposable syringe by a medical laboratory technologist before and after the experiments. A total of 2 mL of the 5 mL of venous blood was added to an anticoagulant tube (EDTA bottle), shaken, and centrifuged at 3000 rpm for 5 minutes. The remaining 3 mL was left at room temperature for 1 hour and centrifuged at 1000 rpm for 15 minutes. Isolated plasma and serum were kept frozen until the test. The samples were taken to the laboratory for analysis, as follows.

Interleukin-6 (IL-6) from the serum was analyzed using an ELISA kit (Cohesion Biosciences, London, UK). The minimum detectable dose of Human IL-6 is typically less than 1 pg/mL. The Human IL-6 ELISA Kit allows for the detection and quantification of endogenous levels of natural and/or recombinant Human IL-6 proteins within the range of 3.9 to 250 pg/mL. Tumor necrosis factor- α (TNF- α) from serum was analyzed using an enzyme-linked immunospecific assay (ELISA) kit (Cohesion Biosciences, London, UK). The serum was allowed to clot in a serum separator tube at room temperature, was centrifuged at approximately 1000 \times g for 15 min, and was immediately analyzed [24]. The minimum detectable dose of Human TNF- α is typically less than 7 pg/mL. The Human TNF- α ELISA Kit allows for the detection and quantification of endogenous levels of natural and/or recombinant Human TNF- α proteins within the range of 15.6 to 1000 pg/mL. C-reactive protein (CRP) from the serum was also analyzed using an ELISA kit (Cohesion Biosciences, London, UK). The minimum detectable dose of Human CRP is typically less than 10 pg/mL. The Human CRP ELISA Kit allows for the detection and quantification of endogenous levels of natural and/or recombinant Human CRP proteins within the range of 15.6 to 1000 pg/mL. Resistin (RSTN), known as adipose tissue-specific secretory factor, from the serum was analyzed using an ELISA kit (Phoenix Pharmaceuticals, London, UK). The standard solution and sample for RSTN were added to a microplate coated with a specific RSTN monoclonal antibody and bound to RSTN (#EK-S-028-36) to form immobilized antibodies. Afterwards, unbound material was removed by washing and a biotinylated polyclonal antibody specific for biomarkers was added to each well. Unbound antibody-enzyme for biomarkers was removed by washing and Horseradish Peroxidase was added to each well, respectively. The carcinoembryonic antigen (CEA) was measured by the Sandwich principle of quantitative chemiluminescence assay, with a Cobas 8000 e801 (Roche Diagnostics, Mannheim, Germany). For reference, a normal value of CEA is ≤ 3.8 ng / mL for nonsmokers and ≤ 5.5 ng / mL for smokers. A binding substance was developed after the washing process and addition of substrate solution (TMB) [19].

High-density lipoprotein-cholesterol (HDL-C) and low-density lipoprotein-cholesterol (LDL-C) were also measured by the homogeneous enzymatic colorimetric assay, with a Cobas C702 (Roche, Mannheim, Germany). For reference, a normal value of HDL-C is over 40 mg/dL and a normal range of LDL-C is <100 to 129 mg/dL.

2.5. Aerobic Dance Program with EMS Administration

All patients completed a supervised progressive program for 8 weeks. Based on recommendations from available literature [9,20,25-27], the stimulation frequency was selected at 85 Hz, the impulse-width at 350 μ s, the impulse-rise as a rectangular application, and variable electrostimulation intensities relative to the maximum peak voltage. This study used 1 MT (maximal tolerance) as the maximum peak voltage, similar to calculating the maximal voluntary contraction as 1 maximal repetition [13]. In measuring 1 MT, each participant in the EMSG stood still while wearing EMS suits. In order to prevent the patients from being surprised or uncomfortable with the electrical stimulus, the 1 MT level was gradually increased while providing a low stimulation current [17,28-30]. In other

words, the electric stimulation was stopped at the request of the participant when reaching an unbearable level, at which point the intensity was set as 1 MT.

The patients of EMSG were assigned to 60% of 1 MT from the baseline to Week 2, 70% of 1 MT from Week 3 to Week 5, and 80% of 1 MT from Week 6 to Week 8, respectively. Although the patients in CON performed aerobic dancing while wearing EMS suits, they did not receive any electrical stimuli. All patients were asked to express the difficulty level of exercise during aerobic dancing with an EMS suit. An instructor asked for the ratings of perceived exertion (RPE) every 5 minutes, and an assistant recorded them. Meanwhile, the impulse duration was 6 s, with a 4-s break between impulses. For EMSG, an instructor conducted EMS sessions three times a week on two non-consecutive days (Mon., Wed., and Fri.) to allow for a rest interval of 48 hours between each session.

In order to provide effective muscular contractions and to prevent harmful joint injury, the dance movements were simplified and composed of clapping and tapping, bending and rotating, aerobic and anaerobic exercises, and stretching exercises, which were performed according to the instructor's directions. The aerobic dance with an EMS suit was as follows. Warm-up was performed at RPE of 9-11 for 5 minutes of walking in place. After that, upper and lower leg stretching was performed with the same intensity and time. Work-out with dance consisted of clapping and tapping (8 min), bending and rotating (12 min), aerobic and anaerobic exercises (10 min), and stretching exercises (10 min). Initially, clapping and tapping with dance began with applause for 1 minute, followed by tapping of the head, shoulders, torso, back, and legs. At this time, the exercise intensity was between RPE 9 and 11. Second, bending and rotating with dance was performed between RPE 11 and 13 for 12 minutes with kneeling, followed by standing, standing on toes, waving arms, lifting legs, bending the waist upward, and rotating shoulders. Third, aerobic and anaerobic exercises with dance were performed between RPE 11 and 13 for 10 minutes with stepping forward and sideways, stepping backward and sideways, heel raises, lifting knees, curling legs, front lunges, and cross lunges. Fourth, stretching exercises with dance began with clapping and drawing an X-shape, followed by 10 minutes of RPE 11-15 for rotating arms, stretching arms, rotating wrists, shaking wrists, bowing, and rotating shoulders. Finally, the exercise was finished by stretching the upper and lower bodies for 5 minutes.

