

Supplementation and therapeutic use of vitamin D in patients with multiple sclerosis: Consensus of the Scientific Department of Neuroimmunology of the Brazilian Academy of Neurology

Suplementação e uso terapêutico de vitamina D nos pacientes com esclerose múltipla: Consenso do Departamento Científico de Neuroimunologia da Academia Brasileira de Neurologia

Doralina Guimarães Brum¹, Elizabeth Regina Comini-Frota², Cláudia Cristina F. Vasconcelos³, Elza Dias-Tosta⁴

ABSTRACT

Multiple sclerosis (MS) is an inflammatory, autoimmune, demyelinating, and degenerative central nervous system disease. Even though the etiology of MS has not yet been fully elucidated, there is evidence that genetic and environmental factors interact to cause the disease. Among the main environmental factors studied, those more likely associated with MS include certain viruses, smoking, and hypovitaminosis D. This review aimed to determine whether there is evidence to recommend the use of vitamin D as monotherapy or as adjunct therapy in patients with MS. We searched PUBMED, EMBASE, COCHRANNE, and LILACS databases for studies published until September 9th, 2013, using the keywords “multiple sclerosis”, “vitamin D”, and “clinical trial”. There is no scientific evidence up to the production of this consensus for the use of vitamin D as monotherapy for MS in clinical practice.

Keywords: vitamin D, multiple sclerosis, experimental autoimmune encephalitis.

RESUMO

A esclerose múltipla (EM) é uma doença inflamatória, autoimune, desmielinizante e degenerativa do sistema nervoso central. Estudos epidemiológicos têm identificado associações de hipovitaminose D com doenças autoimunes. O principal objetivo desta revisão é responder se há evidências que indiquem o uso terapêutico de vitamina D em monoterapia para pacientes com EM. Por meio dos sites PUBMED, EMBASE, LILACS e Scielo foram realizadas buscas usando os descritores “vitamin D”, e “multiple sclerosis” até 12/09/2013. Estudos clínicos randomizados, controlados e duplo-cegos foram selecionados para avaliar a resposta terapêutica da vitamina D na EM. Não foram encontradas evidências científicas que justifiquem o uso da vitamina D em monoterapia no tratamento da EM, na prática clínica.

Palavras-chave: Vitamina D, esclerose múltipla, encefalite autoimune experimental.

¹Departamento de Neurologia, Psicologia e Psiquiatria, Faculdade de Medicina de Botucatu, Universidade Estadual Paulista, Botucatu SP, Brazil;

²Hospital das Clínicas, Universidade Federal de Minas Gerais, Belo Horizonte MG, Brazil;

³Universidade Federal do Rio de Janeiro, Rio de Janeiro RJ, Brazil;

⁴Hospital de Base do Distrito Federal, Brasília DF, Brazil.

Correspondence: Doralina G. Brum. Departamento de Neurologia, Psicologia e Psiquiatria da Faculdade de Medicina de Botucatu, Universidade Estadual Paulista - UNESP. Distrito de Rubião Júnior, s/n; 18618-970 Botucatu SP, Brasil. E-mail: dbrum@fmb.unesp.br

Conflict of interest: The Brazilian Academy of Neurology (ABN) is committed to produce clinical guidelines critically and independently. This guideline is part of ABN's continuing education activity. It is based on review of scientific and clinical knowledge. Its purpose is not to address the subject in its entirety. Treatment decision is shared between patient and physician and according to each situation.

Support: This guideline was developed with financial support from the Brazilian Academy of Neurology. None of the authors received honoraria for their participation.

The conflict of interest form for the issue specifically addressed in this article was signed by all authors.

