

## Comparison of Eight Weeks Exposure to Sunlight and Home-Based Pilates Training on Serum Vitamin D , CCL20 and Body Composition of Overweight Multiple Sclerosis Women

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### Abstract

**Objective:** The purpose of this study was to compare eight weeks of exposure to sunlight and Home-Based Pilates Training serum vitamin D, CCL20, and the body composition of women with multiple sclerosis.

**Materials and Methods:** This study was a quasi-experimental study. Among women with Multiple sclerosis (with 2-5 EDSS), 44 were purposefully and voluntarily selected and randomly divided into three groups: Pilates at home (n=15), Pilates exposed to the sun (n=15) and control (n=14) were divided. The training program consisted of three sessions per week for eight weeks, consisting of two parts, Pilates training at home and Pilates training in the environment. An exercise program on the DVD was provided. The Serum vitamin D, CCL20 indices, and body composition of patients with MS were measured 48 hours before and after eight Pilates exercises at home and under sunlight. To analyze the data, analysis of covariance and Bonferroni post hoc test, SPSS software, and  $P \leq 0.05$  significance level.

**Results:** Comparing the results in the two stages showed that there was a significant difference between CCL20 ( $P= 0.001$ ), serum vitamin D ( $P= 0.001$ ), BMI ( $P= 0.001$ ), weight ( $P= 0.001$ ) and WHR ( $P= 0.001$ ) indices of women with MS in the studied groups. These differences were only between the training groups and the control group. However, there was a significant difference in the CCL20 index and serum vitamin D between the two training groups (Respectively  $P= 0.037$ ,  $P= 0.001$ ).

**Conclusion:** Patients with MS can improve their vitamin D level and weight-related and inflammatory indicators by using Pilates exercises exposed to sunlight and at home with minimal cost.


**Keywords:** Pilates training, Chemokine CCL20, Body composition, Multiple sclerosis, Sunlight, Vitamin D

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## Introduction

The pathological cause of multiple sclerosis (MS) is unknown (1). Between 25 and 33% of human beings with MS have weight problems. Depression, fatigue, and decreased mobility are associated with MS and obesity separately. Most behavioral weight reduction trials exclude people with neurologic disease. Therefore, little research has tested the outcomes of weight reduction on symptom presentation and fitness results among human beings with MS and obesity (2). Losing weight improves health, and reduces infection, stroke, MS, diabetes, certain cancers, and death among the overweight and obese. Weight loss not only improves mobility and energy but also increases stamina and mood. Mild obesity at age 20 doubles the risk of developing MS, according to studies (3). Among the cytokines, chemokines are responsible for leukocyte migration into inflammatory sites (4). CCL20 is expressed in a domain referred to as the choroid plexus where T cells enter the CNS. CCR6, the receptor of CCL20, is selectively expressed through CD4 + T cells that produce the cytokine IL-17. as a consequence, the CCL20-CCR6 interplay is involved within the blood mind barrier (BBB) disruption and follow-up migration of pathogenic T cells into the CNS (5).

Moreover, IL-17 secreted by circulating Th17 cells expands the pro-inflammatory response via NF- $\kappa$ B signaling to enhance CCL20 transcription (6). In one study by Fathiaet al., serum CCL20 levels in patients with Secondary progressive multiple sclerosis (SPMS) and relapsing-remitting multiple sclerosis (RRMS) are obviously higher than in healthy subjects. This trend is identical to the results of the 2017 research on CCL20 serum levels in RRMS patients and controls by Li R et al, as well as the results that higher CCL20 serum levels in patients with SPMS, RRMS, and primary progressive multiple sclerosis (PPMS) (5). Decreased exposure to sunlight and low 25-hydroxyvitamin D indicates that

there may be several risk factors for MS (7). In addition to receiving vitamin D from the skin's immune cell environment, are exposed to ultraviolet radiation from the sun which will affect the immune system of these patients (8). On the other hand, due to the physical condition of these patients, in order to strengthen their immune systems, they consider it necessary to conduct exercise training with appropriate intensity and duration for these people (9). Todd et al. (2017) in a study showed that taking 12 weeks of oral vitamin D supplementation had no effect on the immune system and inflammatory parameters of footballers (10).

