



## Vitamin D Levels and Their Relationship with Ambulation in Patients with Amyotrophic Lateral Sclerosis

*Amyotrofik Lateral Sklerozlu Hastalarda Vitamin D Düzeyleri ve Ambulasyonla İlişkisi*

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### Abstract

**Objective:** Vitamin D is a steroid hormone that exerts many effects on human health. Amyotrophic lateral sclerosis is a progressive neurodegenerative disease characterized by the loss of motor neurons. This can result in a number of complications, including muscle weakness, spasticity, difficulty swallowing, speech disturbances, and, in some cases, respiratory failure. A number of studies have reported an association between vitamin D and various neurological diseases. This study aimed to assess vitamin D levels in patients diagnosed with amyotrophic lateral sclerosis and compare them with those of a control group comprising healthy individuals. In addition, the correlation between vitamin D levels and functional ambulation levels will be examined.

**Materials and Methods:** This retrospective cross-sectional study was conducted involving 29 male and female patients diagnosed with definite amyotrophic lateral sclerosis using the El Escorial criteria and 28 healthy controls from the physical medicine and rehabilitation clinic of medicine. The medical records of the patients were consulted to obtain relevant demographic, clinical, and vitamin D levels. Subsequently, vitamin D levels were compared with those of healthy controls.

**Results:** The results are presented in the following section. The mean vitamin D level observed in the patient group with amyotrophic lateral sclerosis was  $20.21 \pm 9.53$ , while in the healthy control group, it was  $26.69 \pm 8.90$ . A statistically significant difference in vitamin D levels was observed between the two groups ( $p=0.034$ ). No correlation was observed between patients' ambulation levels and vitamin D levels ( $p=0.202$ ).

**Conclusion:** It can be concluded that patients with amyotrophic lateral sclerosis are at risk of vitamin D deficiency or insufficiency. It is therefore recommended that regular vitamin D levels are measured and treatment plans are developed for these patients.

**Keywords:** Amyotrophic lateral sclerosis, vitamin D, ambulation

### Öz

**Amaç:** Vitamin D insan sağlığı üzerine çok yönlü etkileri olan steroid yapıda bir hormondur. Amyotrofik lateral skleroz ilerleyici kas güçsüzlüğü, spastisite, yutma, konuşma bozuklukları ve solunum yetmezliğine yol açan motor nöronların progresif kaybıyla karakterize nörodejeneratif hastalıktır. Çalışmalarda vitamin D'nin nörolojik pek çok hastalıkla da ilişkili olduğu bildirilmiştir. Amacımız amyotrofik lateral skleroz hastalarında vitamin D düzeylerini değerlendirmek ve sağlıklı kontrollerle karşılaştırmak, vitamin D düzeyinin fonksiyonel ambulasyon düzeyleriyle ilişkisini değerlendirmektir.

**Gereç ve Yöntem:** Retrospektif, kesitsel çalışmaya tıp fakültesi fiziksel tıp rehabilitasyon kliniğinde takipli El Escorial kriterlerine göre kesin amyotrofik lateral skleroz tanılı kadın ve erkek 29 hasta ve 28 sağlıklı kontrol alındı. Hastaların kayıtlı dosyalarından demografik, klinik verileriyle vitamin D düzeyleri kaydedildi. Vitamin D düzeyleri sağlık kontrollerle karşılaştırıldı.

**Bulgular:** Amyotrofik lateral skleroz hastalarının vitamin D düzeyi:  $20,21 \pm 9,53$ , sağlıklı kontrol grubunun  $26,69 \pm 8,90$  idi. Gruplar arası vitamin D düzeylerindeki fark istatistiksel olarak anlamlıydı ( $p=0,034$ ). Hastaların ambulasyon düzeyi ile vitamin D düzeyleri arasında ilişki saptanmadı ( $p=0,202$ ).

**Sonuç:** Amyotrofik lateral skleroz hastaları vitamin D eksiklik/yetersizlik açısından risk altında olan bir gruptur. Bu hastalara vitamin D düzeylerinin düzenli ölçümlerinin yapılması ve tedavilerinin planlanmasının uygun olacağını düşünmekteyiz.

