Comparison of Vitamin D_3 Serum Levels in New Diagnosed Patients with Multiple Sclerosis *versus* Their Healthy Relatives

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Abstract- Multiple risk factors are introduced for Multiple Sclerosis (MS). Recent studies have suggested a possible correlation between vitamin D deficiency and an increase risk of MS. This study was therefore undertaken to compare vitamin D levels in new cases of MS and their relatives as healthy controls. Seventy five new diagnosed MS patients and 100 matched healthy controls among their relatives were enrolled in this study. Two groups were matched for gender, age, season in which serum level of vitamin D was checked and region and diet. Serum levels of 25-hydroxy vitamin D was measured, recorded and analyzed. Seventy five patients (57 female and 18 male) and 100 healthy subjects (75 female and 25 male) were enrolled in this study. The mean serum levels of 25-hydroxy vitamin D in case and control groups were 11.31 and 17.9 ng/ml, respectively (P=0.003). Compared to the healthy subject, serum levels of vitamin D were significantly lower in patients with MS. This difference was only significant in women. Observed difference of vitamin D levels of both groups were significant in summer. This study continues to support the role of vitamin D deficiency in MS.

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Introduction

Multiple sclerosis (MS) is one of the most common chronic inflammatory autoimmune diseases of the central nervous system which is characterized by demielinization and fibrosis in brain and spinal cord (1).

It is suggested that vitamin D prevents MS and maintaining proper blood levels of vitamin D in patients with MS could decrease disease activity (2).

An immunosuppressant activity is suggested for vitamin D and inhibitory effects of vitamin D on Th1 and stimulatory effects on Th2 are reported in recent in vitro studies (3).

It has not yet established whether vitamin D is an immune regulator or it could support the treatment of MS (3,4). Correlation between MS and vitamin D is documented (5) but it is not demonstrated whether vitamin D acts as a risk factor for MS or vitamin D deficiency is one of MS consequences. It has been shown in a later study that low levels of vitamin D is observed in patients suffering from MS many years before the time of diagnosis; and concluded that vitamin D deficiency is considered as a risk factor for MS (6). Additionally opposing correlation between serum vitamin D levels and MS clinical activity is also reported (7). A strong relationship between disability and exposure to sunlight is also showed in a recent study in Australia. Vitamin D supplementation for MS patients is also suggested in recent studies in order to decrease disease activity, flare ups and resulted disabilities (8).

However results of published studies are still contradictory. This study was therefore undertaken to compare vitamin D levels in new cases of MS and their relatives as healthy controls.

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Materials and Methods

In a two years period of time, seventy five consecutive new diagnosed cases of MS were enrolled in this retrospective case-control study. In order to minimize confounding factors of vitamin D levels such as economic level and diet, one hundred healthy age and sex matched controls among relatives of the patients were also enrolled to the study.

Diagnosis of MS was confirmed using McDonald 2010 criteria, (9) with Lumbar puncture (LP) and Cerebrospinal fluid (CSF) analysis plus two times of Magnetic resonance imaging (MRI) with and without Gadolinium contrast.

Exclusion criteria were as follows:

1) Vitamin D supplementation during the last 6 months before study

2) Patients treated with cyclosporine, lipid lowering agents and hormones

3) Patients with chronic kidney disease, malabsorption and endocrine diseases

4) History of recurrent pregnancy

Sampling from the patients and their relatives (controls) was performed at the same time in order to eliminate effect of time (seasons) on vitamin D levels.

Serum vitamin D levels were determined with electrochemiluminescence method using commercial kits (Roche Elecsys 2010 Chemistry Analyzer, Roche, Germany). Laboratory technician was blinded to the samples history whether they were obtained from the patients or their controls.

Regarding serum vitamin D levels, the subjects first categorized into three groups of normal (vitamin D levels> 30 ng per milliliter), insufficiency (vitamin D levels 20-30 ng per milliliter), and deficiency (vitamin D levels<20 ng per milliliter) and compared with each other. Subjects of insufficiency and deficiency groups then were combined and analysis was repeated to compare vitamin D deficient and normal subjects.

Statistical Package for the Social Sciences, Version 17.0 (SPSS Inc. Chicago, IL, USA) was used for statistical analysis. Statistical significance was set at 0.05. Results are expressed as mean ± Standard Deviation (SD). Student's t-test was applied to compare means of two groups.

