

Original Article

The Effect of Aerobic Training and Vitamin D Supplementation on Blood Pressure in Elderly Women with Nonalcoholic Fatty Liver and Vitamin D Deficiency

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<u>Abstract</u>

Background & Objective: The aim of this study was to investigate the effects of aerobic training (AT) and vitamin D (Vit D) supplementation on cardiovascular variables in elderly women with nonalcoholic fatty liver disease (NAFLD) and vitamin D deficiency.

Materials & Methods: A total of forty women (60-65 years) with NAFLD (second or third grade) and vitamin D deficiency were recruited for the study. Participants were randomly assigned to four AT+Vit D, AT, Vit D, and control (C) groups and were allocated to receive either vitamin D (~50000 IU • week for eight weeks) or placebo. The aerobic training protocol consisted of 40-60 minutes of aerobic training at 60% - 75% of HRmax, 3 times a week for eight weeks. Anthropometric indices along with systolic blood pressure (SBP), diastolic blood pressure (DBP), and heart rate (HR) were measured and mean of arterial pressure (MAP) was calculated.

Results: AT+Vit D, Vit D and AT groups demonstrated a significant decrease in SBP, DBP, and HR, (P < 0.05) while, C group showed a significant increase in the mentioned variables (P < 0.05). Also after 8 weeks, AT + Vit D and AT groups showed significant decrease in the mean of MAP (P = 0.001; P = 0.018). Compared to the control group, significant differences in the SBP, DBP, and HR were observed in all groups. The mean SBP was significantly lower in AT+Vit D group comparing with AT, Vit D, and C groups. Also, no significant differences were observed between AT and Vit D groups in all variables.

Conclusion: AT developed significant improvement in cardiovascular variables indicating SBP, DBP, MAP, and HR in elderly women with NAFLD and vitamin D deficiency. Meanwhile, AT combined with Vit D decreased SBP more significantly that might be incorporated in the management programs of the patients suffering from NAFLD in order to augment improvement in their blood pressure.

Keywords: Exercise, Vitamin D, Blood Pressure, NAFLD

Introduction

Currently, the elderly represent a large and growing portion of the population (1). According to the 2011 census in Iran, 7.7% of the total population of the country aged over 65, which is projected to increase by 25 to 35% by

2031 (2). The older one becomes, the higher the prevalence of chronic diseases like Non-Alcoholic Fatty Liver Disease (NAFLD) especially in women (3). NAFLD is the leading cause of chronic liver disease, with increased prevalence up to 20–30% worldwide (4). It is characterized by the accumulation of fat more than 5% of the hepatocytes in the liver (5), which encompasses various conditions ranging from simple steatosis, nonalcoholic steatohepatitis, to

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cirrhosis (6). Recent studies suggested that NAFLD is an independent risk factor for hypertension, the major cause of Cardiovascular disease (CVD), and its progression is associated with the development of hypertension (7). The deficiency of vitamin D has been implicated as the predictive factor for the occurrence of NAFLD (8, 9). Furthermore, Vitamin D deficiency is known to be associated with hypertension, obesity, atherosclerosis, type 2 diabetes mellitus, oxidative stress, cancer and CVD (10-13). Moreover, studies suggest that a low level of vitamin D is associated with high blood pressure. They also revealed that a high chance of developing hypertension is correlated with the increased risk of mortality and high levels of cardiovascular events in the future (12, 14, 15). It has been reported that vitamin D regulates calcium and bone homeostasis, and its receptor is present in many tissues, such as vascular endothelium and myocardial tissue. This indicates the biological role and function of vitamin D, which can suppress renin production by affecting the juxtaglomerular system (16, 17). Vitamin D supplementation can lead to the suppression of parathyroid hormone and proinflammatory cytokine production. In addition, there are possible pathophysiological pathways that show the effects of vitamin D on vascular protection against hypertension (18). A number of previous studies associated with NLFAD have reported that regular aerobic training reduces hepatic fat content in NLFAD patients, and helps control hypertension (19, 20). Among the principal physiological benefits of physical training in hypertensive patients, there are reductions in heart rate (HR) and peripheral vascular resistance (21). These factors are related to the reduction of systolic and diastolic blood pressure (SBP and DBP) (22). In addition to the improvements in blood pressure, aerobic training is associated with a reduction of serum cholesterol, which often is above the reference values in hypertensive subjects (23). Generally, there are contradictory results about the effect of aerobic training on hypertension. Some studies indicate beneficial effects (24, 25) and others report no significant changes (26, 27). In addition, limited amount of studies have been conducted to show the effects of vitamin D supplementation on blood pressure in NAFLD. On the other hand, to our knowledge, there is no information in the literature comparing the separate and interactional effects of aerobic