One of the suitable training methods for these patients is due to their physical condition and problems with commuting at home and Pilates activity. Therefore, considering that Pilates is a safe and low-risk exercise and the fear of falling into this disease is low, and also training, learning, and controlling the intensity of this exercise at home is easier and more possible than other exercises, it is also possible to perform activities in the environment. There is also an opening for the absorption of vitamin D (1). Deep and diaphragmatic breathing in Pilates exercise, a better oxygen supply to the body, increases insulin sensitivity in people; there is no need to secrete a lot of insulin, and in this way fat oxidation in the body increases. Therefore, since deep and diaphragmatic breathing is one of the important principles in Pilates exercises, it is expected that as a result of these exercises, the amount of weight and body fat mass will decrease (11). On the other hand, one of the advantages of Pilates exercises compared to other exercises is that you can use muscles, joints, and organs in a targeted and focused manner. Focusing on the muscles of the central region of the body and abdominal muscles when performing Pilates exercises has an effective role in weight loss and body composition (12). This training method is applicable to standing, sitting, and lying down,

and will also involve the body, mind, and soul of people at the same time. On the other hand, considering the importance of sun exposure and vitamin D absorption in MS patients, this study examines the question of whether eight weeks of Pilates training at home and sun exposure on the levels of vitamin D, serum CCL20 in the blood and body composition of women with will it affect multiple sclerosis?

## Materials and Methods

### Participants

This study was a quasi-experimental method. Inclusion criteria included no smoking, no regular exercise for the past six months and at least two years of MS. All patients consisted of all female MS patients referred to the MS Association of Fasa with 2-5 EDSS. In this study, 44 people were purposefully and voluntarily selected to participate. Subjects were randomly divided into three groups Pilates exercises at home (n=15), Pilates exercises outdoors (n=15), and the control group (n=14). The conditions for entering the study included not smoking, not doing regular sports activity in the past year, being overweight, and having a history of MS disease for at least two years. Exclusion criteria also included muscle injuries, severe recurrence of the disease, inability to perform the exercise, lack of regular participation in the training protocol, participation in a training program other than the training protocol of the research, pregnancy, recurrence of MS in the last 12 weeks (13), Change in MS medication or steroid therapy in the past 12 weeks and failure to cooperate until the end of the research process. Then, the people who were eligible for the study were selected with an age range of 25 to 40 years. At the beginning of the work, written consent based on voluntary and conscious participation in training sessions was received from the subjects. Before starting the exercises, two or three introductory sessions were held to inform the patients about the possible benefits and side effects, as well as how to participate, and the required factors

(height, weight, Body mass index (BMI)) were measured.

### Exercise protocol

The exercise program consisted of 5 minutes of warm-up with stretching exercises (six stretches were maintained for at least 30 seconds) and the main body of the exercise consisted of 14 movements, selected from Pilates (the first week started with 10 movements and even weeks one movement added to the movements). In the eighth week, it ended with 14 movements Participants who were not comfortable using the DVD player were provided with an online Pilates link. Participants also contacted the researcher by phone or email, where they were allowed to ask about the DVD or the movements. Each Pilates session lasted approximately one hour and consisted of seven warm-up exercises and fourteen beginner-level exercises using a mat (exercise mat), as shown in Table 1 (1). The intensity used increased during the training period (increasing repetition and adding movement to the movements). The repetitions progressed gradually at two-week intervals, resulting in ten repetitions in the final two weeks (seventh and eighth weeks). Reporting, intensity, honesty, and adherence to the exercise program should be recorded through the individual exercise diary, which includes the date of the session, the number of repetitions performed in each exercise, and the rated perceived exertion) RPE (of each session, mentioned by the participant immediately after the exercise. Exercise intensity was measured by RPE and heart rate (1). Thus, RPE was 10 in the first phase of training in the first session and 12 in the eighth session (initially, before the start of the training program, the intensity of training was piloted by both the heart rate and the RPE index by the subjects). This is how to control the intensity of the exercise; Each person was told to stop moving before reaching a difficult stage (equivalent to the RPE of the target number). The intensity of exercise was also controlled by heart rate in each session. First,

the subjects' maximum heart rate was calculated using the formula ( $\text{age} \times 220 = \text{maximum heart rate (HRmax)}$ ), then they worked with a percentage of those subjects (intensity 50-55% in the first week, up to 65-70% of maximum heart rate in last week); (8).

