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ORIGINAL ARTICLE

The Effects of Pilates Training and Vitamin D Supplementation on Lipid Profile and Flexibility in Men with Multiple Sclerosis

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K E Y W O R D S

Body composition;
Flexibility;
Lipid profile;
MS;
Pilates training;
Vitamin D Supplementation

A B S T R A C T

The aim of this study was to investigate the effects of eight weeks of Pilates training and vitamin D supplementation on lipid profile and flexibility in men with MS. For this purpose, 32 people who met the research conditions voluntarily participated in this study as a statistical sample with an age range of 40-45 years and were randomly divided into four groups: supplement, exercise, combined (exercise + supplement) and control groups. One week before the start of the intervention, a pre-test was taken from all four groups, which included the Wells test to measure flexibility and BMI. The training protocol also included Pilates exercises for eight weeks, three times a week on alternate days, with sessions starting at 20 minutes and gradually increasing to one hour. The training load was increased in two ways: increasing the number of repetitions per round and increasing the retention of movement (from the first session, five seconds of maintaining the movement with six repetitions, which increased to one hour, eight seconds of maintaining the movement with 10 repetitions in the eighth week). Descriptive statistics were used in data analysis to calculate the standard deviation, mean, and graph. In the inferential statistics section, the normality of the data distribution was first assessed using the Shapiro-Wilk test, which revealed that the data distribution was normal. Next, the Levene test was used to determine the homogeneity of variances. One-way analysis of variance was used to examine the significance of the difference between the groups, and if significant, the Tukey post hoc test was used to determine the difference between the means of the two groups. The significance level was also considered to be ($P \leq 0.05$). All statistical analyses and graphing were performed using SPSS version 22 software. Eight weeks of Pilates training and vitamin D supplementation had a significant effect on triglycerides (P=0.003), cholesterol (P=0.0001), low-density lipoprotein (P=0.0001), high-density lipoprotein (P=0.0001), body composition (P=0.0001), and flexibility (P=0.0001) in men with MS. According to the results of the study, it can be said that Pilates training combined with vitamin D consumption improves lipid profile indicators and some health-related physical fitness factors.

Introduction

Multiple sclerosis (MS) is a chronic, inflammatory, and autoimmune disease of the central nervous system characterized by the destruction of the myelin sheath. Its global prevalence is estimated at approximately 3.5 million cases, with around 70,000 reported in Iran. The majority of patients are between 20 and 40 years