2.6. Data Analysis

All data are reported as the mean (SD). The sample size was determined using G*Power v 3.1.3, considering an a priori effect size of $f^2(V) = .35$ (medium size effect), α error probability of .05, and power ($1 - \beta$ error probability) of .95. Based on the results of the Shapiro-Wilk test, the non-parametric Mann-Whitney U test and Wilcoxon rank test were used to examine the differences of variables between groups and to examine the changes of variables between times. A significance of $p < 0.05$ was employed. SPSS 18.0 (SPSS Inc., Chicago, IL, USA) was used for all analyses.

3. Results

3.1. Effect of EMS on Body Composition

As shown in Table 2, the body weight of CON did not change, while that of EMSG decreased significantly. These results showed differences between groups after 8 weeks. The skeletal muscle mass of EMSG increased significantly ($p = 0.009$), whereas that of CON did not change, indicating a significant difference between groups ($p = 0.019$). On the other hand, although the fat mass of EMSG was significantly decreased while that of CON was not changed, there was a significant difference between groups (Fig. 1). The percent of fat of CON increased, although not significantly, while that of EMSG showed a tendency to decrease. This result showed a significant difference between groups after the experiment (Fig. 1). Specifically, the basal metabolic rate (BMR) of EMSG significantly increased, whereas that of CON significantly decreased, by the end of the experiment. This result showed a significant difference between groups at post-value ($p = 0.001$). In other words, a significant effect of the EMS intervention concerning body composition was evident.

Table 2. Differences and changes in body composition

Items		Groups		Z (p)*
		CON (n = 12)	EMSG (n = 13)	
Body weight (kg)	Pre	63.75 ± 3.82	63.05 ± 8.78	-0.272 (0.810)
	Post	62.33 ± 4.17	57.54 ± 6.58	-2.124 (0.035)
	Z (p)**	-0.353 (0.724)	-3.041 (0.002)	
Skeletal muscle (kg)	Pre	20.46 ± 0.97	20.94 ± 2.03	-1.774 (0.077)
	Post	19.82 ± 1.71	22.41 ± 2.19	-2.342 (0.019)
	Z (p)**	-0.179 (0.858)	-2.623 (0.009)	
Fat mass (kg)	Pre	25.23 ± 2.93	26.50 ± 5.50	-0.627 (0.538)
	Post	25.42 ± 2.66	22.31 ± 3.89	-2.205 (0.026)
	Z (p)**	-0.314 (0.753)	-2.900 (0.004)	
Percent fat (%)	Pre	38.33 ± 4.31	39.21 ± 1.76	-0.082 (0.936)
	Post	39.66 ± 2.32	34.95 ± 3.82	-3.267 (0.001)
	Z (p)**	-1.603 (0.109)	-2.485 (0.013)	
BMR (kcal)	Pre	1187.00 ± 35.92	1188.69 ± 67.98	-0.300 (0.769)
	Post	1110.17 ± 40.62	1237.77 ± 74.40	-3.601 (0.001)
	Z (p)**	-2.989 (0.003)	-2.134 (0.033)	

All data represents the mean ± standard deviation. EMS, CON, and BMR mean the electromyostimulation group, control group, and basal metabolic rate, respectively. Symbols * and ** were analyzed by the Mann–Whitney U test and Wilcoxon rank test, respectively.

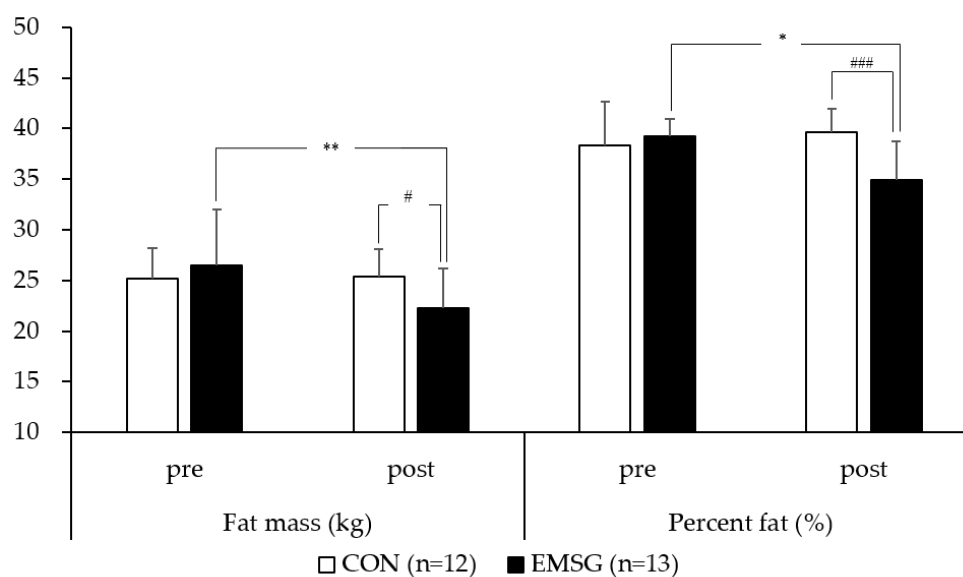


Fig. 1. Changes within a group and differences between groups of fat mass and percent fat. CON and EMSG mean the control group and electromyostimulation group, respectively. Symbols * and ** mean significance between the pre-value and post-value, where * was $p < .05$ and ** was $p < .01$, respectively. Symbols # and ### mean significance between CON and EMSG, where # was $p < .05$ and ### was $p < .001$, respectively.