**Anahtar kelimeler:** Amyotrofik lateral skleroz, vitamin D, ambulasyon

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## Introduction

Vitamin D is a steroid hormone with a wide range of physiological effects on human health. While its most significant and well-documented effects are on the skeletal system, there is also substantial scientific evidence linking deficiency of vitamin D with a range of neurological disorders, including Parkinson's disease, multiple sclerosis, neurodegenerative conditions, migraine headaches, and diabetic neuropathy. Vitamin D receptors are expressed in a number of vital brain regions, including the substantia nigra, hippocampus, hypothalamus, thalamus, and subcortical grey nuclei, in both neurons and glial cells. In these regions, vitamin D has been demonstrated to be effective in the differentiation and maturation of neurons. It has been demonstrated to play a role in the modulation of growth factor biosynthesis, including that of nerve growth factor derived from glial cell lines, and in the biosynthesis of various transmitters, including acetylcholine (1).

It is of the utmost importance to evaluate vitamin D not only in terms of serum 25-hydroxyvitamin D [25(OH)D] levels, but also as an indicator of overall lifestyle.

Indeed, reduced vitamin D levels may indicate dietary habits and levels of physical exercise, as well as being influenced by several health-related factors, such as body mass index (BMI). It is therefore recommended that serum 25(OH)D levels be considered not only as an indicator of vitamin D status, but also as an indicator of overall health status. Furthermore, there is evidence to suggest that vitamin D plays a role in muscle strength. A deficiency in vitamin D has been associated with impaired physical performance, a reduction in bone mineral density, and an elevated incidence of falls (2). The prevalence of amyotrophic lateral sclerosis (ALS) is estimated at 2-3 per 100,000. A range of factors, including genetic, environmental, and lifestyle factors, are implicated in the etiology of this disorder. The range of medical treatment options is restricted, as there are only two approved pharmaceutical agents for the disease: riluzole and edaravone (3). Therapeutic intervention aims to improve quality of life and prolong survival through a multidisciplinary approach, with a particular focus on symptom management (4,5). Recent years have seen a growing body of evidence emerge from scientific research examining the potential role of vitamin D in the context of ALS and its relationship with the disease. The role of vitamin D levels in ALS patients and its role in the disease are subjects of considerable debate and controversy (6). The objective of our study was to evaluate vitamin D levels in patients with ALS and to compare them with those of healthy controls. Additionally, we sought to assess the relationship between patients' vitamin D levels and their functional ambulation levels.

## Materials and Methods

The study was conducted using a cross-sectional, retrospectively designed methodology. The İzmir Bakırçay University Non-Interventional Clinical Research Ethics Committee has reached a decision, numbered 1438 (date: 24.01.2024), in regard to

the aforementioned matter. The study population consisted of female and male patients diagnosed with definite ALS who first sought treatment at the Physical Medicine and Rehabilitation Clinic of İzmir Bakırçay University Faculty of Medicine between May and November of the years 2022-2023. Individuals who had received vitamin D treatment were excluded from the study. A review of the medications used by patients was conducted based on an analysis of their medical records. Patients with nephropathy (serum creatinine level >1.5 mg/dL), tube feeding, gastrostomy use, and those who were taking vitamin D supplements were not included in the study.

A review of the patients' follow-up files was conducted to obtain information regarding their demographic characteristics, the auxiliary devices they were using, their functional ambulation levels, and the concentration of 25(OH)D, calcium, phosphorus, and parathormone in their serum. The serum 25(OH)D levels of the patient group were compared with those of a healthy control group of the same age and gender. Subsequently, the relationship between vitamin D levels and functional ambulation values in the patient group was evaluated.

25(OH)D levels were measured by electrochemiluminescence immunoassay (ECLIA) using the Elecsys Vitamin D total III, version 2.0 kit on the Roche Cobas E 801 analyser (Roche Diagnostics, Mannheim, Germany). 25(OH)D levels were measured by ELISA. Serum 25(OH)D levels were categorised as follows: a level of less than 10 ng/mL indicates vitamin D deficiency, a level of 10-20 ng/mL indicates vitamin D insufficiency, a level of 20 ng/mL or more is considered sufficient for bone health, and a level of 30-50 ng/mL is considered sufficient for extra-bone effects. In this study, a level of 30-50 ng/mL was found to be adequate for extra-bone effects (7).