training and vitamin D supplementation. Since the decrease in physical activity level and the high prevalence of vitamin D deficiency and NAFLD are more significant in elderly women, the present study intends to investigate the effects of aerobic training and vitamin D supplementation on hypertension in elderly women with NAFLD and vitamin D deficiency.

Materials & Methods

This is a semi-experimental research with pretest and post-test design. The target population of this study was elderly women (60 to 65 years) of Kermanshah with NAFLD and vitamin D deficiency. Forty Subjects were divided into four groups including: Aerobic training (AT, n=10); Vitamin D supplementation (Vit D, n=10); Aerobic training with vitamin D supplements (AT+Vit D, n=10); and Control (C, n=10). Inclusion criteria includes: ultrasound (US) confirmation of being in second or third grade of NAFLD and certified by an internist, 25-OHD levels between 10 and 20 ng/ml, not having an specific diet and regular exercise program in the past year. Exclusion criteria includes: significant consumption of alcohol (more than 20 gr per day) (28, 29), presence of other liver diseases (B and C hepatitis), other disorders such as autoimmune hepatitis, joint disease, celiac and Wilson disease, CVD, kidney failure, hypothyroidism, also surgical treatment of obesity or severe weight loss, and the presence of any other chronic disease or skeletal disorders (29).

Three days prior to the start of the study, explanations about research conduction were provided for subjects and consent forms were signed by all 40 subjects. Also, PARQ, 3-day dietary questionnaire, nutritional questionnaires including food history, food frequency, habits and dietary behaviors, and Health History Questionnaire (HHQ) were completed. On the first day, height was measured with an accuracy of 0.5 cm. Body weight (BW, with an accuracy of 0.1 gr), BMI, body fat percentage (BFP), were obtained using the INBODY test in the fasting state. Also waist-hip ratio (WHR) was measured by dividing waist circumference by hip circumference according to WHO criteria.

Ethical considerations

The present study was approved by the Committee of Ethics in Research at Kermanshah University of Medical Sciences and registered in the Iranian Clinical Trial Registration Center under the code IR.KUMS.REC.1397.1059.

Before the study, informed written consent was obtained from all participants after a comprehensive oral and written explanation of the study was provided to them. All patients were allowed to leave the study at any point voluntarily. The demographic data of the participants were kept confidential. Also, all the clinical and paraclinical tests of the study were free of charge, with no costs to the patients.

Training program

The aerobic training program was executed 3 days/week for 8 weeks based on the ACSM's recommendation. Each session lasted 40-60 min consisting 10 minutes of warming-up, 20 to 40 minutes of aerobic exercise and 10 minutes of cooling-down. In the first week, subjects performed 20-min of aerobic exercise including: walking, jogging and running at 60% HRmax. The initial duration and intensity was gradually progressed to a maximum duration of 40 min and 75% HRmax respectively. The training program was performed with 60-75% of the individual HRmax (using Karvonen Formula) and 10-13 of RPE in 6-20 Borg scale (Table 1) (30).

caffeinated products and exercising, 30 min prior to the BP measurement. Throughout assessment, patients remained in a relaxed seated state for 10 min. The cuff was placed on the left arm for all patients and BP was recorded according to standard guidelines (31). A second recording was also taken after 2 min on the same arm. If the measurements had a difference of $\geq 5 \text{ mmHg}$ of BP, further recording were obtained until there were 2 consecutive stable measurements. Final recording was considered as an average of the 2 stable measurements obtained. Measurement took place each day between 8.00 a.m. and 10.00 a.m. in accordance with the protocol of the American Heart Association (31) Mean arterial pressure (MAP) was calculated as DBP + [0.333 (SBP - DBP)].