3.2. Effect of EMS on Biomarkers

As shown in Table 3, IL-6 significantly changed in EMSG, whereas a change in CON did not occur from the baseline to the end of the experiment. This result showed a significant difference between groups ($p = 0.026$; Fig. 2). Similarly, the changes of other cytokines in EMSG were represented in TNF- α ($p = 0.001$; Fig. 2), CRP ($p = 0.001$), RSTN ($p = 0.001$; Fig. 3), and CEA ($p = 0.004$; Fig. 3).

Although the HDL-C of EMSG was not significantly changed, that of CON was significantly decreased. Meanwhile, the LDL-C of EMSG was significantly decreased, but that of CON was not significantly changed. This result demonstrated a significant difference ($p = 0.014$) between the groups after 12 weeks. In other words, significant effects due to the EMS intervention were found concerning biomarker variables in obese elderly women.

Table 3. Differences and changes in biomarkers

Items (units)		Groups		Z (p) *
		CON (n = 12)	EMSG (n = 13)	
IL-6 (pg/mL)	Pre	14.29 ± 7.52	14.51 ± 7.14	-0.355 (0.728)
	Post	15.41 ± 3.79	9.95 ± 6.31	-2.232 (0.026)
	Z (p) **	-1.257 (0.209)	-2.691 (0.007)	
TNF-a (pg/mL)	Pre	28.21 ± 8.51	27.28 ± 12.35	-0.109 (0.936)
	Post	36.68 ± 11.68	20.77 ± 8.64	-3.160 (0.001)
	Z (p) **	-2.826 (0.005)	-2.273 (0.023)	
CRP (pg/mL)	Pre	34.33 ± 15.80	33.19 ± 10.30	-0.218 (0.852)
	Post	54.65 ± 11.66	26.65 ± 8.13	-4.247 (0.001)
	Z (p) **	-3.062 (0.002)	-1.573 (0.116)	
RSTN (ng/mL)	Pre	5.92 ± 1.98	5.56 ± 2.55	-0.654 (0.538)
	Post	8.43 ± 4.06	3.33 ± 1.09	-3.485 (0.001)
	Z (p) **	-1.491 (0.136)	-3.044 (0.002)	
CEA (ng/mL)	Pre	2.12 ± 1.21	2.24 ± 0.66	-0.628 (0.538)
	Post	2.88 ± 1.07	1.42 ± 0.19	-2.833 (0.004)
	Z (p) **	-1.888 (0.059)	-2.972 (0.003)	
HDL-C (mg/dL)	Pre	50.92 ± 9.82	47.08 ± 9.99	-0.300 (0.769)
	Post	46.33 ± 8.63	51.69 ± 7.70	-1.363 (0.186)
	Z (p) **	-2.367 (0.018)	-1.297 (0.195)	
LDL-C (mg/dL)	Pre	134.17 ± 43.47	145.31 ± 35.44	-1.035 (0.320)
	Post	131.83 ± 36.60	97.54 ± 23.88	-2.451 (0.014)
	Z (p) **	-0.236 (0.813)	-2.552 (0.011)	

All data represents the mean ± standard deviation. EMSG, CON, IL-6, TNF, CRP, RSTN, CEA, HDL-C, and LDL-C mean the electromyostimulation group, control group, interleukin-6, tumor necrosis factor, C-reactive protein, resistin, carcinoembryonic antigen, high density lipoprotein-cholesterol, and low density lipoprotein-cholesterol, respectively. Symbols * and ** were analyzed by the Mann–Whitney U test and Wilcoxon rank test, respectively.

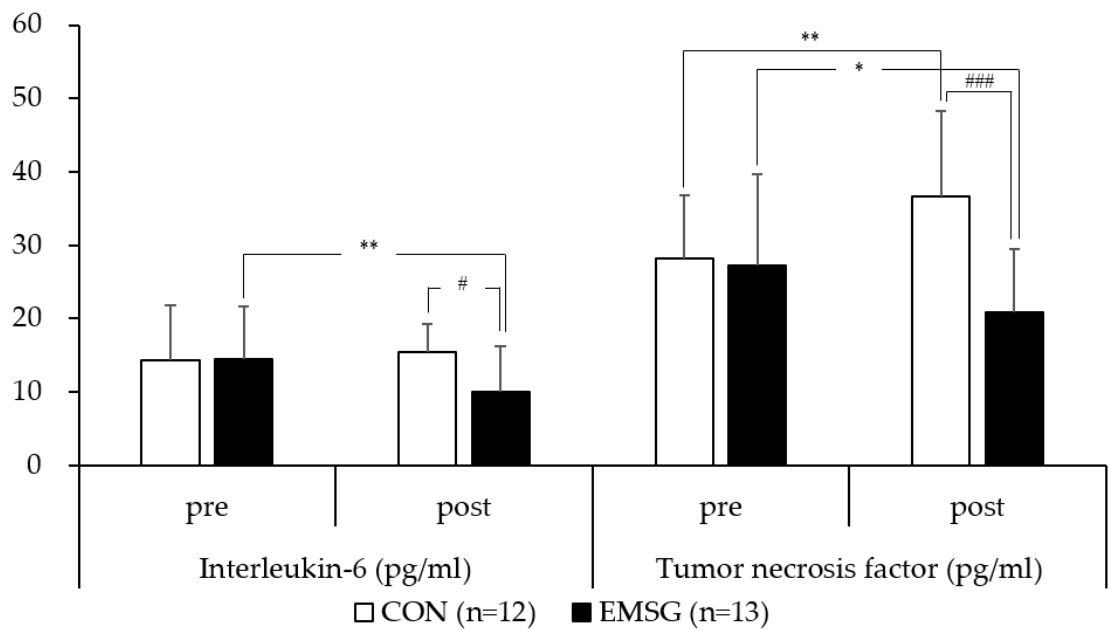


Fig. 2. Changes within a group and differences between groups of interleukin-6 and tumor necrosis factor. CON and EMSG mean the control group and electromyostimulation group, respectively. Symbols * and ** mean significance between the pre-value and post-value, where * was $p < .05$ and ** was $p < .01$, respectively. Symbols # and ### mean significance between CON and EMSG, where # was $p < .05$ and ### was $p < .001$, respectively.