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of age, and women are two to three times more likely to be affected than men. Common symptoms include fatigue, depression, motor dysfunction, and visual disturbances. Although the exact cause of MS remains unknown, genetic, environmental, and immunological factors are believed to play a role. Recent studies suggest that Pilates exercise, lipid regulation, and vitamin D supplementation may contribute to managing symptoms and disease progression. MS is considered one of the most significant progressive neurological disorders, not only directly impairing neural function but also influencing patients' metabolic status and lifestyle. Exploring the role of exercise, nutrition, and biochemical factors in disease management may provide new strategies to improve patients' quality of life (1). MS affects both the brain and spinal cord, leading to demyelination of nerve cells and the formation of sclerotic plaques, which disrupt the conduction of electrical impulses. The disease primarily targets the white matter of the central nervous system, resulting in inflammation, demyelination, and ultimately sclerosis (scarring) (2).Currently, there is no definitive cure for MS. Available treatments mainly aim to alleviate symptoms or slow disease progression. Therefore, early diagnosis and timely intervention are crucial to preventing severe complications and uncontrolled progression (3).As a chronic condition, MS is associated with persistent symptoms and disabilities, requiring long-term management (4). Fatigue is among the most common and distressing symptoms, significantly impacting patients' psychological well-being. It threatens independence and the ability to participate effectively in family and social life, restricts self-care activities, and limits physical capacity. Patti *et al*. (2003) reported that approximately 80% of MS patients suffer from fatigue, with some even losing their jobs due to reduced functional capacity (5). Given the absence of a definitive cure, patients must rely on treatments that primarily reduce symptoms. Advances in physical education research have highlighted exercise as a complementary therapy alongside pharmacological interventions. Incorporating exercise programs such as Pilates can enhance physical health, improve quality of life, and help patients cope with the variable nature of MS. Moreover, exercise may modulate immune function, potentially preventing further myelin and white matter damage before irreversible axonal injury occurs. These aspects underscore the importance of continued research in this field (6). One of the effective ways to support patients with multiple sclerosis is through physical activity, particularly exercises that require minimal equipment, are easy to perform, enhance flexibility, and do not significantly raise core body temperature (which may be harmful for these patients). Core stability exercises in Pilates are especially beneficial, as they help maintain coordination and balance between body and mind (7). These exercises not only reduce total blood cholesterol but also increase high-density lipoprotein cholesterol (HDL-c) and decrease low-density lipoprotein cholesterol (LDL-c) (8). Moreover, they stimulate cardiac activity, thereby promoting a greater sense of vitality and well-being (9). Mahmodirad *et al*. (2025) reported that Pilates training led to significant reductions in body weight, blood pressure, insulin, total cholesterol, and triglycerides in diabetic individuals, although no changes were observed in HDL or LDL concentrations. Similarly, Shahrjerdi *et al*. (2015) found that Pilates exercises reduced glucose, insulin, and triglyceride levels in diabetic patients, but had no significant effects on plasma leptin, HDL, LDL, or total cholesterol. These findings suggest that Pilates may play a preventive role against cardiovascular risk factors. Over the past decades, numerous non-skeletal diseases associated with vitamin D deficiency have been identified, including osteoporosis, cardiovascular diseases, and metabolic disorders (10, 11). Previous research has confirmed the role of vitamin D in endothelial function, blood pressure regulation, and cardiovascular health. Several mechanisms have been proposed to explain the link between vitamin D deficiency and cardiovascular disease, one of which is its role in regulating lipid profiles (12, 13). Some studies indicate that vitamin D deficiency is directly associated with reduced muscle strength and performance (14), as well as diminished finger strength in adolescent girls (15). However, other research suggests that since vitamin D receptors have not yet been identified on muscle membranes, its effects on muscle growth and function may be indirect (16). Given the documented influence of vitamin D on muscular performance, it appears that certain physical fitness indicators—such as flexibility and body composition—may be affected by vitamin D supplementation (17, 18, 19). Therefore, the present study seeks to answer the following question: Does eight weeks of Pilates training combined with vitamin D supplementation have a significant effect on lipid profile and flexibility in men with multiple sclerosis?

Materials and Methods

This study was applied in nature and employed a quasi-experimental design with pre-test and post-test measurements. The study population consisted of men with multiple sclerosis (MS) who were members of the Tehran MS Society. Out of 115 individuals attending sessions, 32 participants who met the inclusion criteria voluntarily enrolled in the study. They were randomly assigned into four groups: supplement, exercise, combined exercise + supplement, and control.

Inclusion criteria were as follows: male patients aged 40–45 years with a body mass index (BMI) of 25–30 kg/m²; diagnosed with relapsing-remitting MS; Expanded Disability Status Scale (EDSS) score ≤ 3; vitamin D deficiency (serum vitamin D < 30 ng/mL based on the most recent test); no use of antispasmodic drugs or prednisone; not in the acute or severe stage of the disease; and at least one year since diagnosis. Exclusion criteria included absence from more than three training sessions, onset of new illness, use of specific medications or other supplements, severe stress, relapse of MS, failure to take vitamin D supplements, or participation in physical activity outside the study protocol. Written informed consent was obtained from all participants prior to the intervention. One week before the intervention, baseline assessments were conducted for all four groups, including blood tests, the Wells sit-and-reach test for flexibility, and body composition analysis. Blood samples were collected to evaluate lipid profiles.