Data Analysis

Data are presented as means \pm SD. Prior to analysis, data normality was checked with the Shapiro-Wilk test. In order to compare the mean SBP and DBP between and within groups, ANOVA and t-test were used, respectively. Tukey's post hoc test was used if significant

Variables		- <u>9</u>			Weeks				
	First	Second	Third	Fourth	Fifth	Sixth	Seventh	Eight	
Intensity (HR _{max})	60%	60%	65%	65%	70%	70%	75%	75%	
Time (min)	20	25	25	30	30	35	35	40	
Borg scale	10	10	11	11	11	12	12	13	

Table 1. Exercise protocol program

Vitamin D supplementation

In this study, both AT+Vit D group and Vit D group received 50000 IU of vitamin D supplement once per week at the beginning of the week, made by the Zahravi Pharmaceutical Company in Iran. The C and AT groups also received a weekly placebo (a paraffin made by the Zahravi Pharmaceutical Company, Iran) with the same shape, color, smell and tastes to the vitamin D supplements pills, over a period of 8 weeks.

Blood pressure measurements

SBP and DBP were measured by standard manual sphygmomanometer. Participants were strictly prohibited from consuming any

differences were found. The SPSS software version 21 was used at a significance level of P< 0.05.

Results

The anthropometric indices of groups are presented in Table 2. There was no statistically significant difference between the four groups at the beginning of the study in weight, BMI, body fat and WHR.

Based on the results of t-test, there were significant differences in the mean of SBP, DBP, and HR between the pre-test and post-test conditions. After 8 weeks SBP, DBP and HR significantly decreased in AT+Vit D, AT, Vit D groups while, in the control group a significant increase in SBP, DBP and HR was observed (Table 3). The results also showed that after 8 weeks, the mean of MAP in AT + Vit D and AT groups decreased significantly (P = 0.001; P = 0.018, respectively) however, there was no

Table 2. Mean \pm SD and frequency distribution of the anthropometric indices before the intervention among
the groups

Variables	AT+Vit D (n = 10)	AT (n = 10)	Vit D (n=10)	C (n = 10)	P Value
Age (years)	63.10 ±2.37	62.60 ±1.89	61.30 ± 1.41	62 ±1.88	0.16
Height (cm)	157.10 ± 5.25	160 ± 5.45	158.30±4.59	159 ±5.79	0.11
Weight (kg)	87.40 ±4.64	85.80 ± 3.35	86.10 ±3.14	87.50 ±4.19	0.679
BMI (kg/h ²)	35.55 ±3.55	33.57 ± 1.92	34.40±1.78	34.65 ± 1.70	0.332
Body Fat (%)	44 ±3.36	43.10 ±3.41	41.20±3.15	42±3.59	0.284
WHR	94.50 ±2.54	95.20 ± 3.79	95.80±2.78	95.70 ±3.16	0.778

AT+Vit D group, Aerobic training and vitamin D supplementation, **AT group,** Aerobic training; **Vit D group,** vitamin D supplementation; **C group,** that had neither aerobic training nor vitamin D supplementation. P values are calculated using one-way analysis of variance test followed by Tukey's Post hoc Test.