The study protocol was reviewed and approved by ethics committee of Hamadan University of Medical Sciences. Study protocol was explained for the patients and their relatives and written informed consent was obtained from them.

Results

One hundred and seventy five subjects were enrolled in this study. Age and sex distribution of the participants in the case and control groups is shown in table 1. Mean ± standard deviation (SD) age of the subjects in the case and control groups were 30.2±0.91 and 34.76±1.07 years respectively.

Season distribution of sampling of the participants in the study was as follows:

Spring (case: 28%, control: 27%), summer (case: 21.33%, control: 23%), autumn (case: 22.66%, control: 23%), winter (case: 28%, control: 27%).

A significant difference was observed between mean serum levels of vitamin D of the case (11.31, 95% CI 9.4-13.1) and control (17.9, 95% CI 14.4-21.3) groups (P=0.003, t=0.052). Serum levels of vitamin D between women of both groups was also significant (P=0.009, t=2.65) while it was not significant between men (P=0.012, t=1.59).

Serum levels of vitamin D was significantly different in case (mean: 11.8, 95% CI 9.04-14.57) and control groups (mean: 23.43, 95% CI 9.03-34.2) which were obtained in summer (P=0.03, t=2.26). There was no significant difference in serum vitamin D levels of samples obtained in the other seasons (P> 0.05).

In 20 to 40 years old subjects of both groups, observed differences between serum levels of vitamin D was statistically significant (P=0.029, t=2.21), while it was not significant amongst the other age groups.

Table 1. Age and sex distribution of the participants in the study.

		Case group (n=75)		Control group (n=100)	
		Number	**Percent**	**Number**	**Percent**
Sex	Male	57	76	75	75
	Female	18	24	25	25
Age	<20 years	3	4	4	4
	20-40 years	61	81.3	66	66
	>40 years	11	14.7	30	30

Ninety six percent of the patients suffered from vitamin D deficiency (deficiency 84%, insufficiency 12%), while 85% of the subjects of control group had vitamin D deficiency (deficiency 65%, insufficiency 20%).

Discussion

Consecutive patients were enrolled in this study and majority of them were women. This is in accordance with the previous findings which were demonstrated a higher prevalence of MS in women (6).

Compared to the healthy controls, serum levels of vitamin D were significantly lower in the patients with MS. Prospective studies were also suggested vitamin D as a risk factor for MS (4). A recent published study reported low levels of 25-hydroxy vitamin D in patients with MS (5). Several studies have also reported low vitamin D levels in patients with MS (10-12).

Despite lower serum levels of vitamin D in women, there was no statistically significant correlation between sex and serum vitamin D levels in our study. However, compared to the healthy women, serum vitamin D levels were significantly lower in women subgroup of the patients with MS. On the other hand, there was no significant difference between serum vitamin D levels of men of both groups. Although lower sample size of men, may explain insignificancy of observed trend of decreased vitamin D levels in men with MS.

Majority of the patients in this study were 20-40 years old. This finding is in accordance with the previous reports of age distribution of MS (7).

Living in high latitudes is correlated to lower serum levels of vitamin D (4,5). Decreased exposure to ultra violet waves of sunlight is suggested as the main mechanism of this finding. Although vitamin D deficiency due to nutritional deficiencies is also prevalent (2,6,13), low levels of serum vitamin D and lesser sun exposure are related to higher MS prevalence in the literature (5,7,14). According to the results of this study, a significant difference in serum vitamin D levels were observed between the two groups in summer, while no statistically significant difference were observed in the other seasons. It shows that in presence of low sunlight exposure (*i.e.* in winter), both healthy subjects and patients with MS suffer from vitamin D deficiency. In seasons with higher sun exposure, only patients with MS were demonstrated vitamin D deficiency and serum levels of vitamin D were near normal and normal in healthy subjects. Results of a similar study in Finland are in line with our findings (15).

*Study limitations***:** MS flare ups and correlation between serum levels of vitamin D could be evaluated in this study. A larger sample size could increase the power of this study and help to achieve significant results in observed trends.