Exercise Protocol

The Pilates training program lasted eight weeks, with sessions held three times per week on alternate days (24 sessions total). Training was conducted indoors at a controlled temperature of 25–26°C. Sessions began at 20 minutes and progressively increased to 60 minutes. Exercises included flexibility-based Pilates movements such as hip flexor and adductor stretches, hamstring stretch, C-curve stretch, cat–camel exercise, trunk rotation in supine position (oblique contraction and stretch), abdominal contraction with knee-to-chest stretch, Superman exercise, contralateral arm–leg extension, prone trunk lift, forward and backward bridge, and cobra pose. Rest periods were limited to the time required to change positions. Training intensity was progressively increased by extending the duration of each movement (from 5 seconds with 6 repetitions in the first session to 8 seconds with 10 repetitions in the final session). Each session began with 10 minutes of warm-up stretching and ended with 10 minutes of cool-down relaxation (Adapted from the study by Rezaei *et al*l (2017) (20). The control group was instructed to continue their usual daily routines and refrain from any exercise. Post-tests were conducted one week after the intervention, identical to the pre-test assessments.

Vitamin D Supplementation

A total of 128 vitamin D supplements (50,000 IU softgel capsules) were purchased from a pharmacy. Each participant in the supplement and exercise + supplement groups received eight capsules. According to physician recommendations and based on participants' vitamin D deficiency, one capsule was consumed weekly for eight weeks. Participants were instructed to take one capsule every Saturday (21).

Blood sampling procedure

Participants were instructed to refrain from any light to moderate physical activity and from following calorie-restricted diets for 24 hours prior to blood collection. Blood samples were drawn between 8:00 and 9:00 a.m. after a 12-hour overnight fast. A volume of 5 mL was collected from the right antecubital vein by a trained laboratory technician. Each sample was transferred into test tubes containing ethylenediaminetetraacetic acid (EDTA) solution (Pars Azmoon, Iran). Plasma was separated by centrifugation at 3,500 rpm for 15 minutes using a Hettich centrifuge (Germany). Plasma samples were then stored at –80°C in a Mihann Azma freezer (Iran). Lipid profile analyses included triglycerides, total cholesterol, LDL, and HDL. Post-test blood sampling was conducted one week after the intervention, under the same conditions as the pre-test. Anthropometric measurements were obtained using a stadiometer (Seca, Germany), a digital scale (Sapor SSB-215D, China), a gym mat, and a Wells bench for flexibility testing.

Biochemical analyses:

Serum 25(OH)D concentration was measured using an enzyme immunoassay kit (Padtan Gostar Isar, Tehran, Iran) with a semi-automated microplate reader (STATFAX 3200; Awareness, Palm City, FL, USA). Total cholesterol was determined by the colorimetric (CHOD-PAP) photometric method using an autoanalyzer (MINDRAY BS 380, China). Triglycerides were measured by the colorimetric (GPO-PAP) photometric method using the same autoanalyzer. HDL-C was assessed enzymatically without VLDL precipitation, and LDL-C was calculated using an enzymatic colorimetric method, both analyzed with the MINDRAY BS 380 autoanalyzer.

Statistical analysis

Descriptive statistics (mean, standard deviation, and charts) were used to summarize the data. Normality of distribution was assessed using the Shapiro–Wilk test, confirming that the data were normally distributed. Homogeneity of variances was tested using Levene's test. One-way ANOVA was applied to examine differences between groups, and when significant, Tukey's post hoc test was used to identify pairwise differences. Statistical significance was set at $p \leq 0.05$. All analyses and graphical representations were performed using SPSS software, version 22.

Results

The findings of the study are presented in the following tables and figures.

Table 1 is shown the mean height of participants in the Pilates training, vitamin D supplementation, combined Pilates training + vitamin D supplementation, and control groups was 170.88 cm, 170.25 cm, 171.38 cm, and 169.38 cm, respectively. In addition, the mean body weight of participants in the same groups was 78.50 kg, 76.75 kg, 77.50 kg, and 72.88 kg, respectively.