Table 3. Comparison of Mean \pm SD of cardiovascular variables within and between the groups under the study

Variables	AT + Vit D (n = 10)	AT (n = 10)	Vit D (n=10)	C (n = 10)	P Value a
SBP (mmHg)					
Before	129.60 ± 2.50	130.30 ± 1.56	128.90 ± 1.79	129.40 ± 1.77	0.454
After	125.40 ± 1.34	127.40 ± 1.64	127.80 ± 1.75	131.10 ± 1.37	0.001 ¥
P Value _b	P=0.001 *	P=0.002 *	P=0.001 *	P=0.002 *	
DBP (mmHg)					
Before	84.40±1.34	83.40±1.42	84.20±1.03	84.10±1.28	0.343
After	82.90±1.37	82.40±1.40	83.30±1.41	85.30±0.94	0.001 ¥
P Value b	P=0.001 *	P=0.023 *	P=0.002 *	P=0.002 *	
MAP (mmHg)					
Before	99.45±1.25	99.01±1.03	99.08±0.94	99.18±1.10	0.821
After	98.55±1.01	98.38±1.15	99.01±0.83	99.35±1.07	0.155
P Value b	0.001 *	0.018 *	0.343	0.096	
HR (mmHg)					
Before	85 ±1.33	83.40 ±1.42	84.20 ± 1.03	84.10 ± 1.19	0.059
After	79.60±1.42	79.70±1.15	80.70±1.63	86.40 ± 0.84	0.001¥
P Value b	P=0.001 *	P=0.001 *	P=0.001 *	P=0.001 *	

AT+Vit D group: Aerobic training and vitamin D supplementation; AT group: Aerobic training; Vit D group: vitamin D supplementation; C group: that had neither aerobic training nor vitamin D supplementation; SBP: systolic blood pressure; DBP: diastolic blood pressure; MAP: mean of arterial pressure; HR: heart rate.

P values with superscript "a" are calculated using one-way analysis of variance test followed by Tukey's post hoc test; superscript letter "b" indicates values were calculated using paired t-test

*Significantly different in comparison with pre and posttest within the groups; ¥ significantly different in comparison with pre and posttest between groups.











Figure 1 (c). Pre-test and post-test value of the MAP among groups

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AT+Vit D group: Aerobic training and vitamin D supplementation; AT group: Aerobic training; Vit D group: vitamin D supplementation; C group: that had neither aerobic training nor vitamin D supplementation; SBP: systolic blood pressure; DBP: diastolic blood pressure; MAP: mean of arterial pressure; HR: heart rate.

*: Significantly different in comparison with pre and posttest groups; \pounds : Significance different between AT+Vit D group compare to AT group in posttest; μ : Significant difference between AT+Vit D group compared to Vit D group in posttest; \pounds : Significant difference between AT+Vit D, AT, and Vit D groups compared to C group.

significant difference in the mean of MAP in Vit D and C groups (P = 0.343; P = 0.096).

The results of one-way ANOVA showed no significant difference in SBP, DBP, MAP and HR between the groups in the pre-test, however in the post-test there was a significant difference in the SBP, DBP and HR between the groups (Table 3). Although, there was no significant difference in the mean of MAP between groups in the post-test (P = 0.155).

The results of Tukey's post hoc test are presented in Figure 1 (a,b,c,d). The lowest levels of SBP, DBP, HR and MAP were in the AT+Vit D group. The results also showed that there was a significant difference in SBP levels between the AT+Vit D group with AT group, Vit D group and control group; However, there was no significant difference between the AT+Vit D group with AT group and Vit D group in the DBP, HR and MAP. The results also showed a significant difference in the SBP, DBP and HR levels between AT+Vit D group, the AT group, and the Vit D group compared to the control group.

Discussion

This study is the first to assess the effect of aerobic training and vitamin D supplementation on hypertension in elderly women with nonalcoholic fatty liver and vitamin D deficiency. Since increased SBP, DBP, HR and MAP are risk factors for the development of CVD in elderly women, thus identifying effective programs in reducing SBP, DBP, HR and MAP has always been a controversial issue. Our study observed high prevalence of hypertension in patients with NAFLD. Based on the results of the studies, after 8 weeks SBP, DBP and HR significantly decreased in AT+Vit D, AT, Vit D groups while, in the control group a significant increase in SBP, DBP and HR were observed.