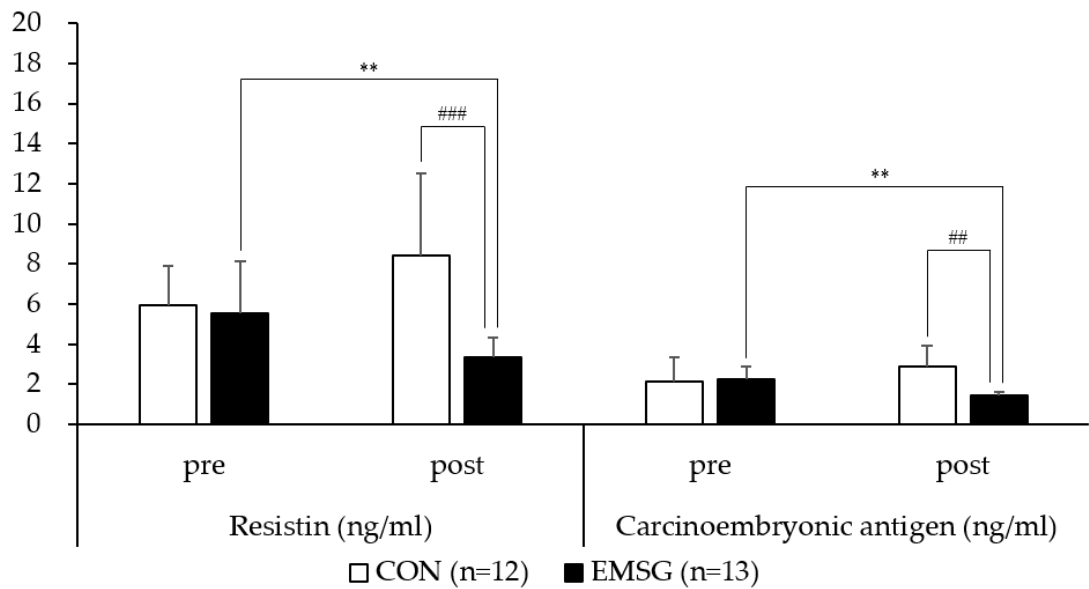


Fig. 3. Changes within a group and differences between groups of resistin and carcinoembryonic antigen. CON and EMSG mean the control group and electromyostimulation group, respectively. Symbols ** means significance between the pre-value and post-value, where ** was $p < .01$. Symbols ## and ### mean significance between CON and EMSG, where ## was $p < .01$ and ### was $p < .001$, respectively.

4. Discussion

This study found some evidence that aerobic dancing with EMS suits improved body composition. Specifically, the body weight and fat mass in EMSG were significantly changed, and those in CON increased significantly. Additionally, almost all biomarkers in CON showed no change after 8 weeks, although the cytokines significantly decreased in EMSG. In addition, LDL-C in EMSG was significantly decreased by the end of the experiment, although it was not changed in CON. These results revealed significant differences between the groups after 8 weeks.

The results of this study showed that the oxidative effect of aerobic dance combined with the muscle contractions provided by EMS had a beneficial effect. In other words, this positive effect in EMSG was thought to be crucial in the increase of skeletal muscle mass and BMR after the end of the experiment. The results of this study are thought to be similar to those of several research studies. Anderson et al. [31] reported that the improvement in anthropometric measures was greater for walking and EMSG compared with walking only and CON in sedentary adult women after 8 weeks. Porcari et al. [32] indicated that there was a significant reduction in body fat when exercise was combined with EMS. In fact, aerobic dance is a type of oxidative exercise that can be enjoyed while listening to music. It can also develop the cardiopulmonary function, as well as muscular strength and endurance. The aerobic dance used in this study was designed to avoid using the full range of motion to prevent joint complications in elderly women. For this reason, we think that the body composition and cytokines in the CON did not change or negatively changed, compared with those of EMSG. In other words, CON was not positively altered in terms of body composition and cytokines due to the lack of EMS's supplementary muscle stimulation. In particular, the biomarkers in CON showed significant increases or no changes, despite participating in aerobic dancing. Specifically, the TNF- α ($p = 0.005$) and CRP ($p = 0.002$) in CON significantly increased after 8 weeks. However, both variables in EMSG significantly decreased. These results revealed significant differences between the groups by the end of the experiment. Meanwhile, the reason that there was negative or no change in the variables of CON is also thought to be due to the fact that the diet was not controlled and configured to cover a small range of dancing activities because of considering their degenerative joints.

When screening any cancer, the CEA is widely investigated [33-35]. CEA, an oncofetal glycoprotein, is overexpressed in adenocarcinomas, and is thus widely used as a tumor marker. CEA may be involved in the release of pro-inflammatory cytokines, probably by stimulating monocytes and macrophages [33] and in the release of endothelial adhesion molecules [34]. Therefore, CEA may contribute to the development of cancer. In addition, this action of CEA may also cause atherosclerosis and cardiovascular disease, as well as the metastasis of malignant cells [36,37]. CEA, for which aging is a major contributing factor, showed an increasing tendency in the CON of this study over the course of the 8-week program. Lee et al. [38] reported that CEA concentrations could be associated with metabolic disturbances and cardiovascular disease, as well as cancer. Several types of diseases are linked to higher levels of the CEA of biomarker components [36,37,39,40]. IL-6 also has many roles essential to the regulation of the immune response, hematopoiesis, and bone resorption. It is involved not only in the hepatic acute phase response, but also in adipose tissue metabolism and lipoprotein lipase activity [41]. The overproduction of IL-6, a pro-inflammatory cytokine, is associated with a spectrum of age-related conditions, including cardiovascular disease, osteoporosis, arthritis, type 2 diabetes mellitus, certain cancers, periodontal disease, frailty, and functional decline [42]. Meanwhile, CRP is a major acute-phase reactant primarily synthesized in the liver hepatocytes. It is composed of five identical, 21,500-molecular-weight subunits. CRP mediates activities associated with pre-immune nonspecific host resistance. It shows the strongest association with cardiovascular events.