Table 1. Descriptive findings related to participants' characteristics.

Variable	Group	Number	Minimum	Maximum	Mean	Standard Deviation
Height	pilates	8	168.5	173	170.88	1.246
	Supplement	8	169	172	170.25	1.436
	Supplement - pilates	8	170	173	171.38	2.383
	contorl	8	168.5	170	169.38	1.963
Weight	pilates	8	77	80	78.50	3.117
	Supplement	8	69	84	76.75	2.068
	Supplement - pilates	8	74.5	80.5	77.5	2.188
	contorl	8	71.5	74	72.88	2.875

Table 2 is shown given the level of statistical significance (p = 0.003, F = 6.033), there was a significant difference among participants across the three groups.

Table 2. Results of One-Way ANOVA for differences in mean triglyceride levels across training groups.

Source of Variation	SS (Sum of Squares)	df	MS (Mean Square)	F-value	p-value
Between groups	2565.754	3	855.583	6.033	0.003
Within groups	3971.573	28	141.813		
Total	6547.510	31			

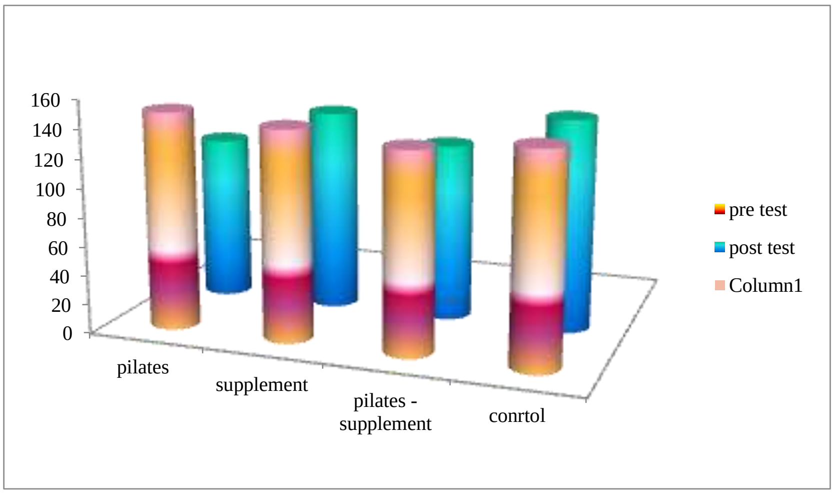

Figure 1. Changes in triglyceride levels across training groups

Table 3 is shown given the level of statistical significance (p = 0.0001, F = 86.565), there was a significant difference among participants across the three groups.

Table 3. Results of One-Way ANOVA for differences in mean cholesterol levels across training groups.

Source of variation	SS (Sum of squares)	df	MS (Mean square)	F-value	p-value
Between groups	839.385	3	279.792	86.565	0.0001
Within groups	90.510	28	3.232		
Total	929.885	31			

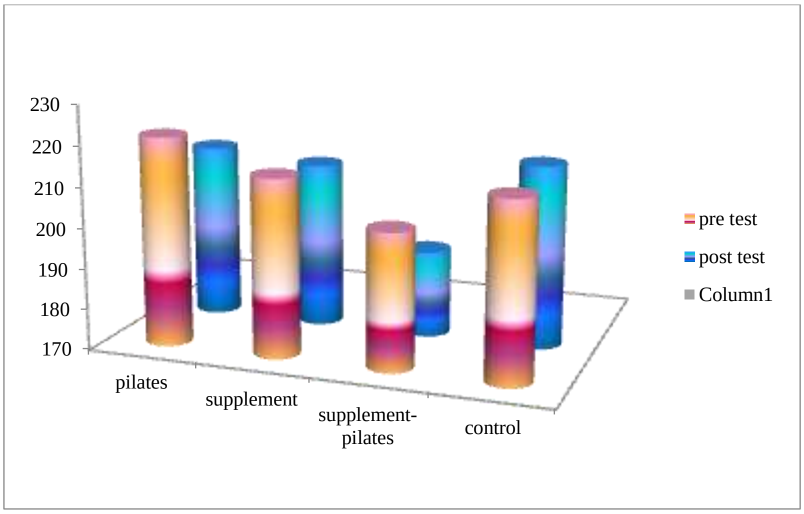

Figure 2. Changes in Cholesterol Levels Across Training Groups

Table 4 is shown given the level of statistical significance (p = 0.0001, F = 538.222), there was a significant difference among participants across the three groups.