There are limited consensus in the literature as to how aerobic training affects cardiovascular variables (SBP, DBP, MAP and HR) in elderly women with NAFLAD patients. Most studies indicate a reduction of BP values (32, 33) and HR (34) during rest after aerobic training in this population. However, other authors conducting similar studies did not observe significant

changes in these parameters (26, 27). Among the factors reported, the main contributors to this disagreement seem to be hypertension stage in addition to the intensity, duration, and frequency of the aerobic training program (35). Our results indicated that elderly women had significant reductions in SBP, DBP, MAP and HR after the completion of an 8-week aerobic training program. A similar result for BP was shown by Lima et al (2011) with significant reductions in both systolic $(142.7 \pm 6.3 \text{ vs. } 130.8 \pm 5.8 \text{ mmHg})$ P < 0.001) and diastolic BP (87.0 ± 4.5 vs. 81.9 \pm 4, 3 mmHg, P = 0.002) in just 30 days of aerobic training with three sessions per week of moderate intensity (50-70% HR reserve + HR exercise (36). Hypertension rest) is multifactorial, and several mechanisms may be involved in the decrease in BP and bradycardia induced by aerobic training. The related chronic adaptations include improvement of endothelial function, (37) the reduction of sympathetic nerve activity, increased baroreflex sensitivity, (22) improvement in the hyperinsulinism state, (38) improvement in lipoprotein profile,(38)weight loss, (39) reduced cardiac output, (39) and reduced peripheral vascular resistance (PVR) (40). In the elderly, the main mechanism involved in the reduction of resting blood pressure in response to aerobic physical training seems to be a reduction in PVR (41).

On the other hand, in the view of the bile aciddependent uptake of vitamin D and its hepatic metabolism, it is reasonable to expect an association between vitamin D status and both cholestatic and non-cholestatic chronic liver disease. Recently, a significant correlation between lower 25(OH) vitamin D levels and an increasing stage of fibrosis and severity of necroinflammatory activity was observed in a population with genotype 1 chronic hepatitis C (42). Interestingly, reduced 25(OH) vitamin D levels have also been noted in patients with NAFLD. A study by Targher et al (2007) found strong association between vitamin D a deficiency and NAFLD with a close association of vitamin D levels with the histological severity of hepatic steatosis, necro-inflammation, and fibrosis (8). 25(OH) vitamin D levels were inversely associated with NAFLD independent of hypertension, suggesting that inadequate vitamin D status might contribute to the development and progression of NAFLD (8).

Results of the present study demonstrate that the lowest levels of SBP, DBP, HR and MAP after 2 months were in the AT+Vit D group. Limited studies have been conducted on the relationship between vitamin D intake and hypertension. Martins et al (2007) have shown that low Vitamin D levels are associated with a higher risk of having hypertension (43). Pfeifer et al. showed that 800 IU of Vitamin D3 plus 1200 mg of calcium significantly reduces blood pressure after 8 weeks compared to the group receiving 1200 mg of calcium alone (44) Vitamin D has also been shown to play a critical role in the regulation of renin-angiotensin system and thus has an influence on the regulation of blood pressure (45, 46). It has also been demonstrated that calcium regulating hormones and reninangiotensin system coordinately mediate blood regulation via altering pressure cellular concentrations of sodium and calcium ions. Thus, it is clear that Vitamin D supplementation has a role in the regulation of blood pressure and that it should be supplemented with the antihypertensive drugs to the patients with hypertension.

One of the limitations of our study was that the diagnosis of NAFLD in our patients was based on the presence of hepatic steatosis on ultrasonography and exclusion of other causes of hepatic steatosis and raised ALT without a histological diagnosis. However, in the absence of liver biopsy, the presence of significant fibrosis and the severity of liver disease were evaluated by transient elastography in our patients, which is now an established modality in patients with NAFLD. Even though the number of patients with raised ALT was small in the study, we could demonstrate significant improvement in ALT in the group that was given vitamin D.

