

# Vitamin D3 Supplementation Improved Physical Performance in Healthy Older Adults in Japan: A Pilot Study

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## Abstract

Japan has the highest proportion of older adults and so-called “Super-aged society” in the world. These results suggested that the prevalence of both cognitive and physical functional impairment increases with age. Therefore, the present study was designed to investigate the effect of vitamin D3 supplementation in cognitive and physical functional impairment in healthy older adults in Japan. We enrolled 7 Japanese male (age:  $76.0 \pm 8.7$ ) and 5 female (age:  $78.3 \pm 9.3$ ) in this study. The physical function of even a person getting on a wheelchair could be tested in all subjects. Treatment group ( $n = 7$ ) consisted of a 500 IU/day vitamin D3 corresponding to twice of daily requirement for 6 months. Blood was collected by venipuncture and the serum 25-hydroxy vitamin D (25 OHD), 1,25-hydroxy vitamin D (1,25 OHD), Ca and PTH concentration were measured. The Mini-Mental State Examination and the Montreal Cognitive Assessment-Japanese version were used for the cognitive function test. Physical function was measured objectively using the Timed Up and Go (TUG), 4-mgait speed test (4 MGS), 5-repetition sit-to-stand (5 STS), 30-second chair stand test (CS-30) and measurement of upper grip strength. Our results show that a 6-month intake of vitamin D3 increased serum 25 OHD within the insufficiency and sufficiency levels without changing 1,25 OHD, Ca and PTH concentration. And it significantly decreased 4 MGS without changing TUG, 5 STS, CS-30, upper grip strength and cognitive function. Serum 25 OHD cut-off values for skeletal muscle index, grip strength and 4 MGS in diagnosis of sarcopenia are 18.6 - 23.4 ng/mL. These findings suggest that serum 25 OHD levels (23.4 ng/mL) might improve moving capacity, lower limb muscle strength, and physical balance functional

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impairment.

## Keywords

Serum 25-Hydroxyvitamin D, Cognitive Function, Physical Function, Aging, Sarcopenia

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## 1. Introduction

Vitamin D is a secosteroid associated with peripheral calcium homeostasis [1]. Vitamin D is available in vitamin D<sub>2</sub> of plants and D<sub>3</sub> of animals. Both vitamin D<sub>2</sub> and D<sub>3</sub> are biologically inert and require activation through two hydroxylation processes involving 25-hydroxylase (CYP2R1) and 1 $\alpha$ -hydroxylase (CYP27B1), located in the liver and kidney, respectively [2]. 1,25-dihydroxyvitamin D (1,25 OHD) is a biologically active metabolite produced by two steps of hydroxylation reactions [3].

Recent evidence has identified a beneficial role of vitamin D in maintaining cognitive and physical functions. Vitamin D receptor and CYP27B1 have been found in the brain (hippocampus) [4] and skeletal muscle [5]. Over expression of reactive oxygen species (ROS) stimulated by disruption of cerebral blood flow is one of the main causes of vascular dementia-induced cognitive deficits in the rat model [6]. 1,25 OHD was reported to have a significant physiological anti-oxidant activity [7].

The low 25-hydroxy vitamin D (25 OHD) level has been recently associated with greater risk of cognitive impairment in older as well as younger adults using the Montreal Cognitive Assessment (MoCA) Arabic version [8]. Vitamin D supplementation caused significant improvement in the cognitive performance by Mini-Mental State Examination (MMSE) score in Alzheimer's disease [9]. Lower serum 25 OHD is associated with poorer functional mobility using walking speed, Timed Up and Go test (TUG), and cognitive function using MMSE [10]. Vitamin D<sub>3</sub> supplementation improves muscle function and physical performance in the elderly population using 4 meter gait speed test (4 MGS) [11]. In our latest study, a significant positive correlation was found between urinary 25 OHD/creatinine and MMSE or Japanese version of the MoCA (MoCA-J), and a significant negative correlation between serum 1,25 OHD and TUG or 4 MGS [12]. Other studies, however, have not found an association between vitamin D and physical performance [13] [14] [15] [16] [17]. Vitamin D supplementation has not been found to increase muscle strength in older adults [18] [19] [20] [21].