The Functional Ambulation scale (FAS) was employed to assess the patients' ambulatory abilities (8). This scale is used to classify the patient's motor skills with regard to ambulation.

- Level 0: The patient is non-functional and unable to ambulate.
- Level 1: The patient is able to walk with constant support and supervision or continuous manual assistance.
- Level 2: The patient is able to ambulate with the assistance of a walking aid, bearing minimal weight on their own limbs.
- Level 3: The patient is able to walk on flat ground without manual assistance, provided that they are under the supervision of a healthcare professional.
- Level 4: The patient is able to walk independently on flat ground, but is unable to climb stairs or traverse uneven surfaces.
- Level 5: The patient is able to walk independently on any surface and at any speed.

Patients were classified into two groups based on their FAS level. The initial cohort comprised individuals with levels 0, 1, or 2, indicative of a markedly impaired capacity for ambulation. In contrast, the subsequent cohort consisted of those with levels 3, 4, or 5, which are typically regarded as indicative of an adequate ambulatory capacity.

Patients' BMI was categorised as follows: A BMI of less than 18.4 is classified as low weight, a BMI of 18.5 to 24.9 is classified as normal weight, a BMI of 25 to 29.9 is classified as excess weight, and a BMI of 30 to 34.9 is classified as obese (9).

### Statistical Analysis

Statistical analysis was performed with SPSS version 25 for Windows. Normality of the data was assessed using the Shapiro-Wilk test. Data with a normal distribution were presented as mean ± standard deviation, whereas non-normally distributed data were presented as median and interquartile range. Student's t-test or Mann-Whitney U test was used to determine whether there were significant differences in means or medians between the groups. Correlations between variables were assessed using the Pearson or Spearman test, as appropriate. Chi-square test was used to assess associations between categorical variables. A p-value of lower than 0.05 was regarded as statistically significant.

### Results

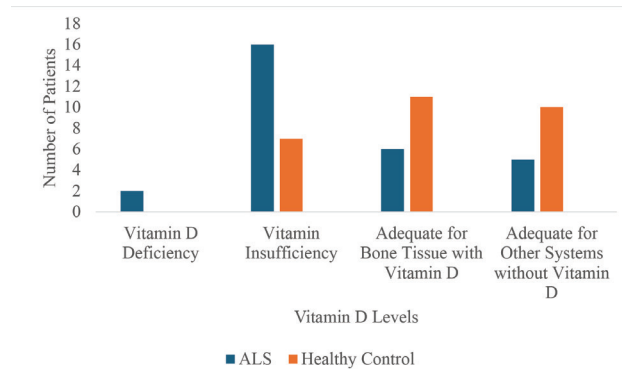
Initially, 30 patients with ALS were to be included in the study. However, one patient's data was excluded as an outlier, resulting in 29 patients being included in the study. Of the 29 patients, 18 were males, and 11 were females. The healthy control group consisted of 28 individuals, of whom 17 were males and 11 were females. The group of ALS patients had a similar age and sex distribution to the control group (p=0.916).

The demographic data of the patients with ALS are shown in Table 1.

The average vitamin D level in men with ALS was 22.96±9.15, while in women, it was 15.70±8.70. There was a statistically significant difference between the two groups (p=0.022).

The ALS patient group and the control cohort had sufficient vitamin D levels to support bone health. However, the levels observed in both groups were below the optimal range for extracellular tissues (<30 ng/mL). A statistically significant difference was observed between vitamin D levels in ALS patients and healthy controls (p=0.034) (Table 2).

Figure 1 shows the levels of 25(OH)D observed in both the ALS cohort and the matched healthy control group. Vitamin D levels in patients with severe ambulatory impairment (n=8) were 16.51±6.5 ng/mL, whereas those who were able to walk (n=21) had levels of 21.62±10.23 ng/mL. There was no statistically significant difference between the two groups (p=0.202).