List of contributors: Amilton Antunes Barreira, Antônio Pereira Gomes Neto, Cláudio Roberto Carneiro, Carlos Augusto de Albuquerque Damasceno, Daniel Lima Varela, Damácio Ramón Kaimen-Maciel, Denis Bernardi Bichuetti, Denise Sisterolli Diniz, Eber Castro Correa, Elizabeth Batista da Silva, Fabio Siquineli, Fernando Coronetti Gomes da Rocha, Felipe von Glehn Silva, Fernando Faria Andrade Figueira, Gutemberg Augusto Cruz dos Santos, Heloíse Helena de Figueiredo Siqueira, Jefferson Becker, Leandro Cortoni Calia, José Mauricio Godoy Barreiros, Luiz Domingos Mendes Melges, Marcos Aurélio Moreira, Marcos Papais-Alvarenga, Maria Cecília Aragon de Vecino, Maria Cristina Brandão de Giacomo, Maria Fernanda Mendes, Maria Lúcia Brito Ferreira, Maria Lúcia Vellutini Pimentel, Monica Koncke Fiuza Parolin, Nívea de Macedo Oliveira Morales, Osvaldo J.M. do Nascimento, Paulo Pereira Christo, Regina Maria Papais Alvarenga, Renata Brandt de Souza, Renato Puppi Munhoz, Rogério de Rizo Morales, Sidney Gomes, Solange Maria da G. G. Camargo, Soniza Vieira Alvez-Leon, Suzana Costa Nunes Machado, Tarso Adoni, Thereza Cristina D'Ávila Winckler, Thiago de Faria Junqueira, Yara Dadalti Fragoso, and Yuna de Ribeiro Araújo.

External collaborators: Alessandro Farias (researcher), Eduardo Antônio Donadi (immunologist), and Marcelo de Paula Corrêa (meteorologist).

Received 10 January 2014; Accepted 20 January 2014.

The therapeutic use of vitamin D for treating multiple sclerosis (MS) is a controversial issue that is of interest to physicians, researchers, and patients. The Scientific Department of Neuroimmunology (DCNI) of the Brazilian Academy of Neurology (ABN) organized a meeting on September 12, 2013, to discuss the basic aspects of vitamin D metabolism, results of *in vitro* and experimental studies on experimental autoimmune encephalomyelitis (EAE), and controlled clinical trials of vitamin D in MS. Neurologists and researchers participating in the meeting approved a guideline consensus to guide Brazilian neurologists in the care of patients with MS.

VITAMIN D, MS, AND EAE

Vitamin D is an important hormone for calcium homeostasis and bone metabolism¹. Besides its action in bone tissue, vitamin D has a role in cell differentiation, cell growth inhibition, and immune system modulation². The main source of vitamin D is ultraviolet-B radiation (95%). However, no consensus has been reached on optimal serum vitamin D levels for human metabolic needs^{3,4}. The association between vitamin D and autoimmune diseases and neoplasms has been established in recent years⁵, but this relationship has not yet been fully elucidated.

Multiple sclerosis is an inflammatory, autoimmune, demyelinating, and degenerative central nervous system (CNS) disease, whose geographic and ethnic distribution is characterized by a higher prevalence in northern hemisphere countries, particularly in populations of Caucasian origin⁶.

The predominantly temperate climate in the northern hemisphere with long periods of low solar radiation and the relatively high prevalence of hypovitaminosis D observed in population studies⁷ have led to the hypothesis that this deficiency may explain the geographical distribution of MS. Moreover, it has been suggested that adequate serum levels of vitamin D could help reduce the risk of developing MS^{8,9}.

Even though the etiology of MS has not yet been fully elucidated, there is evidence that genetic^{10,11} and environmental¹² factors interact to cause the disease. Among the main environmental factors studied, those more likely associated with MS include certain viruses¹³, smoking¹⁴, and hypovitaminosis D^{15,16}. The latter is particularly important in the northern hemisphere, where the seasonal variation and subsequent reduction in ultraviolet-B radiation in winter may lead to a higher prevalence of hypovitaminosis D. Some conditions represent risk of hypovitaminosis D in the general population such as long stay indoors, use of sunscreen, and skin pigmentation^{17,18}. Motor limitations associated with later stages of MS may contribute to the occurrence of hypovitaminosis D in this group of patients¹⁹.

Unlike northern hemisphere countries, solar radiation in Brazil is believed to be plentiful in all seasons and regions to

prevent hypovitaminosis D. Thus, the amount of sunlight one is exposed in Brazil should be enough to avoid hypovitaminosis D in healthy individuals when exposed to sunlight even for short periods. Nevertheless, no studies have compared serum vitamin D levels among Brazilian regions, whereas few studies have analyzed serum vitamin D levels in a selected risk group²⁰.

Preliminary experimental studies have demonstrated an immunomodulatory role of vitamin D on human immune cells *in vitro*^{21,22} and in an experimental animal model (EAE)^{23,24}. An *in vitro* study with peripheral blood cells of patients on vitamin D therapy showed that serum levels above 40 ng/ml may exert modulatory action on immune cells²⁰. Additional studies are underway to better understand this immunomodulatory effect on autoimmune diseases.