Therefore, the present study was designed to investigate the effect of vitamin D<sub>3</sub> supplementation corresponding to twice of the daily requirement for 6 months in cognitive and physical functional impairment in healthy older adults in Japan.

## 2. Methods

### 2.1. Subjects and Setting

Prior to the study, the ethical approval was obtained from the ethics committee of Kyoto Bunkyo Junior College (project registration number in 2016: 7) and Aichi Medical College for Physical and Occupational Therapy (Project registration number in 2016: 468). A total of 12 healthy adults age  $\geq 65$  years were included in adult day-care center clients in Fukui ( $n = 4$ ) and Aichi Prefectures ( $n = 8$ ). These areas with varying daylight hours were selected. The annual daylight hours were maximum in Aichi (2255 hrs/year: the 4th in Japan), and minimum in Fukui (1788 hrs/year: the 37th in Japan) between the two areas. The researchers attended the adult day-care center and assured the proper management of safety and confidentiality of the study. Eligible participants were required to not have had sarcopenia, osteoporosis and functional disorder of thyroid. The manager of the adult day-care center invited participation in the study, and all the subjects whose participation was requested were selected from September in 2017 to May in 2018. After obtaining informed consent from a family member belonging to the same household, we enrolled 7 Japanese male (age:  $80.8 \pm 6.1$ ) and 5 female (age:  $75.9 \pm 7.5$ ) in this study. The physical function of even a person getting on a wheelchair could be tested among all subjects. All the participants eat at home. Treatment group ( $n = 7$ ) consisted of a vitamin D supplements (500 IU/day of the vitamin D3 purchased from UHA Mikakuto Co., Ltd., Osaka) for 6 months. The hypercalcemia at vitamin D intake level is lower than 10,000 IU/day (Dietary Reference Intake for Japanese, 2015).

### 2.2. Cognitive Function Test

The MMSE was used for the cognitive function test. It consists of five downstream items of orientation, memory, attentiveness for calculations, speech function, and design capacity. The maximum score for the MMSE is 30 points, and cutoff score for dementia were of 23 to 24 points [22]. The MoCA-J may be better at detecting early cognitive dysfunction [23] and was used for the cognitive function test. The maximum score for the MoCA-J is 30 points, and cutoff score for dementia were of 25 to 26 points [9]. These tests were performed by verbal questioning of 5- to 10-min duration by skilled occupational and physical therapists.

### 2.3. Muscle Weight and Physical Function

Muscle weight was measured using Inbody 430 (Inbody Japan, Tokyo) and calculated as skeletal muscle index (SMI). Physical performance tests included balance, lower limbs muscular strength, 4 MGS and upper grip strength. The TUG of functional mobility is assessed by asking the participant to stand up from a standard chair, walk a 3 meter distance, turn, walk back to the chair and sit down again [24]. The lower limbs muscular strength was measured against the time to complete 5-repetition sit-to-stand (5 STS) and the number to chair stand

for 30 sec (CS-30) [25]. All tests were performed by skilled physical therapists.

#### 2.4. Serum 1,25 OHD, 25 OHD, Ca and PTH Assay

Blood was collected by venipuncture and serum 1,25 OHD, 25 OHD, Ca and PTH concentration were measured by Nikken Igaku Co. (Fukui, Japan) and Falco Holdings Co. (Kyoto, Japan).

#### 2.5. Statistics

Results are expressed as the mean  $\pm$  S.D., and differences between before and after intervention with vitamin D3 supplements were evaluated using the Wilcoxon test. A  $p$ -value  $< 0.05$  was considered to be statistically significant. Analyses were carried out using SPSS 21 for Windows (IBM, Japan).