Conclusions

In conclusion, our study demonstrates that low physical activity and vitamin D deficiency is common in patients with NAFLD as it is in the general population. Aerobic training and vitamin D supplementation are a suitable intervention leading to an improvement in the cardiovascular variables. In general terms, our results reinforce the recommendations available in the literature about non-pharmacological treatment of hypertension with planned aerobic training programs in the elderly with NAFLD and vitamin D deficiency.

More studies are needed to verify the effectiveness of different physical training

programs and different doses of vitamin D on cardiovascular and metabolic parameters, specifically for the elderly hypertensive population with NAFLD.

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Conflict of Interests

The authors declare no competing interests.

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مقاله پژوهشی

تأثیر تمرینات هوازی و مکملدهی ویتامین D بر فشار خون در زنان سالمند مبتلا به کبد چرب غیرالکلی و کمبود ویتامین D

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چکیدہ

زمینه و هدف: این مطالعه به منظور تعیین تأثیر تمرینات هوازی و مکملدهی ویتامین D بر متغیرهای قلبی-عروقی در زنان سالمند مبتلا به بیماری کبد چرب غیر الکلی (NAFLD) و کمبود ویتامین D انجام گردید.

مواد و روشها: در مجموع ۴۰ نفر از زنان مبتلا به NAFLD (مرحله ۲ و ۳) و کمبود ویتامین D برای مطالعه انتخاب شدند. شرکت کنندگان به صورت تصادفی به ۴ گروه تمرین هوازی+ مکمل دهی ویتامین D (AT + Vit D)، تمرین هوازی (AT)، مکمل دهی ویتامین D (Vit D) و کنترل (C) تقسیم شدند، همچنین دریافت ویتامین D (حدود ۵۰۰۰۰ IU در هفته به مدت هشت هفته) و یا دارونما مشخص گردید. پروتکل تمرین هوازی شامل ۴۰ تا ۶۰ دقیقه تمرین هوازی با شدت ۶۰ تا ۷۵ درصد ضربان قلب بیشینه، ۳ بار در هفته به مدت هشت هفته به مدت هشت هفته بود. شاخص های تن سنجی همراه با فشار خون سیستولیک (SBP)، فشار خون دیاستولیک (DBP) و ضربان قلب (HR) اندازه گیری شد و میانگین فشار شریانی (MAP) محاسبه شد.

نتایج: گروه های Vit D ،AT+Vit D و AT کاهش معنیداری در متغیرهای BPP ،SBP و HE داشتند (P<۰/۰۵)، در حالی که متغیرهای ذکر شده در گروه C به طور معنیداری افزایش یافت (P<۰.۰۵). همچنین پس از ۸ هفته در گروه های AT+Vit D و AT کاهش معنیداری در میانگین MAP (P=۰/۰۱۹؛ P=۰/۰۱۸) مشاهده شد. در همه گروهها در مقایسه با گروه کنترل اختلاف معنیداری در متغیرهای BP، GP و HT مشاهده شد. میانگین SBP در گروه AD+Vit D به طور معنیداری کمتر از گروههای AT ، AT و C بود. همچنین بین گروه های AT و Vit D اختلاف معنیداری مشاهده نشد.

نتیجه گیری: تمرینات هوازی در زنان سالمند مبتلا به NAFLD و کمبود ویتامین D باعث بهبود معنیداری در متغیرهای قلبی-عروقی شامل SBP، MAP ،DBP و HR شد. علاوه بر این، تمرینات هوازی در ترکیب با ویتامین D باعث کاهش بیشتری در SBP شد و میتواند به منظور بهبود فشار خون در برنامهی کنترل بیماران مبتلا به NAFLD گنجانده شود.

كلمات كليدى: فعاليت ورزشى، ويتامين D، بيمارى كبد چرب غير الكلى، فشارخون

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