This review aimed to determine whether there is evidence to recommend the use of vitamin D as monotherapy or as adjunct therapy in patients with MS. We searched PUBMED, EMBASE, COCHRANNE, and LILACS databases for studies published until September 9th, 2013, using the keywords “multiple sclerosis”, “vitamin D”, and “clinical trial”. Randomized controlled clinical trials with vitamin D in patients with MS were included in the analysis.

RANDOMIZED AND CONTROLLED CLINICAL TRIALS WITH VITAMIN D IN THE TREATMENT OF MS

To evaluate the therapeutic response of vitamin D in MS patients, we selected double-blind, randomized, controlled clinical trials from the literature^{25,26-28}. These studies are still scarce and most were not designed to evaluate therapeutic response to vitamin D. Next, we discuss the most relevant studies.

A clinical study conducted in Finland²⁵ in 66 patients with relapsing-remitting multiple sclerosis (RRMS) compared a group with 34 patients using 20,000 IU/week of vitamin D and interferon beta-1b (IFN β -1b) to another group with 32 patients using IFN β -1b only. In that study, primary outcomes included tolerability and safety aspects, and number of new lesions and gadolinium enhancing lesions on MRI scans. Secondary outcomes included clinical parameters such as annual relapse rate and changes in the Expanded Disability Scale Score (EDSS), in addition to other imaging parameters. The authors observed that the treated group showed fewer new T2 lesions, but there were no significant differences in clinical parameters between the two groups after 12 months. However, there was a significant reduction in the number of gadolinium enhancing lesions in the vitamin D group.

Another study, conducted in Norway²⁶, compared bone mineral density, relapse frequency, disease progression, and motor function measures between 35 patients with MS using 20,000 IU of cholecalciferol per week associated with 500

mg/day of calcium and a control group of 33 patients with MS using 500 mg/day of calcium only for two years²⁶. Patients in both groups had been previously using immunomodulatory drugs (interferon beta or glatiramer acetate) for a similar period of time. No differences in annual relapse rate and changes in functional capacity measured by EDSS were observed between the two groups, even though vitamin D levels ranged from 24.72 ng/ml in the placebo group to 49.26 ng/ml in the vitamin D group. The study was not powered to address clinical outcomes¹².

A phase II study developed in Iran²⁷ compared 25 patients with RRMS receiving the active form of vitamin D (calcitriol) at a dose of 0.25 µg/day with patients receiving placebo²⁷. Both groups used conventional immunomodulators. There was no difference in the EDSS between the calcitriol and placebo groups after 12 months followup¹³. It should be noted in that study the small sample size and inclusion criterion of serum 25-hydroxyvitamin D level >40 ng/ml.

A randomized study in Australia²⁸ compared 11 patients with RRMS treated with vitamin D2 in a dose of 6,000 IU twice daily in addition to a daily low-dose (1,000 IU) with 12 patients receiving the 1,000 IU/day dose only²⁸. The neuraxial index of inflammatory activity on MRI was compared between the high-dose and low-dose groups. No significant differences between the groups were detected.

A meta-analysis of the studies cited above detected no difference in the number of relapses between the groups²⁹. The number of new lesions and gadolinium enhancing lesions were compared to serum vitamin D levels in other two studies and the findings were conflicting^{26,30}. Limitations of the studies include different dosages and forms of vitamin D administered.

In contrast to epidemiological and experimental studies, randomized trials on the use of vitamin D in MS showed no significant differences in the parameters of disease activity – relapse rate, EDSS progression, and new or gadolinium enhancing lesions on MRI – between the group receiving vitamin D and groups receiving placebo or a smaller dose of vitamin D. These differences and other contradictions indicate the need to conduct double-blind, randomized, controlled trials in large groups of patients, considering the differences between clinical, neuroimaging, biological, and immunological variables, and powered to accurately estimate the therapeutic efficacy and possible side effects of vitamin D in MS.

VITAMIN D AND OTHER ISSUES

Normal range

The Institute of Medicine (IOM) and the American Society for Endocrinology advocate different levels of vitamin D to maintain bone health: ≥20 ng/ml and ≥30ng/ml,

respectively^{3,4}. There is no consensus on whether bone cells and immune cells require different levels of vitamin D. In addition to the lack of consensus on the normal range values for vitamin D, the toxic serum concentration and the concentration leading up to this condition are also controversial. In adults, doses greater than or equal to 10,000 IU/day are associated with hypercalcemia^{31,32}.