Table 4. Results of one-Way ANOVA for differences in mean LDL levels across training groups.

Source of variation	SS (Sum of Squares)	df	MS (Mean Square)	F-value	p-value
Between groups	865.100	3	288.333	538.222	0.0001
Within groups	15.020	28	0.536		
Total	882.00	31			

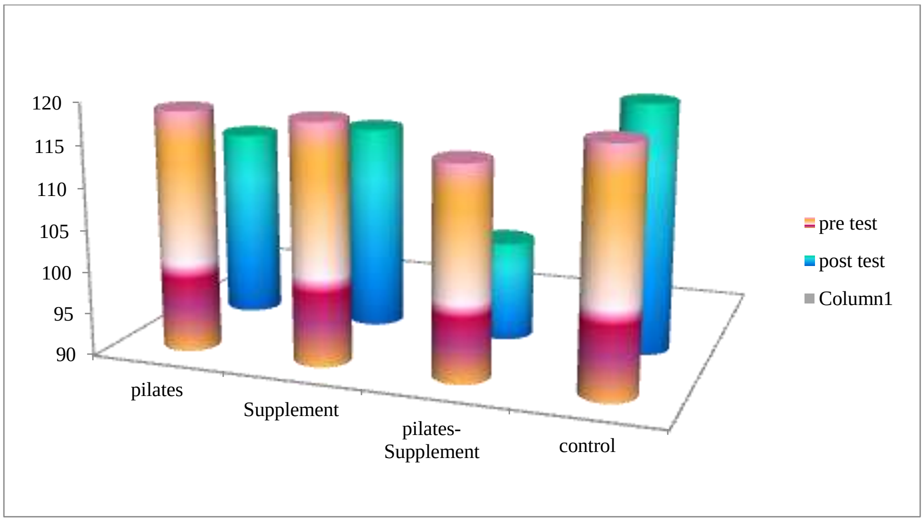

Figure 3. Changes in LDL Levels across training groups

Table 5 is shown given the level of statistical significance (p = 0.0001, F = 542.284), there was a significant difference among participants across the three groups.

Table 5. Results of One-Way ANOVA for differences in mean HDL levels across training groups.

Source of variation	SS (Sum of squares)	df	MS (Mean square)	F-value	p-value
Between groups	**196.194**	**3**	**65.365**	**542.284**	**0.0001**
Within groups	**3.476**	**28**	**0.121**		
Total	**199.569**	**31**			