### 3. Results

#### 3.1. Study Subjects

Characteristics of the study subjects are shown in **Table 1**. Mean age was 76.0 years for males ( $n = 7$ ) and 78.3 years for females ( $n = 5$ ). Obesity was defined as a body-mass index (BMI)  $\geq 25.0$  kg/m<sup>2</sup>. The prevalence of obesity defined BMI was 0% in males and 26.7% in females (2 subjects in each group).

#### 3.2. Change in Serum 25 (OH) D, 1,25 (OH) D, Ca and PTH

A 6 month intake of vitamin D3 supplements significantly increased serum 25 OHD concentration within the insufficiency and sufficiency level (3 subjects:  $>20$  ng/mL and  $<30$  ng/mL, 3 subjects:  $>30$  ng/mL) without changing serum Ca and PTH concentration (**Figure 1**). These parameters in control group were not changed for 6 months. These results suggested that vitamin D3 supplements intake maintained serum 25 OHD levels.

#### 3.3. Serum 25 OHD and Cognitive Function

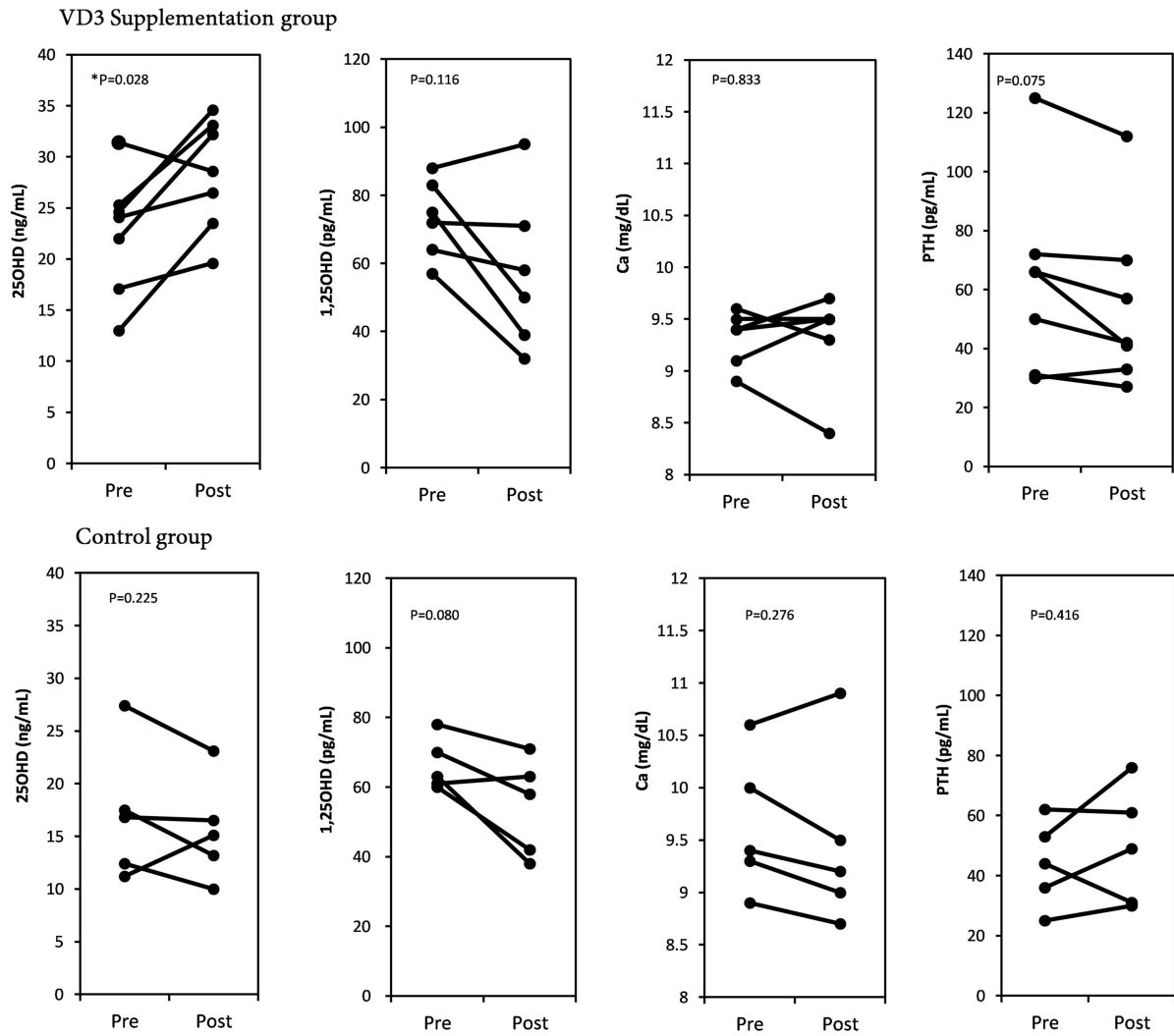
Significant changes in cognitive function in supplementation and control groups were not seen (**Figure 2**). These findings suggest that vitamin D3 supplementation was not associated with change in cognitive function.