### Measuring indicators

In this study, to measure serum vitamin D and CCL20, venous blood samples were taken from a brachial vein (12 hours fasting) before and 48 hours after the last training session (to prevent the acute effect of the last training session on blood variables). Immediately after blood sampling, the samples were centrifuged at 3000 rpm for 10 minutes and the isolated serum was kept at  $-80^{\circ}\text{C}$  until the parameters were measured. Blood samples were measured

to measure vitamin D levels and resilience index and sleep quality. Levels of 25-hydroxyvitamin D were measured using the ELISA method and instructions of the manufacturer (Padten Gostar Ishar Company Kit) with a sensitivity of 1.9 ng/ml. Also, the serum CCL20 index was measured by the ELISA method using the Zellbio Germany kit. Also, digital scales (model digital scales (YAGMI) with an accuracy of 0.1 kg made in Japan); (People in light clothing and without shoes) were used to measure weight before and after eight weeks of training. BMI is also calculated by dividing weight in kilograms by height squared in meters. Also, a tape measure was used to calculate the WHR (waist-to-hip ratio). The food recall questionnaire was used to control the nutrition of the subjects. All

**Table 1. Warm-up movements, main body training and cooling**

Warm-up	Main body training	Cooling
Head Nods	Ab Prep	Quadricep
Sliding Leg	C-Curve	Lower Back
Knee Drops	Single Leg Circle	Hamstrings
Knee Folds	Single Leg Stretch	Glutes
Hip Rolls	Double Leg Stretch	Adductors
Scapula Stabilisation	Hamstring Challenge	Calves
Arm Circles	Spine Stretch	Shoulders
	The Saw	Chest
	The Dart	Upper Back
	Prone Hip Extension	
	Cat Stretch	
	Shoulder Bridge	
	Side Lying Arm	
	Opening	
	Adductor Raise	

**Table 2. Pilates exercise protocol (overload, sets, repetitions, and rest)**

Exercise variables	Weeks							
	First	Second	Third	Fourth	Fifth	Sixth	Seventh	Eighth
Number of sessions per week	3	3	3	3	3	3	3	3
Repetition	4	4	6	6	8	8	10	10
The number is set	3	3	3	3	3	3	3	3
Rest time (Seconds)	30-60	15-30	30-60	15-30	30-60	15-30	30-60	15-30
Number of movements	10	11	11	12	12	13	13	14
Rest between sets (minutes)	4	4	4	4	3	3	3	3
RPE	10	10	10	11	11	11	12	12
Intensity% (HRmax)	50-55%	50-55%	55-60%	55-60%	60-65%	60-65%	65-70%	65-70%
Total test time (minutes)	60	60	60	60	60	60	60	60
Breathing	Surface	Surface	Deep	Deep	Deep	Deep and long	Deep and long	Deep and long
Warm-up (minutes)	10	10	10	10	10	10	10	10
Cooling (minutes)	5	5	5	5	5	5	5	5

were asked to write down all the food and beverages they had consumed in the past 24 hours. This questionnaire was completed for each subject in 3 non-consecutive times (one day off and two days off) (the first three days and the last three days of the program). The mentioned quantities of foods were converted to hot using the home scale guide. Then, each food was coded according to the instructions of the food processing software program (N4) and the nutritionist analyzed them in terms of energy and nutrients.

All data were analyzed using SPSS software version 26. Descriptive statistics were used to examine the mean and standard deviation. The Shapiro-Wilk method was also used to normalize the data. Analysis of covariance was used for comparison between groups. The Bonferroni post hoc test was also used to determine the differences between the groups. The significance level was considered  $P < 0.05$  for all statistical analyzes.

### Ethical considerations

This research has the ethical code IR.HSU.REC.1400.019 and the clinical code IRCT20220316054315N1.