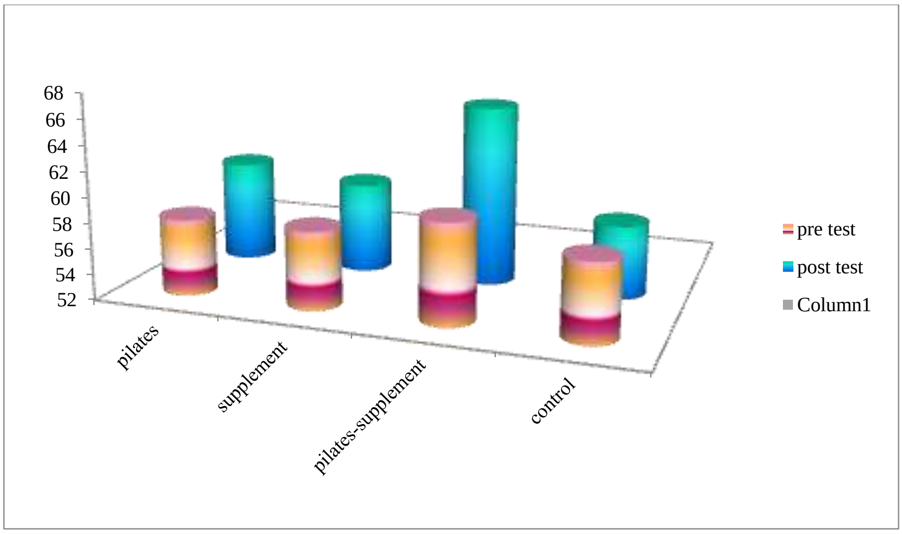

Figure 4. Changes in HDL levels across training groups.

Table 6 is shown given the level of statistical significance (p = 0.0001, F = 13.245), there was a significant difference among participants across the three groups.

Table 6. Results of One-Way ANOVA for differences in mean flexibility across training groups

Source of Variation	SS (Sum of Squares)	df	MS (Mean Square)	F-value	p-value
Between groups	71.854	3	23.948	13.245	0.0001
Within groups	50.634	28	1.808		
Total	122.470	31			

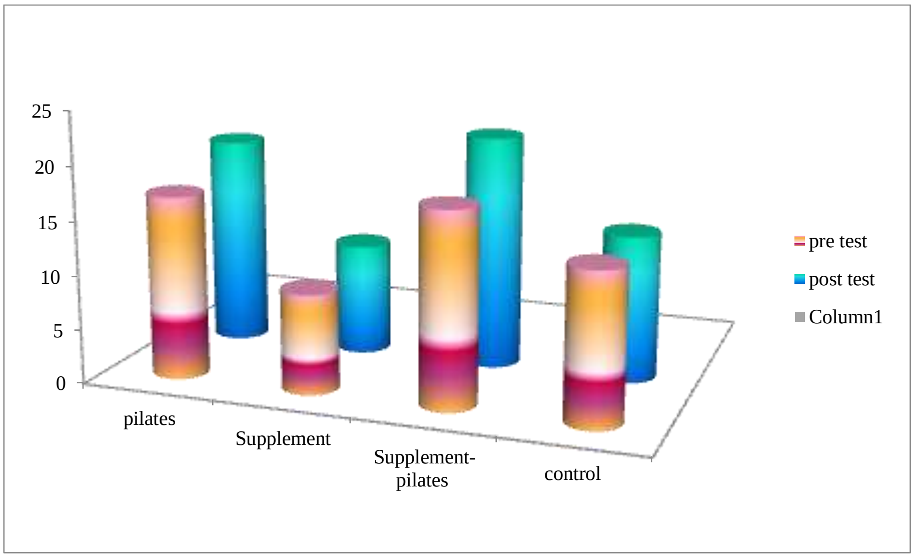

Figure 5. Changes in flexibility across training groups

Discussion

The findings of this study demonstrated that Pilates training combined with vitamin D supplementation had a significant effect on triglyceride and cholesterol indices in men with multiple sclerosis (MS). These results are consistent with the findings of Bahmani *et al*. (2022), Dehestani *et al*. (2019), and Farah Bakhsh *et al*. (2025). Similarly, Gombolay *et al*. (2025) reported that Pilates training significantly reduced triglyceride levels. In addition, Balaghi *et al*. (2021) observed a significant reduction in triglycerides among women with type 2 diabetes following Pilates training. The present study also revealed only a slight reduction in cholesterol levels. In line with this, Kaabomeir *et al*. (2024) reported a significant decrease in cholesterol following Pilates training in patients with type 2 diabetes, while Kasmari *et al*. (2024) found a 4.9% reduction in cholesterol among postmenopausal women after six weeks of aerobic exercise. Furthermore, Moravejolahkami *et al*. (2023) reported the effect of walking on cholesterol levels. A possible explanation for these findings is that cholesterol cannot be utilized as a direct fuel source during exercise; therefore, plasma cholesterol concentrations are not expected to change substantially with physical activity.