#### 3.4. Serum 25 OHD and Physical Function

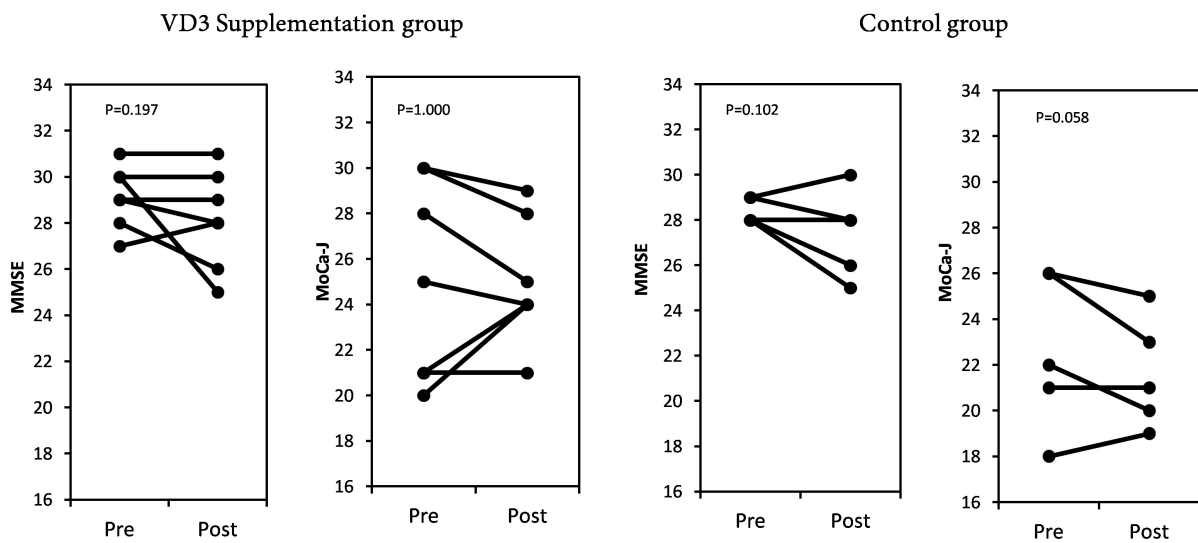
Vitamin D3 supplementation decreased 4 MGS without changing TUG, 5 STS, CS-30 and upper grip strength (**Figure 3**). These findings suggest that serum 25 OHD levels might improve moving capacity, lower limb muscle strength, and physical balance functional impairment.