It is no exaggeration to say that most of the diseases associated with the aging process and obesity or caused by both processes are associated with vascular disease, cardiovascular disease, and tumorigenic diseases. Such chronic degenerative diseases can be prevented and treated by medication, surgery, and healthy lifestyles, but above all, it has been reported that exercise habits and regular exercise are more necessary for sustaining a healthy life [24]. Regular physical activity for

elderly people can help them to maintain or sustain a healthy body weight, enhance muscle mass, and strengthen their immune system. According to the above theories, Rogers et al. [43] and McTiernan [44] showed that the modulation of energy balance by increasing physical activity can contribute to the reduction of cancer risks through numerous epidemiological reviews. Kobayashi et al. [45] also suggested that high levels of moderate and vigorous physical activity during adolescence may contribute to a lower risk of breast cancer in both pre- and post-menopausal women. Therefore, an emerging body of evidence suggests a strong inverse association between higher levels of fitness, or greater amounts of exercise, and cancer occurrence, or mortality [46]. However, there are some discrepancies regarding the exercise volume (intensity, time, and frequency) required for the prevention of cancer [24].

Many studies have reported that exercise can inhibit cancer development through the enhancement of immunity, or promote cancer through the suppression of immunity [45,47]. Banerjee et al. [48] suggested that EMS was capable of eliciting a cardiovascular exercise response without loading the limbs or joints and inducing rhythmical contractions in the leg muscles. According to their results, they demonstrated significant improvements in peak oxygen consumption, walking distance, and quadricep strength after 6 weeks. In their findings, the EMS was only attached to specific parts of the body and the tolerance strength was only about 50%. In other words, most subjects in their study selected an impulse intensity that was consistent with the lower end of the training intensity zone. It is important that the exercise intensity is high enough to improve body composition and produce benefits of metabolism-related exercises [49]. This study combined aerobic dancing with EMS, which used progressive impulse intensities. The use of EMS has been reported to be an effective complementary method to conventional exercise programs [50,51]. Although the favorable effects of EMS on neuromuscular parameters have been previously shown in elderly subjects [52,53], the effects of EMS on body composition and cytokines in elderly obese women are confirmed by the results of this study. Several studies have found that although the various impulse intensities of EMS showed improved tendencies in adipokine profiles, there was an increased effectiveness when using higher electrical impulse intensities [54,55]. One researcher reported that adipokines fluctuated irregularly when low and moderate EMS impulse intensities were applied during the experimental period. However, RSTN decreased regularly and sequentially for 6 weeks when a high EMS impulse intensity was applied [20]. The results of other research studies also show a decrease in RSTN due to exercise [19,56-59]. In other words, the stimulation that was applied in this study with impulses greater than a moderate intensity can be regarded as an exercise stimulus that activates lipid metabolism reported by other researchers [60,61]. Previous researchers have reported that a higher level of physical activity is associated with an improved lipoprotein profile and increased fat oxidation [61]. Considering the results of this study, the effects of EMS with aerobic dancing performed for 40 minutes per session with low to high impulse intensities provided benefits at the end of the experiment.

Ultimately, we suggest that EMS suits available for aerobic dancing can contribute to oxidative fat metabolism and cytokine reduction by effectively stimulating the muscles of obese elderly women, who may be less active. In other words, the effects of the progressive application of EMS intensities in this study were similar to the results of research [62] on endurance exercise training that showed increased lipid oxidation leading to positive effects on cytokines related to inflammatory substances and metabolic indicators, as well as body composition, in obese women.

5. Conclusions

Based on the confirmed homogeneity of this study, the results suggest that the progressive electrical impulse of EMS for 8 weeks may improve body composition and tumor- or inflammation-related cytokines in elderly obese women. Since most of the biochemical variables did not show positive changes in most of the results of this study, we suggest that the long-term aerobic dancing and EMS program should be used to find changes in body composition and biomarkers. In particular, in order to observe changes in biomarkers that are harmful to health, it is necessary to conduct research for a longer experimental period or greater exercise frequency.