Mahmodirad *et al*. (2025) suggested that vitamin D may indirectly enhance metabolism and reduce body fat by increasing muscle mass, stimulating sympathetic nerves, and lowering insulin levels. It may also directly influence adipogenesis and adipocyte differentiation, reduce fatty acid absorption in the intestine, and regulate genes involved in fatty acid oxidation and mitochondrial metabolism(22). These mechanisms collectively contribute to increased fat oxidation, prevention of weight gain, and reductions in body weight and BMI. The results further indicated that eight weeks of Pilates training combined with vitamin D supplementation significantly affected low-density lipoprotein (LDL) levels in men with MS. These findings are consistent with those of Mahmodirad *et al.* (2025). In the Pilates training group, mean LDL levels decreased. Supporting this, Lasheen *et al*. (2022) and Lopez *et al*. (2023) reported that Pilates training reduced LDL in individuals with type 2 diabetes. Improvements in LDL following Pilates may be attributed to increased hepatic lipase and lipoprotein lipase activity at the cellular level, which enhance lipid metabolism and promote fat uptake by adipose tissue. Conversely, Ispoglou *et al*. (2023) reported no significant effect of Pilates training on LDL in patients with type 2 diabetes. Such discrepancies may be explained by differences in sample size, intervention duration, and dietary habits of participants. On the other hand, vitamin D has been shown to potentially reduce the formation of new adipocytes (23), suppress fat storage, and effectively limit fat accumulation. Moreover, vitamin D may increase serotonin and testosterone levels, both of which play critical roles in enhancing metabolism, increasing energy expenditure, regulating appetite, and promoting satiety (24).

The results of this study demonstrated that eight weeks of Pilates training combined with vitamin D supplementation had a significant effect on high-density lipoprotein (HDL) levels in men with multiple sclerosis (MS). These findings are consistent with those of Mahmodirad *et al*. (2025), who reported a significant increase in HDL among MS patients. Similarly, Vahdatpoor *et al*. (2023) observed notable improvements in HDL particle size and concentration, although these effects persisted only for 15 days after cessation of exercise. In contrast, Woo *et al*. (2024) reported no significant effect of Pilates training on HDL. Such discrepancies across studies may be attributed to differences in sample size, intervention duration, and dietary habits of participants. The present study also revealed that eight weeks of Pilates training combined with vitamin D supplementation significantly improved flexibility in men with MS. These findings are in line with those of Mahmodirad *et al*. (2025), Shahrjerdi *et al*. (2015), and Zarezadeh *et al*. (2017). Dorstyn *et al*. (2019) similarly reported significant improvements in flexibility in experimental groups compared to controls. Pilates exercises incorporate static active stretches with alternating ranges of motion, which contribute to maintaining joint health and reducing stiffness. The focused nature of Pilates movements allows the mind to guide the body through its full range of motion, thereby enhancing joint flexibility. This mechanism explains the observed improvements in flexibility following eight weeks of Pilates training(26).

Overall, the increase in flexibility observed in this study can be attributed to the repeated use of static active stretches and range of motion (ROM) exercises inherent in Pilates. These stretching effects help maintain youthfulness and vitality by counteracting joint weakness and neuromuscular reflex decline(26).

Conclusions

Based on the findings, vitamin D supplementation combined with regular exercise—particularly aerobic activity and Pilates—can significantly reduce metabolic risk factors such as triglycerides, cholesterol, and LDL, while increasing HDL in men with MS. In addition, this combination contributes to weight reduction, alleviates obesity associated with physical inactivity, enhances flexibility, and improves muscle weakness and range of motion. Therefore, Pilates training accompanied by vitamin D supplementation may play an effective role in promoting health, reducing disease-related complications, and improving the quality of life in men with multiple sclerosis.

Conflict of interests

No conflict.

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