### Results

The results of the Shapiro test one showed that all the data of this research were normal. The results of anthropometric and physical indices are presented in Table 3. Statistical results related to demographic indicators showed a significant difference between the weight and BMI of the subjects after eight weeks ( $P = 0.001$ ). This was the difference between the two training groups and the control group, but there was no significant difference between the two training groups. Also, no difference was observed between the age and height of the subjects in the initial measurement and comparison between the three groups ( $P = 0.98$  and  $P = 0.99$ , respectively), and the data of all three groups were homogeneous. The results of the analysis of covariance showed that there was a significant difference between the BMI and

WHR indices of the three groups after eight weeks ( $P = 0.003$ ). This was the difference between the two groups of exercise in the sun and at home with the control group ( $P = 0.001$  and  $P = 0.028$ , respectively). But there was no significant difference in WHR indices between the two training groups ( $P = 0.44$ ). Statistical results showed that there was no significant difference between the calories received by the subject ( $P = 0.72$ ).

The covariance results showed that after eight weeks of Pilates training, a significant difference was observed in the CCL20 index of the groups ( $P = 0.001$ ). The results of the post hoc test showed that the amount of CCL20 in the Pilates training group exposed to sunlight was significantly reduced compared to the Pilates training group at home ( $P = 0.037$ ). Also, there was a significant difference between the exposure to sunlight Pilates training group with the control group and the Pilates home training group with the control group ( $P = 0.001$ ). Blood serum CCL20 decreased by 20.07% in the home-based Pilates group and 10.93% in the Pilates exercise group at home. While in the control group, it increased by 1.88%. Both exercises significantly reduced the CCL20 in women with multiple sclerosis. On the other hand, due to the percentage change in the two exercise programs, the Pilates exercise program in the sun was more effective in reducing the CCL20 variable in the blood serum of women with multiple sclerosis. The results of the analysis of covariance showed that after eight weeks of training, a significant difference was observed between vitamin D levels ( $P = 0.001$ ). Additionally, after eight weeks, vitamin D levels in the Pilates group exposed to sunlight were significantly increased compared to the control group and the exercise group at home ( $P = 0.001$ ). But there was no significant difference between the control group and home exercise ( $P = 0.87$ ). Blood serum vitamin D increased by 78.43% in the Pilates exercise group exposed to sunlight and by 9.61% in the Pilates exercise group at home. While in the control group, it had increased by 3.11%.

According to the percentage of changes, the Pilates training program exposed to sunlight was more effective in increasing the serum vitamin D variable of women with multiple sclerosis. The results of covariance analysis in Table 4, and post hoc test results in Table 5 are presented.

## Discussion

The outcomes of this research showed that after eight weeks of outdoor and home-based Pilates training, there was a significant difference in the weight indices, body mass index, WHR, and CCL20 of the three groups. There were no significant differences in

**Table 3. Mean and standard deviation of physiological variables**

Variable	Groups	Mean ( $\pm$ SD)		P	Homogeneity
		Before intervention	After the intervention		
Age (years)	Pilates in the Sun.	35.16 ( $\pm$ 4.44)	-	0.93	0.11
	Pilates at home	35.75 ( $\pm$ 5.15)	-		
	Control	35.50 ( $\pm$ 3.62)	-		
Weight (kg)	Pilates in the Sun.	68.41 ( $\pm$ 3.77)	61.95 ( $\pm$ 2.19)	0.001*	0.85
	Pilates at home	71.41 ( $\pm$ 3.52)	67.00 ( $\pm$ 3.54)		
	Control	70.60 ( $\pm$ 5.71)	71.10 ( $\pm$ 5.70)		
Height (cm)	Pilates in the Sun.	161.75 ( $\pm$ 5.59)	-	0.90	0.90
	Pilates at home	162.75 ( $\pm$ 6.12)	-		
	Control	162.00 ( $\pm$ 5.05)	-		
Body Mass Index (BMI)	Pilates in the Sun.	26.41 ( $\pm$ 2.05)	23.91 ( $\pm$ 3.08)	0.001*	0.38
	Pilates at home	27.25 ( $\pm$ 3.01)	25.57 ( $\pm$ 2.47)		
	Control	26.94 ( $\pm$ 4.68)	27.13 ( $\pm$ 4.11)		
Calories received	Pilates in the Sun.	2555.80 ( $\pm$ 145.78)	2548.23 ( $\pm$ 140.76)	0.72	0.77
	Pilates at home	2601.16 ( $\pm$ 128.37)	2589.83 ( $\pm$ 180.17)		
	Control	2658.80 ( $\pm$ 93.00)	2631.60 ( $\pm$ 135.33)		