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References

- Ettinger, W.H.Jr.; Fried, L.P.; Harris, T.; Shemanski, L.; Schulz, R.; Robbins, J. Self-reported causes of physical disability in older people: the Cardiovascular Health Study. CHS Collaborative Research Group. *J. Am. Geriatr. Soc.* **1994**, *42*, 1035-1044. doi: 10.1111/j.1532-5415.1994.tb06206.x
- Haslam, D.W.; James, W.P. Obesity. *Lancet* **2005**, *366*, 1197-1209. doi: 10.1016/S0140-6736(05)67483-1.
- Keddie, A.M. Associations between severe obesity and depression: results from the National Health and Nutrition Examination Survey, 2005-2006. *Prev. Chronic Dis.* **2011**, *8*, A57.
- Soon, H.K.; Saad, H.A.; Taib, M.N.; Rahman, H.A.; Mun, C.Y. Effects of combined physical activity and dietary intervention on obesity and metabolic parameters in adults with abdominal obesity. *Southeast Asian J. Trop. Med. Public Health* **2013**, *44*, 295-308.
- Davidson, L.E.; Tucker, L.; Peterson, T. Physical activity changes predict abdominal fat change in midlife women. *J. Phys. Act. Health*, **2010**, *7*, 316-322.
- Kellgren, J.H. Osteoarthritis in Patients and Populations. *Br. Med. J.* **1961**, *2*, 1-6. doi: 10.1136/bmj.2.5243.1
- Baker, K.R.; Nelson, M.E.; Felson, D.T.; Layne, J.E.; Sarno, R.; Roubenoff, R. The efficacy of home based progressive strength training in older adults with knee osteoarthritis: A randomized controlled trial. *J. Rheumatol.* **2001**, *28*, 1655-1665.
- Kovar, P.A.; Allegrante, J.P.; MacKenzie, C.R.; Peterson, M.G.; Gutin, B.; Charlson, M.E. Supervised fitness walking in patients with osteoarthritis of the knee. A randomized, controlled trial. *Ann. Intern. Med.* **1992**, *116*, 529-534. doi: 10.7326/0003-4819-116-7-529
- Kemmler, W.; Schliffka, R.; Mayhew, J.L.; von Stengel, S. Effects of whole-body electromyostimulation on resting metabolic rate, body composition, and maximum strength in postmenopausal women: the Training and ElectroStimulation Trial. *J. Strength Cond. Res.* **2010**, *24*, 1880-1887. doi: 10.1519/JSC.0b013e3181ddaee.
- Lexell, J.; Taylor, C.C.; Sjöström, M. What is the cause of the ageing atrophy? Total number, size and proportion of different fiber types studied in whole vastus lateralis muscle from 15-to 83-year-old men. *J. Neurol. Sci.* **1988**, *84*, 275-294. doi: 10.1016/0022-510x(88)90132-3.
- Porcari, J.P.; McLean, K.P.; Foster, C.; Kernozek, T.; Crenshaw, B.; Swenson, C. Effects of electrical muscle stimulation on body composition, muscle strength, and physical appearance. *J. Strength Cond. Res.* **2002**, *16*, 165-172.
- Enoka, R.M. Activation order of motor axons in electrically evoked contractions. *Muscle Nerve* **2002**, *25*, 763-764. doi: 10.1002/mus.10117.
- Gondin, J.; Guette, M.; Ballay, Y.; Martin, A. Electromyostimulation training effects on neural drive and muscle architecture. *Med. Sci. Sports Exerc.* **2005**, *37*, 1291-1299. doi: 10.1249/01.mss.0000175090.49048.41.
- Bailey, H.R. Localized tissue reduction. *Med. J. Aust.* **1976**, *1*, 780-781.
- Gondin, J.; Cozzone, P.J.; Bendahan, D. Is high-frequency neuromuscular electrical stimulation a suitable tool for muscle performance improvement in both healthy humans and athletes? *Eur. J. Appl. Physiol.* **2011**, *111*, 2473-2487. doi: 10.1007/s00421-011-2101-2.
- Thakral, G.; Lafontaine, J.; Najafi, B.; Talal, T.K.; Kim, P.; Lavery, L.A. Electrical stimulation to accelerate wound healing. *Diabet. Foot Ankle* **2013**, *4*. doi: 10.3402/dfa.v4i0.22081.
- Jee, Y.S. The efficacy and safety of whole-body electromyostimulation in applying to human body: based from graded exercise test. *J. Exerc. Rehabil.* **2018**, *14*, 49-57. doi: 10.12965/jer.1836022.011.
- Maffiuletti, N.A. Physiological and methodological considerations for the use of neuromuscular electrical stimulation. *Eur. J. Appl. Physiol.* **2010**, *110*, 223-234. doi: 10.1007/s00421-010-1502-y.
- Jee, Y.S. The effect of high-impulse electromyostimulation on adipokine profiles, body composition and strength: A pilot study. *Isokin. Exerc. Sci.* **2019**, *27*, 163-176. doi 10.3233/IES-183201.