#### 3.5. Serum 25 OHD Cut-Off Values for Diagnosis of Sarcopenia

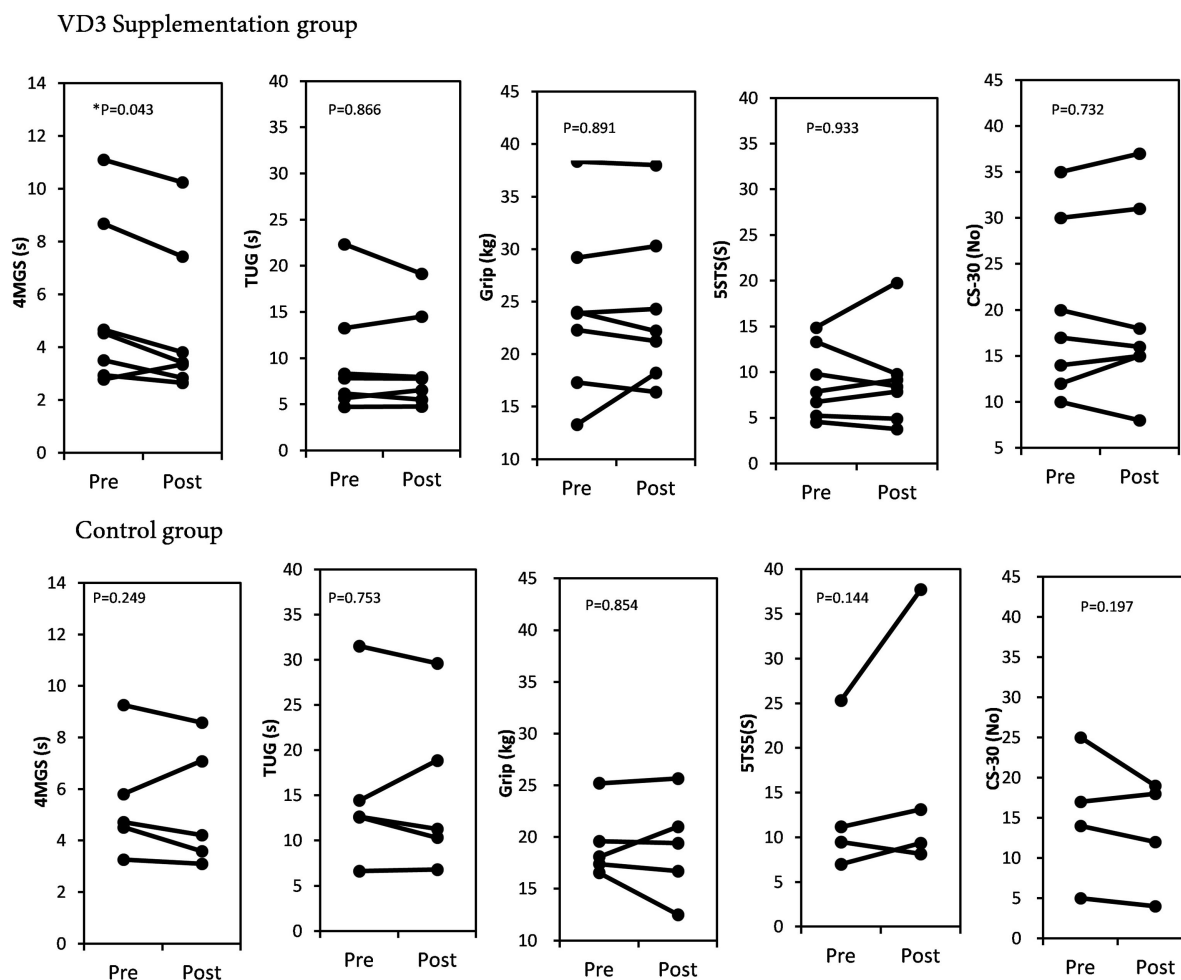
The cut-off values of each test types for the diagnosis of sarcopenia are shown in **Table 2**. From the correlation coefficient and  $p$ -values, serum 25 OHD cut-off values are calculated. These cut-off values were 18.6 - 23.4 ng/mL.



**Figure 1.** Effects of vitamin D3 supplementation on serum biological parameter. \*, *p*-value < 0.05 was considered to be statistically significant.