\* The symbol is significant

**Table 4. Results of analysis of covariance of vitamin D, serum CCL20 and WHR in patients with MS**

Variable	Groups	Mean ( $\pm$ SD)		P between groups
		Before intervention	After the intervention	
WHR (%)	Pilates in the Sun.	0.77 ( $\pm$ 0.03)	0.73 ( $\pm$ 0.03)	0.003*
	Pilates at home	0.77 ( $\pm$ 0.03)	0.75 ( $\pm$ 0.03)	
	Control	0.77 ( $\pm$ 0.02)	0.79 ( $\pm$ 0.01)	
CCL20 (pg/ml)	Pilates in the Sun.	86.39 ( $\pm$ 2.25)	69.8 ( $\pm$ 5.87)	0.001*
	Pilates at home	87.35 ( $\pm$ 3.04)	77.80 ( $\pm$ 10.34)	
	Control	87.00 ( $\pm$ 3.83)	88.64 ( $\pm$ 7.79)	
Serum Vitamin D	Pilates in the Sun.	18.36 ( $\pm$ 2.78)	32.76 ( $\pm$ 5.13)	0.001*
	Pilates at home	19.44 ( $\pm$ 2.40)	21.31 ( $\pm$ 2.32)	
	Control	18.44 ( $\pm$ 3.53)	19.22 ( $\pm$ 3.64)	

\* The symbol is significant

**Table 5. Results of post hoc analysis of vitamin D index, serum CCL20 and WHR in patients with MS**

Variable	Groups	Groups	Difference of averages	P
Weight (kg)	Pilates in the Sun.	Control	-7.58	0.001*
	Pilates at home	Pilates at home	-2.14	0.16
	Control	Control	-5.43	0.001*
BMI	Pilates in the Sun.	Pilates at home	-2.74	0.001*
	Pilates at home	Control	-0.59	0.45
	Control	Control	-2.14	0.001*
WHR (%)	Pilates in the Sun.	Pilates at home	-0.065	0.001*
	Pilates at home	Control	-0.022	0.44
	Control	Control	-0.043	0.028*
CCL20 (pg/ml)	Pilates in the Sun.	Pilates at home	-19.40	0.001*
	Pilates at home	Control	-8.45	0.037*
	Control	Control	-10.94	0.008*
Serum Vitamin D	Pilates in the Sun.	Pilates at home	13.88	0.001*
	Pilates at home	Control	12.76	0.001*
	Control	Control	1.21	0.87

\* The symbol is significant

weight indices, body mass index, and WHR between two training groups in two different environments. The only difference was between the training groups and the control group. Also, a significant distinction turned into discovered in the CCL20 index between the 2 training groups and the control group. In the CCL20 index, a significant difference was observed between the two exercise groups in two different environments, and the exercise group exposed to the sun had a greater decrease in CCL20 than the exercise group in the home environment. These modifications showed a significant decrease within the training groups in comparison to the control group, but no enormous difference turned into found among the two Pilates training groups within the different environments.

In this regard, Wamberg et al. (2013) in a study consistent with the present study showed that taking 26 weeks of vitamin D supplementation did not have a significant effect on weight loss and BMI of obese women (13). Considering the present study, after eight weeks, the level of vitamin D in the blood had significantly increased in the group exposed to the sun, but no significant difference was observed between the body weight, BMI, and WHR of the two groups after eight weeks. Bozoğlu, et al. (2018) After 10 weeks of Pilates exercises observed a significant decrease in the indicators of body weight, BMI, and WHR in obese women (14). In another study inconsistent with the present study, Siraj et al. (2013) showed that eight weeks of Pilates exercise had no significant effect on the WHR index of non-athlete women. Among the reasons for insignificance, we can mention the type of subject and the appropriate body composition according to the initial BMI (BMI=22). Therefore, eight weeks of Pilates did not significantly alter the WHR. One of the reasons given by the researcher for not changing the WHR index was their lack of control over the subject's feeding schedule (15).