**Figure 1.** Vitamin D Levels of amyotrophic lateral sclerosis and healthy control group  
ALS: Amyotrophic lateral sclerosis

**Table 1. Demographic data of patients with amyotrophic lateral sclerosis**

	n	%
Mean age in years	59.20±7.77	
<b>Gender</b>		
Male/female	18/11	63/37
Average year of diagnosis (year) (mean ± SD)	4.17±3.14	
<b>Body mass index</b>		
Low weight	1	1.8
Normal weight	12	21.1
Excess weight	9	15.8
Obese	7	12.3
<b>Functional ambulation level</b>		
FAS 0	7	24.1
FAS 1	0	0.0
FAS 2	1	3.4
FAS 3	5	17.2
FAS 4	11	37.9
FAS 5	5	17.2
<b>Assistive device usage</b>		
No device used	18	62.1
Cane	2	6.9
Walker	1	3.4
Wheelchair	8	27.6
FAS: Functional Ambulation scale, n: Patient count, %: Percentage rate, SD: Standard deviation		

**Table 2. Biochemical data of amyotrophic lateral sclerosis and healthy control group**

	ALS	Control	p-value
Vitamin D (ng/mL)	20.21±9.53	26.69±8.90	<b>0.034*</b>
Phosphorus	3.53±0.55	3.24±0.46	<b>0.035*</b>
Calcium	9.48±1.43	9.55±0.28	0.106
Parathormone	44.55±15.60	41.42±12.73	0.322

\*p<0.05 is statistically significant. ALS: Amyotrophic lateral sclerosis

The aim of this study was to analyse vitamin D concentrations between ALS patients and a healthy control group. The association between vitamin D levels and walking was investigated. Vitamin D levels were found to be significantly lower in the ALS patient group compared to the healthy control group. While vitamin D levels were sufficient for bone health (>20 ng/mL) in both ALS patients and the healthy group, they were low for extracellular systems in both groups. No correlation was found between the vitamin D levels and functional ambulation in ALS patients.

## Discussion

Vitamin D is a powerful hormone. It has a profound effect on all systems and organs in the human body. Serum levels should be considered an indicator of lifestyle in assessments. Low vitamin D levels are indicative of dietary habits and physical activity levels, so it is recommended that vitamin D levels be taken as an indicator of lifestyle in assessments. Vitamin D exerts neuroprotective effects in numerous pathophysiological processes by modulating oxidative stress, mitochondrial dysfunction, and apoptotic pathways. In ALS, increased levels of glutamate-induced excitotoxic neuronal damage, reactive oxygen radicals, and apoptosis have been implicated in the regulation of intracellular signalling and the maintenance of interneuronal balance (10). The role of vitamin D in ALS is a topic of ongoing debate, with studies investigating its levels, potential as a biomarker, impact on disease progression, and the impact of vitamin D supplementation on clinical outcomes (6). Vitamin D inadequacy and insufficiency are recognised as global epidemics. In our country, the prevalence of vitamin D deficiency is 51.8% (9). Several studies have reported significantly lower vitamin D concentrations in patients with ALS compared to healthy controls (10-12). Cortese et al. (13) determined the average vitamin D levels in 71 patients and assessed them using the ALS Functional Rating scale-revised (ALSFR-R). The prevalence of vitamin D deficiency was found to be related to disease severity. It has also been postulated that the different degrees of ALS symptom onset may be explained by different disturbances in vitamin D signalling pathways in cortical/spinal motor neurons (13,14). In their systematic review, Plantone et al. (15) evaluated twelve observational studies and one randomised controlled trial involving 1,280 patients with ALS and compared their vitamin D levels with those of controls. The researchers found that people with ALS had reduced vitamin D levels compared with healthy controls. On average, ALS patients had vitamin D concentrations that were -6.1 ng/mL lower than those in the healthy controls (15). Regarding the frequency of vitamin D deficiency in patients with ALS, although these individuals generally have lower concentrations of vitamin 25(OH)D compared with the normal range, there are studies that report no statistically significant difference in vitamin 25(OH)D concentrations between ALS patients and controls (16,17).

Vitamin D supplementation has been shown to improve motor function and reduce the incidence of falls (18). In contrast to the findings of our study, vitamin D supplementation has been shown to have a beneficial impact on motor function in ALS mouse models, while vitamin D deficiency has been associated with a detrimental effect on motor function and disease severity (19). The researchers also looked at the association between vitamin D levels and gender in people with ALS. The results showed that men had higher vitamin D levels. It is possible that changes in hormone levels in postmenopausal women, traditional clothing and limited sun exposure in older female patients may be contributing factors (20). The present study did not find a correlation between walking and vitamin D levels. However, the progressive deterioration of ambulation in ALS patients may lead to reduced exposure to direct sunlight, which could contribute to vitamin D deficiency.