**Figure 2.** Effects of vitamin D3 supplementation on cognitive function.



**Figure 3.** Effects of serum 25 OHD on physical function. \*,  $p$ -value < 0.05 was considered to be statistically significant.

**Table 1.** Characteristics of the study subjects.

	VD3 Supplementation group	Control group
No. of participants (% male)	7 (33.3)	5 (40.0)
Age (y)	77.8 ± 8.5	77.6 ± 5.0
Body height (cm)	156.2 ± 7.8	152.4 ± 3.6
Body weight (kg)	55.9 ± 9.8	56.2 ± 9.4
BMI (kg/m <sup>2</sup> )	22.8 ± 2.8	24.3 ± 4.3

**Table 2.** Serum 25 OHD cut-off values for physical functions in diagnosis of sarcopenia.

Test types	Test cut-off values	Correlation coefficient and $p$ values between test and serum 25 OHD	Serum 25 OHD cut-off values (ng/mL)
Grip strength (female)	<20 kg <sup>27)</sup>	0.522/0.038	18.6
4 MGS (s)	<5 sec <sup>13)</sup>	-0.58/0.003	23.4

## 4. Discussion

In the present study, the number of the vitamin D3 supplement group with 25 OHD > 30 ng/mL (normal) increased from 1 to 3 and the number of vitamin D3 supplement group with 25 OHD < 20 ng/mL (deficient) decreased from 2 to 1 during 6 months vitamin D supplementation. These data showed that vitamin D3 supplementation was associated with improvement of serum vitamin D levels and the vitamin D3 dose used in the intervention (500 IU/day) was low for typical improvement of cognitive and physical functions.

In the present study, vitamin D3 supplementation significantly decreased 4 MGS without changing 5 STS, TUG and cognitive function. The effect of vitamin D3 on 4 MGS was especially clear. However, vitamin D3 was not associated with change in CS-30. These results suggested that vitamin D3 was found to improve moving capacity, but not endurance. The cut-off values for predicting the level of risk of falls in community-dwelling elders are 1.0 m/sec in 4 m walking speed [26]. The number of vitamin D3 supplement group with 4 MGS > 4 sec decreased 4 to 2 during 6 months vitamin D3 supplementation. Vitamin D bound vitamin D receptor in skeletal muscle and increased mitochondria function [27]. The functional decline of the nervous system would greatly affect the decrease of walking speed in elderly with weaker muscular strength [28]. These findings suggest that serum 25 OHD levels could contribute to improve neuro-muscular system and physical performance.

In 1989, Rosenberg proposed the term “sarcopenia” to describe age-related decrease of muscle mass [29]. Sarcopenia represents an important risk factor for disability and mortality. Vitamin D deficiency appears to enhance intramuscular adipose tissue impacting as reduced functionality in skeletal tissues [30]. These findings suggested that vitamin D affects skeletal muscle mass and morphology. Sarcopenia still has no broadly accepted clinical definition. But in this study subjects ( $77.6 \pm 7.4$  years), 9 (75% of subjects) have values lower than sarcopenia diagnostic criteria (muscle weight, upper grip strength and walking speed). In the present study, the cut-off values of serum 25 OHD for the diagnosis of sarcopenia were 18.6 - 23.4 ng/mL. Our results suggested that maintained 25 OHD levels above 23.4 ng/mL prevent sarcopenia in the elderly.

Recent evidence suggests a potential beneficial role of vitamin D in maintaining cognitive function [31]. However, prospective studies have not found an association between 25 OHD concentration and cognitive function. In the present study, daily 500 IU vitamin D3 (serum 25 OHD  $\leq$  30 ng/mL) supplementation was not associated with change in cognitive function. These results suggested that the threshold value for improvement of physical function is lower than that for improvement of cognitive function.

Further study of optimal 25 OHD levels for maintaining physical, cognitive function and preventing falls is needed.

The limitation of this study includes small sample size and possible selection bias. Further subjects are needed.

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## Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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