High performance liquid chromatography (HPLC) followed by mass spectrometry is considered the gold standard for analysis of serum 25-OH vitamin D levels. However, the technique is laborious, expensive, and is not available in most Brazilian laboratories. Other methods such as chemiluminescence, enzyme immunoassay, and radioimmunoassay are also used. Thus, variability in results can occur depending on the assay used³³. In Brazil, there is no efficient inter-laboratory validation system, which can also result in great variability in results. Moreover, certain medications such as anti-convulsants and corticosteroids may have a role in reducing serum levels of vitamin D.

SAFETY PROFILE

The safety profile of different serum vitamin D levels has been evaluated in an open, randomized study conducted in Canada³¹. In that study, a group of 25 patients with MS used escalating cholecalciferol (vitamin D3) doses up to 40,000 IU/day, whereas a second group of 24 patients used 4,000 IU/day. Patients in both groups used immunomodulators (interferon beta and glatiramer acetate) in combination with cholecalciferol. The maximum 40,000 IU/day dose was used for up to six months, followed by 10,000 IU/day for three months and gradual suspension over three months. Both groups received calcium (1,200 mg/day) throughout the trial, and serum calcium was determined. Serum 25-hydroxyvitamin D (25-OH-vitamin D) reached a maximum mean above 250 nmol/l (100ng/ml) during the 40,000 IU/day dosing period. No hypercalcemia was detected during the 10,000 IU/day dosing period, even with serum levels ≥ 100 ng/ml, suggesting that that dose is safe (Class level II evidence). In addition, neither serum calcium nor parathormone urinary levels were altered, even when serum concentrations were higher. Further studies are needed to confirm these findings.

VITAMIN D – SIDE EFFECTS

Clinical picture of vitamin D intoxication may include signs and symptoms originating in different systems: nausea and vomiting, anorexia, abdominal pain, constipation; polydipsia, polyuria, dehydration, nephrolithiasis, nephrocalcinosis, nephrogenic diabetes insipidus, chronic interstitial nephritis, acute and chronic renal failure; hypotonia,

paresthesia, confusion, seizures, apathy, coma; arrhythmia, bradycardia, hypertension, cardiomyopathy; muscle weakness, calcification, osteoporosis; and conjunctival calcification³⁴⁻³⁶. Hypercalcemia is the most important side effect, and when observed in the laboratory is suggestive of intoxication³⁷.

During use of vitamin D, in addition to serum calcium, urinary calcium should be assayed periodically. Serum concentration of parathyroid hormone (PTH) should also be determined and must not exceed the lower reference values of normality indicative of suppression, which is a non-recommended condition³⁵.

FINAL CONSIDERATIONS

Considering the body of information presented here, the DCNI/ABN defines the consensus that:

1. It is recommended to dose vitamin D in patients with clinically isolated syndrome and MS, regardless of the stage of disease, particularly those making frequent use of corticosteroids or anticonvulsivants.
2. Peripheral blood levels of vitamin D lower than 30ng/ml should be corrected in patients with MS, at any stage, or in patients with demyelinating isolated syndrome (grade D recommendation).
3. Peripheral blood levels of vitamin D higher than 100 ng/ml should be avoided until new guidelines are established (grade D recommendation).
4. **There is no scientific evidence up to the production of this consensus for the use of vitamin D as monotherapy for MS in clinical practice.** Therefore, currently, vitamin D monotherapy for MS is considered experimental. For its use in clinical trials, these must be approved by the Human Research Ethics Committee, regulated by the National Commission for Ethics in Research (CONEP), approved by the Regional Medical Board, and informed consent should be provided by patients.
5. According to data from *in vitro* studies with peripheral blood cells of patients using vitamin D, serum levels

above 40 ng/ml are likely to cause modulating action on immune cells¹⁷. Based on that evidence, vitamin D supplementation at doses that maintain serum levels of patients between 40 ng/ml and 100 ng/ml may be recommended, as these are safe levels (grade D recommendation).

6. Considering the individual differences in replacement needs and serum levels of vitamin D, that a study in healthy subjects showed that 5,000 IU/day of vitamin D for 15 weeks increased serum levels up to 60ng/ml, and that doses up to 10,000 IU/day were considered safe, we recommend individualized doses until reaching serum levels between 40 ng/ml and 100 ng/ml (grade D recommendation).
7. Considering that low vitamin D serum levels in patients with isolated demyelinating syndrome could affect the relative risk of conversion to MS¹⁶, we recommend the analysis of serum vitamin D levels in those patients and that a correction is made whenever necessary (grade D recommendation).
8. Because vitamin D3 is a secosteroid hormone, its use should be escalated. Moreover, monitoring serum 25-hydroxvitamin D would be extremely important before increasing dosage to determine whether supplementation is actually effective (grade D recommendation).