In the present study, functional ambulation was assessed in terms of gross motor function. No association was found between vitamin D levels and walking disability or respiratory function tests. This finding is consistent with the observations reported by Yang et al. (21). Nevertheless, Camu et al. (22) conducted an evaluation of vitamin D levels and their correlation with ALS prognosis using a total of nine observational studies. The researchers correlated vitamin D levels with a clinical marker used to assess ALS prognosis, the ALSFRS-R, forced vital capacity and survival. A correlation was found between low vitamin D levels in ALS patients and impairment of gross motor function and ALSFRS-R scores. Camu et al. (22) reported an association between lower vitamin D levels and more severe disease progression. Vitamin D levels were found to be positively related to survival, highlighting its neuroprotective function with respect to motor neurons (22). The available literature suggests that vitamin D supplementation has no effect on prognosis. However, a single randomised controlled trial reports an increase in ALSFRS-R with high dose vitamin D. Therefore, regular assessment of vitamin D levels in patients is recommended (23).

## Study Limitations

Our study's value lies in its pioneering evaluation of vitamin D levels in ALS patients in our country. The main limitation of the study is the relatively small number of patients included. It is recommended that future studies with larger numbers of patients be planned to evaluate vitamin D levels and clinical outcomes such as ALSFR-R after vitamin D supplementation.

## Conclusion

In conclusion, patients with ALS are at risk of developing vitamin D deficiency or insufficiency. It is recommended that regular assessment of vitamin D levels and supplementation be undertaken in patients with ALS, given the beneficial effects of vitamin D on all systems, with particular benefit to bone health.

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### Ethics

**Ethics Committee Approval:** The İzmir Bakırçay University Non-Interventional Clinical Research Ethics Committee has reached a decision, numbered 1438 (date: 24.01.2024).

**Informed Consent:** Retrospective study.

### Authorship Contributions

Concept: F.M.S., F.D.A., Design: F.M.S., F.D.A., Data Collection or Processing: F.M.S., F.D.A., Analysis or Interpretation: F.M.S., F.D.A., Literature Search: F.M.S., Writing: F.M.S.