General effects found that Pilates results in an enormous decrease in body weight, BMI,

and WHR in adults with overweight. Reasons for weight loss and body mass include increased calorie intake by exercise and possibly changes in body composition. In not changing the circumference of the waist to the pelvis, we can point to the difference in the distribution of fat in different areas and the cost per calorie of exercise. The reduction in body weight and BFP seems to be more reported in research consisting of individuals with obesity most effective, and the efficacy of Pilates for the improvement of body weight and BMI seems to be extra obvious in longer intervention duration (16). Due to the fact that multiple sclerosis patients participate less in sports activities that require leaps and bounds due to imbalance. As a result, they gain more weight. On the other hand, according to the results of this research and the nature of Pilates exercise, this exercise can lead to less risk of falling for these patients. Therefore, Pilates exercises can cause weight loss in these patients by engaging the large and central muscles of the body.

The results of this study showed that eight weeks of Pilates training under the sun and at home caused a significant decrease in serum CCL20 index in both groups compared to the control group. Based on the reviews, similar research on CCL20 was not observed in the field of exercise exposed to sunlight and at home. Sports studies are also very limited in relation to this index and people with MS. Therefore, we tried to use limited studies in relation to patients who have a similar course of MS. The results of this study were consistent with the results of Jafarzadeh et al. (2019); (17) and inconsistent with the results of Ziegler et al. (2019); (18). Jafarzadeh et al. (2019) in a study the effect of vitamin D supplementation on the expression of They showed that the consumption of vitamin D3 in mice with EAE reduced the expression of CCL20, CCL22, and CCR4 receptor chemokines in the spinal cord and serum compared to the group receiving olive oil. But it was slightly higher than the healthy control group (17). CCL20 performs a critical function

in the migration of Th17 cells to inflamed tissues (19). After Th17 cells enter the target tissues, they secrete large amounts of inflammatory cytokines, causing a cytokine cascade by the inflammatory cells. This activates the inflammatory process and stimulates the production of CCL20. This rationalization affords an ability interpretation for the chronic presence of Th17 in the lesions of MS sufferers through a superb feedback loop by way of C.

According to the results of Pryor et al. (2015), a decrease in the CCL20 index after Pilates training may be the result of the effect of long-term exercise on the inactivation of interleukins, especially IL-17, resulting in decreased CCL20 expression in patients with MS. Comparison of eight weeks of Pilates training exposed to sunlight and home showed a significant increase in vitamin D in the sun group compared to the two groups of exercise at home and the control group. These results showed the difference between the two groups of outdoor training with home training and the control group. But after eight weeks, there was no significant change in the amount of vitamin D in the home exercise group compared to the control group. Bahmani et al. (2021) in a study showed that aerobic exercise at home does not significantly change the level of vitamin D in the blood of women with M (8). But Bauer et al showed physical activity and sun exposure for 14 days Increased the level of vitamin D in the serum of patients with MS (20). They reported a strong positive correlation between physical activity and vitamin D levels in the blood of people with MS. Their results also showed that physical activity independent of exposure to sunlight improved serum vitamin D levels in patients with low-grade MS (0 to 1.5), Therefore, Bauer et al., The reasons for the lack of change in vitamin D levels of people exposed to sunlight were skin type, ambient temperature, genetic makeup, type of clothing, use of sunscreen, age and height (20).

There may be proof that exercise-stimulated parathyroid hormone (PTH) activates renal

calcitriol synthesis. Also, a decrease in exercise-precipitated serum phosphate (a diet D inhibitor) may additionally result in accelerated vitamin D levels (21). The difference is in the level of the subject's ability to activate hormones and activates the desired pathway, as well as the effect of the time of training sessions on the secretion of hormones and activating the production and secretion of vitamin D. The mentioned cases can be the reasons for the difference between the results of the research and the results of the present study in the field of the effect of exercise on the vitamin D levels of the subjects. The strength of this research is the simultaneous examination of exercise and absorption of vitamin D through the environment for patients with MS. Among the limitations of this research, we can mention the lack of control over the motivation and the daily activity level of the subjects, which in future research can achieve more accurate results.

## Conclusions

The results of the present study showed that eight weeks of Pilates exercise at home and under the sun reduced the inflammatory index and reduced weight and body composition in women with MS. According to the results of exercise under sunlight, it had a greater effect on reducing the inflammatory index CCL20 and increasing vitamin D. Therefore, according to the favorable results of this study, it is recommended that MS patients regularly perform Pilates under the sun due to its low cost, controllable intensity, easy learning of movements and no need for special facilities and equipment.

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## Conflict of Interest

The authors hereby state that there is no struggle of interest within the present have a look at.

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