20. Kemmler, W.; Teschler, M.; Weißenfels, A.; Bebenek, M.; Fröhlich, M.; Kohl, M.; von Stengel, S. Effects of whole-body electromyostimulation versus high-intensity resistance exercise on body composition and strength: A randomized controlled study. *Evid. Based Complement Alternat. Med.* **2016**, 9236809. doi: 10.1155/2016/9236809.
21. von Stengel, S.; Bebenek, M.; Engelke, K.; Kemmler, W. Whole-Body electromyostimulation to fight osteopenia in elderly females: The randomized controlled Training and Electrostimulation Trial (TEST-III). *J. Osteoporos.* **2015**, 2015:643520. doi: 10.1155/2015/643520.
22. Zheng, A.; Sakari, R.; Cheng, S.M.; Hietikko, A.; Moilanen, P.; Timonen, J.; Fagerlund, K.M.; Kärkkäinen, M.; Alèn, M.; Cheng, S. Effects of a low-frequency sound wave therapy programme on functional capacity, blood circulation and bone metabolism in frail old men and women. *Clin. Rehabil.* **2009**, 23, 897-908. doi: 10.1177/0269215509337273.
23. Cha, J.Y.; Kim, J.H.; Hong, J.; Choi, Y.T.; Kim, M.H.; Cho, J.H.; Ko, I.G.; Jee, Y.S. A 12-week rehabilitation program improves body composition, pain sensation, and internal/external torques of baseball pitchers with shoulder impingement symptom. *J. Exerc. Rehabil.* **2014**, 10, 35-44. doi: 10.12965/jer.140087.
24. Ko, I.G.; Park, E.M.; Choi, H.J.; Yoo, J.; Lee, J.K.; Jee, Y.S. Proper exercise decreases plasma carcinoembryonic antigen levels with the improvement of body condition in elderly women. *Tohoku J. Exp. Med.* **2014**, 233, 17-23. doi: 10.1620/tjem.233.17.
25. Filipovic, A.; Kleinöder, H.; Dörmann, D.; Mester, J. Electromyostimulation--a systematic review of the influence of training regimens and stimulation parameters on effectiveness in electromyostimulation training of selected strength parameters. *J. Strength Cond. Res.* **2011**, 25, 3218-3238. doi: 10.1519/JSC.0b013e318212e3ce.
26. Kemmler, W.; Froehlich, M.; von Stengel, S.; Kleinöder, H. Whole-Body Electromyostimulation - The need for common sense! Rationale and guideline for a safe and effective training. *Dtsch. Z. Sportmed.* **2016**, 67, 218-221. doi: 10.5960/dzsm.2016.246.
27. Lam, H.; Qin, Y.X. The effects of frequency-dependent dynamic muscle stimulation on inhibition of trabecular bone loss in a disuse model. *Bone* **2008**, 43, 1093-1100. doi: 10.1016/j.bone.2008.07.253.
28. Bily, W.; Trimmel, L.; Mödlin, M.; Kaider, A.; Kern, H. Training program and additional electric muscle stimulation for patellofemoral pain syndrome: a pilot study. *Arch. Phys. Med. Rehabil.* **2008**, 89, 1230-1236. doi: 10.1016/j.apmr.2007.10.048.
29. Kraemer, W.J.; Ratamess, N.A. Fundamentals of resistance training: Progression and exercise prescription. *Med. Sci. Sports Exerc.* **2004**, 36, 674-688. doi: 10.1249/01.mss.0000121945.36635.61.
30. Levinger, I.; Goodman, C.; Hare, D.L.; Jerums, G.; Toia, D.; Selig, S. The reliability of the 1 RM strength test for untrained middle-aged individuals. *J. Sci. Med. Sport.* **2009**, 12, 310-316. doi: 10.1016/j.jsams.2007.10.007.
31. Anderson, A.G.; Murphy, M.H.; Murtagh, E.; Nevill, A. An 8-week randomized controlled trial on the effects of brisk walking, and brisk walking with abdominal electrical muscle stimulation on anthropometric, body composition, and self-perception measures in sedentary adult women. *Psychol. Sport Exerc.* **2006**, 7, 437-451. doi.org/10.1016/j.psychsport.2006.04.003.
32. Porcari, J.P.; Miller, J.; Cornwell, K.; Foster, C.; Gibson, M.; McLean, K.; Kernozek, T. The effects of neuromuscular electrical stimulation training on abdominal strength, endurance, and selected anthropometric measures. *J. Sports Sci. Med.* **2005**, 4, 66-75.
33. Ganguly, A.; Yeltsin, E.; Robbins, J. Identification of a carcinoembryonic antigen binding protein on monocytes. *Biochem. Biophys. Res. Commun.* **2003**, 311, 319-323. doi: 10.1016/j.bbrc.2003.09.213.
34. Aarons, C.B.; Bajenova, O.; Andrews, C.; Heydrick, S.; Bushell, K.N.; Reed, K.L.; Thomas, P.; Becker, J.M.; Stucchi, A.F. Carcinoembryonic antigen-stimulated THP-1 macrophages activate endothelial cells and increase cell-cell adhesion of colorectal cancer cells. *Clin. Exp. Metastasis* **2007**, 24, 201-209. doi: 10.1007/s10585-007-9069-7.
35. Ishizaka, N.; Ishizaka, Y.; Toda, E.; Koike, K.; Yamakado, M.; Nagai, R. Are serum carcinoembryonic antigen levels associated with carotid atherosclerosis in Japanese men? *Arterioscler. Thromb. Vasc. Biol.* **2008**, 28, 160-165. doi: 10.1161/ATVBAHA.107.155465.
36. Piro, M.; Giubilato, G.; Pinnelli, M.; Giordano Sciacca, P.; Biasucci, L.M. Endothelium and inflammation. *Panminerva. Med.* **2005**, 47, 75-80.
37. Steeg, P.S. Tumor metastasis: mechanistic insights and clinical challenges. *Nat. Med.* **2006**, 12, 895-904. doi: 10.1038/nm1469.