**Conflict of Interest:** No conflict of interest was declared by the authors.

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### References

1. Theodoratou E, Tzoulaki I, Zgaga L, Ioannidis JP. Vitamin D and multiple health outcomes: umbrella review of systematic reviews and meta-analyses of observational studies and randomised trials. *BMJ*. 2014;348:g2035.
2. Yao P, Bennett D, Maffham M, Lin X, Chen Z, Armitage J, et al. Vitamin D and Calcium for the Prevention of Fracture: A Systematic Review and Meta-analysis. *JAMA Netw Open*. 2019;2:e1917789.
3. Mazahery H, von Hurst PR. Factors Affecting 25-Hydroxyvitamin D Concentration in Response to Vitamin D Supplementation. *Nutrients*. 2015;7:5111-42.
4. Masrori P, Van Damme P. Amyotrophic lateral sclerosis: a clinical review. *Eur J Neurol*. 2020;27:1918-29.
5. Hardiman O, Al-Chalabi A, Chio A, Corr EM, Logroscino G, Robberecht W, et al. Amyotrophic lateral sclerosis. *Nat Rev Dis Primers*. 2017;3:17071.
6. Lanznaster D, Bejan-Angoulvant T, Gandia J, Blasco H, Corcia P. Is There a Role for Vitamin D in Amyotrophic Lateral Sclerosis? A Systematic Review and Meta-Analysis. *Front Neurol*. 2020; 11: 697.
7. Osteoporoz ve Diğer Metabolik Kemik Hastalıkları Çalışma Grubu. Osteoporoz ve Metabolik Kemik Hastalıkları Tanı ve Tedavi Kılavuzu. Türkiye Endokrinoloji ve Metabolizma Derneği; 2020. Available from:URL: [https://file.temd.org.tr/Uploads/publications/guides/documents/20210104143325-2021tbl\\_kilavuz5e76bb3d16.pdf](https://file.temd.org.tr/Uploads/publications/guides/documents/20210104143325-2021tbl_kilavuz5e76bb3d16.pdf)
8. Gündüz NE, Dilek B, Ergin O, Şahin E, Dadaş Ö, Kızıl R, et al. "Nöromusküler Hastalıklarda Aktivite Kısıtlanması ve Fonksiyonel Değerlendirme: Tersiye Bir Merkez Sonuçları." *J PMR Sci*. 2023;26:199-205.
9. Çubukcu M, Türe E, Yazıcıoğlu B, Yavuz E. *Türk Aile Hek Derg*. 2021;25:47-52.
10. Di Somma C, Scarano E, Barrea L, Zhukouskaya VV, Savastano S, Mele C, et al. Vitamin D and Neurological Diseases: An Endocrine View. *Int J Mol Sci*. 2017;18:2482.
11. Libonati L, Onesti E, Gori MC, Ceccanti M, Cambieri C, Fabbri A, et al. Vitamin D in amyotrophic lateral sclerosis. *Funct Neurol*. 2017;32:35-40.
12. Elf K, Askmark H, Nygren I, Punga AR. Vitamin D deficiency in patients with primary immune-mediated peripheral neuropathies. *J Neurol Sci*. 2014;345:184-8.
13. Cortese R, D'Errico E, Introna A, Schirosi G, Scarafino A, Distaso E, et al. Vitamin D levels in serum of amyotrophic lateral sclerosis patients. (P2.069). *Neurology*. 2015;84(Suppl 14).
14. Crick PJ, Griffiths WJ, Zhang J, Beibel M, Abdel-Khalik J, Kuhle J, et al. Reduced Plasma Levels of 25-Hydroxycholesterol and Increased Cerebrospinal Fluid Levels of Bile Acid Precursors in Multiple Sclerosis Patients. *Mol Neurobiol*. 2017;54:8009-20.
15. Plantone D, Primiano G, Manco C, Locci S, Servidei S, De Stefano N. Vitamin D in Neurological Diseases. *Int J Mol Sci*. 2022;24:87.
16. Karam C, Barrett MJ, Imperato T, Macgowan DJ, Scelsa S. Vitamin D deficiency and its supplementation in patients with amyotrophic lateral sclerosis. *J Clin Neurosci*. 2013;20:1550-3.
17. Öğüş E, Sürer H, Kılınc AŞ, Fidancı V, Yılmaz, Dindar N, et al. Evaluation of Vitamin D Levels by Months, Sex and Age. *Ankara Med J*. 2015;5(1):1-5.
18. Chevalley T, Brandi ML, Cashman KD, Cavalier E, Harvey NC, Maggi S, et al. Role of vitamin D supplementation in the management of musculoskeletal diseases: update from an European Society of Clinical and Economical Aspects of Osteoporosis, Osteoarthritis and Musculoskeletal Diseases (ESCEO) working group. *Aging Clin Exp Res*. 2022;34:2603-23.
19. Gianforcaro A, Solomon JA, Hamadeh MJ. Vitamin D(3) at 50x AI attenuates the decline in paw grip endurance, but not disease outcomes, in the G93A mouse model of ALS, and is toxic in females. *PLoS One*. 2013;8:e30243.
20. Paganoni S, Macklin EA, Karam C, Yu H, Gonterman F, Fetterman KA, et al. Vitamin D levels are associated with gross motor function in amyotrophic lateral sclerosis. *Muscle Nerve*. 2017;56:726-31.
21. Yang J, Park JS, Oh KW, Oh SI, Park HM, Kim SH. Vitamin D levels are not predictors of survival in a clinic population of patients with ALS. *J Neurol Sci*. 2016;367:83-8.
22. Camu W, Tremblier B, Plassot C, Alphantery S, Salsac C, Pageot N, et al. Vitamin D confers protection to motoneurons and is a prognostic factor of amyotrophic lateral sclerosis. *Neurobiol Aging*. 2014;35:1198-205.
23. Mentis AA, Bougea AM, Chrousos GP. Amyotrophic lateral sclerosis (ALS) and the endocrine system: Are there any further ties to be explored? *Aging Brain*. 2021;1:100024.