Acknowledgments

We thank the contributors who performed critical review of the manuscript: Amilton Antunes Barreira, Danilo Lima Varela, Denis B. Bichueti, Felipe von Glehn, Eduardo Antônio Donadi, Gutemberg Augusto Cruz dos Santos, Marcos Papais-Alvarenga, Maria Fernanda Mendes, Maria Cecilia Vecino, Maria Lúcia Vellutini, Paulo Pereira Christo, Thiago Faria Junqueira, Soniza Vieira Alvez Leon, and Yara Dadalti Fragoso (Fragoso, YD). We also thank the invaluable expert assistance of Paulo S. Moraes Júnior in using Microsoft Lync for support in the online meeting.

This manuscript was reviewed by a professional science editor and by a native English-speaking copy editor to improve readability.

References

1. Grant WB, Holick MF. Benefits and requirements of vitamin D for optimal health: a review. *Altern Med Rev* 2005;10:94-111.
2. Bikle DD. Vitamin D regulation of immune function. *Vitam Horm* 2011;86:1-21.
3. Holick MF, Binkley NC, Bischoff-Ferrari HA, et al. Evaluation, treatment, and prevention of vitamin D deficiency: an Endocrine Society clinical practice guideline. *J Clin Endocrinol Metab* 2011;96:1911-1930.
4. Rosen CJ, Abrams SA, Aloia JF, et al. IOM committee members respond to Endocrine Society vitamin D guideline. *J Clin Endocrinol Metab* 2012;97:1146-1152.
5. Holick MF. Vitamin D: important for prevention of osteoporosis, cardiovascular heart disease, type 1 diabetes, autoimmune diseases, and some cancers. *South Med J* 2005;98:1024-1027.
6. Kurtzke J. A reassessment of the distribution of multiple sclerosis. *Acta Neurol Scand* 1975;51:137-157.
7. Hossein-nezhad A, Holick MF. Vitamin D for health: a global perspective. *Mayo Clin Proc* 2013;88:720-755.
8. Goldberg P, Fleming MC, Picard EH. Multiple sclerosis: decreased relapse rate through dietary supplementation with calcium, magnesium and vitamin D. *Med Hypotheses* 1986;21:193-200.
9. Martinelli V, Dalla Costa G, Colombo B, et al. Vitamin D levels and risk of multiple sclerosis in patients with clinically isolated syndromes. *Mult Scler* 2013, Epub Ahead of Print
10. Ebers GC, Bulman DE, Sadovnick AD, et al. A population-based study of multiple sclerosis in twins. *N Engl J Med* 1986;315:1638-1642.