38. Lee, J.W.; Park, K.D.; Im, J.A.; Hwang, H.J.; Kim, S.H. Serum carcinoembryonic antigen is associated with metabolic syndrome in female Korean non-smokers. *Clin. Chim. Acta.* **2011**, *412*, 527-530. doi: 10.1016/j.cca.2010.11.033.
39. Johnson, P.J. The role of serum alpha-fetoprotein estimation in the diagnosis and management of hepatocellular carcinoma. *Clin. Liver Dis.* **2001**, *5*, 145-159.
40. Stray-Pedersen, A.; Borresen-Dale, A.L.; Paus, E.; Lindman, C.R.; Burgers, T.; Abrahamsen, T.G. Alpha fetoprotein is increasing with age in ataxia-telangiectasia. *Eur. J. Paediatr. Neurol.* **2007**, *11*, 375-380. doi: 10.1016/j.ejpn.2007.04.001.
41. Kiecolt-Glaser, J.K.; Preacher, K.J.; MacCullum, R.C.; Atkinson, C.; Malarkey, W.B.; Glaser, R. Chronic stress and age-related increases in the proinflammatory cytokine IL-6. *Proc. Nat. Acad. Sci.* **2003**, *100*, 9090-9095. doi: 10.1073/pnas.1531903100.
42. Ferrari, S.L.; Ahn-Luong, L.; Garnerio, P.; Humphries, S.E.; Greenspan, S.L. Two promoter polymorphisms regulating interleukin-6 gene expression are associated with circulating levels of C-reactive protein and markers of bone resorption in postmenopausal women. *J. Clin. Endocr. Metab.* **2003**, *88*, 255-259. doi: 10.1210/jc.2002-020092.
43. Rogers, C.J.; Colbert, L.H.; Greiner, J.W.; Perkins, S.N.; Hursting, S.D. Physical activity and cancer prevention: pathways and targets for intervention. *Sports Med.* **2008**, *38*, 271-296. doi: 10.2165/00007256-200838040-00002.
44. McTiernan, A. Mechanisms linking physical activity with cancer. *Nat. Rev. Cancer.* **2008**, *8*, 205-211. doi: 10.1038/nrc2325.
45. Kobayashi, L.C.; Janssen, I.; Richardson, H.; Lai, A.S.; Spinelli, J.J.; Aronson, K.J. Moderate-to-vigorous intensity physical activity across the life course and risk of pre- and post-menopausal breast cancer. *Breast Cancer Res. Treat.* **2013**, *139*, 851-861. doi: 10.1007/s10549-013-2596-9.
46. Winningham, M.L.; MacVicar, M.G.; Burke, C.A. Exercise for cancer patients: guidelines and precautions. *Phys. Sportsmed.* **1986**, *14*, 125-134. doi: 10.1080/00913847.1986.11709201.
47. Shephard, R.J.; Verde, T.J.; Thomas, S.G.; Shek, P. Physical activity and the immune system. *Can. J. Sport Sci.* **1991**, *16*, 169-185.
48. Banerjee, P.; Caulfield, B.; Crowe, L.; Clark, A. Prolonged electrical muscle stimulation exercise improves strength and aerobic capacity in healthy sedentary adults. *J. Appl. Physiol.* (1985) **2005**, *99*, 2307-2311. doi: 10.1152/japplphysiol.00891.2004.
49. Garber, C.E.; Blissmer, B.; Deschenes, M.R.; Franklin, B.A.; Lamonte, M.J.; Lee, I.M.; Nieman, D.C.; Swain, D.P. American College of Sports Medicine position stand. Quantity and quality of exercise for developing and maintaining cardiorespiratory, musculoskeletal, and neuromotor fitness in apparently healthy adults: guidance for prescribing exercise. *Med. Sci. Sports Exerc.* **2011**, *43*, 1334-1359. doi: 10.1249/MSS.0b013e318213febf.
50. Amiridis, I.; Arabatzis, F.; Violaris, P.; Stavropoulos, E.; Hatzitaki, V. Static balance improvement in elderly after dorsiflexors electrostimulation training. *Eur. J. Appl. Physiol.* **2005**, *94*, 424-433. doi: 10.1007/s00421-005-1326-3.
51. Paillard, T.; Lafont, C.; Soulat, J.M.; Montoya, R.; Costes-Salon, M.C.; Dupui, P. Short-term effects of electrical stimulation superimposed on muscular voluntary contraction in postural control in elderly women. *J. Strength Cond. Res.* **2005**, *19*, 640-646. doi: 10.1519/15354.1.
52. Paillard, T.; Lafont, C.; Costes-Salon, M.C.; Dupui, P. Comparison between three strength development methods on body composition in healthy elderly women. *J. Nutr. Health Aging.* **2003**, *7*, 117-119.
53. Vivodtzev, I.; Pépin, J.L.; Vottero, G.; Mayer, V.; Porsin, B.; Lévy, P.; Wuyam, B. Improvement in quadriceps strength and dyspnea in daily tasks after 1 month of electrical stimulation in severely deconditioned and malnourished COPD. *Chest* **2006**, *129*, 1540-1548. doi: 10.1378/chest.129.6.1540.
54. Esposito, K.; Pontillo, A.; Di Palo, C.; Giugliano, G.; Masella, M.; Marfella, R.; Giugliano, D. Effect of weight loss and lifestyle changes on vascular inflammatory markers in obese women. *JAMA* **2003**, *289*, 1799-1804. doi: 10.1001/jama.289.14.1799.
55. Pittas, A.G.; Joseph, N.A.; Greenberg, A.S. Adipocytokines and insulin resistance. *J. Clin. Endocrinol. Metab.* **2004**, *89*, 447-452. doi.org/10.1210/jc.2003-031005.
56. Choi, K.M.; Kim, J.H.; Cho, G.H.; Baik, S.H.; Park, H.S.; Kim, S.M. Effect of exercise training on plasma visfatin and eotaxin levels. *Eur. J. Endocrinol.* **2007**, *157*, 437-442. doi: 10.1530/EJE-07-0127.

57. Haider, D.G.; Schindler, I.C.; Schaller, G.; Prager, G.; Wolzt, M.; Ludvik, B. Increased plasma visfatin concentrations in morbidly obese subjects are reduced after gastric banding. *J. Clin. Endocrinol. Metab.* **2006**, *91*, 1578-1581. doi: 10.1210/jc.2005-2248.
58. Moon, B.; Kwan, J.J.; Duddy, N.; Sweeney, G.; Begum, N. Resistin inhibits glucose uptake in L6 cells independently of changes in insulin signaling and GLUT4 translocation. *Am. J. Physiol. Endocrinol. Metab.* **2003**, *285*, 106-115. doi: 10.1152/ajpendo.00457.2002.
59. Prestes, J.; Shiguemoto, G.; Botero, J.P.; Frollini, A.; Dias, R.; Leite, R.; Pereira, G.; Magosso, R.; Baldissera, V.; Cavaglieri, C.; Perez, S. Effects of resistance training on resistin, leptin, cytokines, and muscle force in elderly post-menopausal woman. *J. Sports Sci.* **2009**, *27*, 1607-1615. doi: 10.1080/02640410903352923.
60. Kraus, W.E.; Houmard, J.A.; Duscha, B.D.; Knetzger, K.J.; Wharton, M.B.; McCartney, J.S.; Bales, C.W.; Henes, S.; Samsa, G.P.; Otvos, J.D.; Kulkarni, K.R.; Slentz, C.A. Effects of the amount and intensity of exercise on plasma lipoproteins. *N. Engl. J. Med.* **2002**, *347*, 1483-1492. doi: 10.1056/NEJMoa020194.
61. Schrauwen, P.; van Aggel-Leijssen, D.P.; Hul, G.; Wagenmakers, A.J.; Vidal, H.; Saris, W.H.; van Baak, M.A. The effect of a 3-month low-intensity endurance training program on fat oxidation and acetyl-CoA carboxylase-2 expression. *Diabetes* **2002**, *51*, 2220-2226. doi: 10.2337/diabetes.51.7.2220.
62. Samjoo, I.A.; Safdar, A.; Hamadeh, M.J.; Raha, S.; Tarnopolsky, M.A. The effect of endurance exercise on both skeletal muscle and systemic oxidative stress in previously sedentary obese men. *Nutr. Diabetes.* **2013**, *3*, e88. doi: 10.1038/nutd.2013.30.