Study strength points

The controls of this study were selected from the patients' healthy relatives (sister or brother). It could eliminate confounding factors such as diet, economic status, and genetics of vitamin D metabolism. Although there is an increased likelihood of MS in the relatives of the patients with MS, this fact increases value of observed significant differences in this study. In conclusion, compared to the healthy subject, serum levels of vitamin D were significantly lower in patients with MS. This difference was only significant in women. Observed difference of vitamin D levels of both groups were significant in summer. This study continues to support the role of vitamin D deficiency in MS.

Suggestions

Measurement of serum levels of vitamin D at the time of diagnosis of MS is recommended; higher exposure to sunlight and vitamin D supplementation is suggested for the patients with MS. Further larger follow up cohorts are recommended to investigate effect of vitamin D deficiency and also vitamin D supplementation in prevalence and control of MS.

References

1. Weinstock-Guttman B, Bakhshi R. Combination Therapy for Multiple sclerosis: The Treatment Strategy of the future? CNS Drugs 2004;8(12):777-92.
2. Zittermann A. Vitamin D in preventive medicine: are we ignoring the evidence? Br J Nutr 2003;89(5):552-72.
3. Berlanga-Taylor AJ, Ramagopalan SV. Vitamin D and multiple sclerosis: what is the clinical impact? Expert Opin Med Diagn 2013;7(3):227-9.
4. Hewer S, Lucas R, van der Mei I, Taylor BV. Vitamin D and multiple sclerosis.J Clin Neurosci 2013;20(5):634-41.
5. Pierrot-Deseilligny C, Souberbielle JC. Contribution of vitamin D insufficiency to the pathogenesis of multiple sclerosis. Ther Adv Neurol Disord 2013;6(2):81-116.
6. Munger KL, Levin LI, Hollis BW, Howard NS, Ascherio A. Serum 25-Hydroxyvitamin D levels and Risk of Multiple sclerosis. JAMA 2006;296(23):2832-8.

7. Soilu-Hanninen M, Laksonen M, Laitinen I, Erlinna J, Lilius E, Mononen I. A longitudinal study of serum 25-hydroxyvitamin D and intact parathyroid hormone levels indicate the important of vitamin D and Calcium homeostasis regulation in Multiple sclerosis. J Neurol Neurosurg Psychiatr 2008;79:152-7.
8. van der Mei IA, Ponsonby AL, Dwyer T, Blizzard L, Taylor BV, Kilpatrick T, Butzkueven H, McMichael AJ. Vitamin D levels in people with multiple sclerosis and community controls in Tasmania, Australia. J Neurol 2007;254(5):581-90.
9. Chris H Polman, Stephen C Reingold, Brenda Banwell, Michel Clanet, Jeffrey A Cohen, Massimo Filippi, Ludwig Kappos, Fred D Lublin, Xavier Montalban, Paul O'Connor, Magnhild Sandberg-Wollheim, Alan J Thompson, Emmanuelle Waubant, Brian Weinshenker, Jerry S Wolinsky. Diagnostic criteria for multiple sclerosis: 2010 Revisions to the McDonald criteria Ann Neurol 2011;69(2): 292-302.
10. van der Mei IA, Ponsonby AL, Engelsen O, Pasco JA, McGrath JJ, Eyles DW, Blizzard L, Dwyer T, Lucas R, Jones G. The high prevalence of vitamin D insufficiency across Australian populations is only partly explained by season and latitude. Environ Health Perspect 2007;115(8):1132–9.
11. Smolders J, Damoiseaux J, Menheere P, Hupperts R. Vitamin D as an immune modulator in multiple sclerosis, a review. J Neuroimmunol 2008;194(1-2):7–17.
12. Gale CR, Martyn CN. Migrant studies in multiple sclerosis. Prog Neurobiol 1995;47(4-5):425–48.
13. Van Amerongen BM, Dijkstra CD, Lips P, Polman CH. Multiple sclerosis and vitamin D. Eur J Clin Nutr 2004;58:1095-109.
14. Lucas RM, Ponsonby AL, Dear K, Valery PC, Pender MP, Taylor BV, Kilpatrick TJ, Dwyer T, Coulthard A, Chapman C, van der Mei I, Williams D, McMichael AJ. sun exposure and vitamin D levels may have Role in MS. Neurology 2011;76(6):540-8.
15. Soilu-Hanninen M, Airas L, Mononen I, Heikkila A, Viljanen M. 25-Hydroxyvitamin D level in serum of the onset of Multiple sclerosis. J Multiple Sclerosis 2005;11(3):266-71.