11. Sawcer S, Hellenthal G, Pirinen M, et al. Genetic risk and a primary role for cell-mediated immune mechanisms in multiple sclerosis. *Nature* 2011;476:214-219.
12. Cantorna MT, Mahon BD. Mounting evidence for vitamin D as an environmental factor affecting autoimmune disease prevalence. *Exp Biol Med (Maywood)* 2004;229:1136-1142.
13. Lucas RM, Ponsonby AL, Dear K, et al. Current and past Epstein-Barr virus infection in risk of initial CNS demyelination. *Neurology* 2011;77:371-379.
14. Hedström AK, Sundqvist E, Bäärnhielm M, et al. Smoking and two human leukocyte antigen genes interact to increase the risk for multiple sclerosis. *Brain* 2011;134:653-664.
15. Ascherio A, Munger K. Epidemiology of multiple sclerosis: from risk factors to prevention. *Semin Neurol* 2008;28:17-28.
16. Munger KL, Levin LI, Hollis BW, Howard NS, Ascherio A. Serum 25-hydroxyvitamin D levels and risk of multiple sclerosis. *JAMA* 2006;296:2832-2838.
17. Maeda SS, Saraiva GL, Kunii IS, et al. Factors affecting vitamin D status in different populations in the city of Sao Paulo, Brazil: the Sao Paulo vitamin D Evaluation Study (SPADES). *BMC Endocr Disord* 2013;13:14.
18. Libon F, Cavalier E, Nikkels AF. Skin color is relevant to vitamin D synthesis. *Dermatology* 2013;227:250-254.
19. Smolders J, Menheere P, Kessels A, Damoiseaux J, Hupperts R. Association of vitamin D metabolite levels with relapse rate and disability in multiple sclerosis. *Mult Scler* 2008;14:1220-1224.
20. Arantes HP, Kulak CA, Fernandes CE, et al. Erratum to: correlation between 25-hydroxyvitamin D levels and latitude in Brazilian postmenopausal women: from the Arzoxifene Generations Trial. *Osteoporos Int* 2013;24:2899-2900.
21. Kimball S, Vieth R, Dosch HM, et al. Cholecalciferol plus calcium suppresses abnormal PBMC reactivity in patients with multiple sclerosis. *J Clin Endocrinol Metab* 2011;96:2826-2834.
22. Allen AC, Kelly S, Basdeo SA, et al. A pilot study of the immunological effects of high-dose vitamin D in healthy volunteers. *Mult Scler* 2012;18:1797-1800.
23. Farias AS, Spagnol GS, Bordeaux-Rego P, et al. Vitamin D3 induces IDO(+) tolerogenic DCs and enhances Treg, reducing the severity of EAE. *CNS Neurosci Ther* 2013;19:269-277.
24. Correale J, Ysrraelit MC, Gaitán MI. Vitamin D-mediated immune regulation in multiple sclerosis. *J Neurol Sci* 2011;311:23-31.
25. Soilu-Hänninen M, Aivo J, Lindström BM, et al. A randomised, double blind, placebo controlled trial with vitamin D3 as an add on treatment to interferon β -1b in patients with multiple sclerosis. *J Neurol Neurosurg Psychiatry* 2012;83:565-571.
26. Kampman MT, Steffensen LH, Mellgren SI, Jørgensen L. Effect of vitamin D3 supplementation on relapses, disease progression, and measures of function in persons with multiple sclerosis: exploratory outcomes from a double-blind randomised controlled trial. *Mult Scler* 2012;18:1144-1151.
27. Shaygannejad V, Janghorbani M, Ashtari F, Dehghan H. Effects of adjunct low-dose vitamin d on relapsing-remitting multiple sclerosis progression: preliminary findings of a randomized placebo-controlled trial. *Mult Scler Int* 2012;2012:452-541.
28. Stein MS, Liu Y, Gray OM, et al. A randomized trial of high-dose vitamin D2 in relapsing-remitting multiple sclerosis. *Neurology* 2011;77:1611-1618.
29. James E, Dobson R, Kuhle J, Baker D, Giovannoni G, Ramagopalan SV. The effect of vitamin D-related interventions on multiple sclerosis relapses: a meta-analysis. *Mult Scler* 2013;19:1571-1579.
30. Mowry EM, Waubant E, McCulloch CE, et al. Vitamin D status predicts new brain magnetic resonance imaging activity in multiple sclerosis. *Ann Neurol* 2012;72:234-240.
31. Gallo S, Comeau K, Vanstone C, et al. Effect of different dosages of oral vitamin D supplementation on vitamin D status in healthy, breastfed infants: a randomized trial. *JAMA* 2013;309:1785-1792.
32. Burton JM, Kimball S, Vieth R, et al. A phase I/II dose-escalation trial of vitamin D3 and calcium in multiple sclerosis. *Neurology* 2010;74:1852-1859.
33. Roth HJ, Schmidt-Gayk H, Weber H, Niederau C. Accuracy and clinical implications of seven 25-hydroxyvitamin D methods compared with liquid chromatography-tandem mass spectrometry as a reference. *Ann Clin Biochem* 2008;45:153-159.
34. Koul PA, Ahmad SH, Ahmad F, Jan RA, Shah SU, Khan UH. Vitamin d toxicity in adults: a case series from an area with endemic hypovitaminosis d. *Oman Med J* 2011;26:201-204.
35. Zittermann A, Prokop S, Gummert JF, Börgermann J. Safety issues of vitamin D supplementation. *Anticancer Agents Med Chem* 2013;13:4-10.
36. Ashizawa N, Arakawa S, Koide Y, Toda G, Seto S, Yano K. Hypercalcemia due to vitamin D intoxication with clinical features mimicking acute myocardial infarction. *Intern Med* 2003;42:340-344.
37. Bell DA, Crooke MJ, Hay N, Glendenning P. Prolonged vitamin D intoxication: presentation, pathogenesis and progress. *Intern Med J* 2013;43:1148-1150.