http://www.pjbs.org

PJBS

ISSN 1028-8880

Pakistan Journal of Biological Sciences

ANSInet
Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

OPEN ACCESS Pakistan Journal of Biological Sciences

ISSN 1028-8880
DOI: 10.3923/pjbs.2019.406.411

Research Article
The Combined Effect of Vitamin D Deficiency and Hyperparathyroidism on Postural Stability among Healthy Adult Males

[1]Osama A. Kensara, [2]Omar F. Helal, [2,3]Ehab M. Abd El-Kafy, [1]Khloud J. Ghafouri, [4]Mazen M. Ghaith, [5]Fatmah J. Alsolami, [1]Alaa H. Qadhi, [6]Hamed R. Takruri, [1]Abdelelah S. Jazar, [7]Mohammed O. Ibrahim, [8]Awfa Y. Alazzeh and [1]Firas S. Azzeh

[1]Department of Clinical Nutrition, Faculty of Applied Medical Sciences, Umm Al-Qura University, P.O. Box 7067, 21955 Makkah, Saudi Arabia
[2]Department of Physical Therapy, Faculty of Applied Medical Sciences, Umm Al Qura University, Makkah, Saudi Arabia
[3]Department of Physical Therapy for Disturbances of Growth and Developmental Disorders in Children and Its Surgery, Faculty of Physical Therapy, Cairo University, Giza, Egypt
[4]Department of Laboratory Medicine, Faculty of Applied Medical Sciences, Umm Al-Qura University, Makkah, Saudi Arabia
[5]Faculty of Nursing, Umm Al-Qura University, Saudi Arabia
[6]Department of Dietetics and Nutritional Science, Faculty of Agriculture, University of Jordan, Jordan
[7]Department of Nutrition and Food Technology, Faculty of Agriculture, Mu'tah University, Karak, Jordan
[8]Department of Clinical Nutrition, Faculty of Applied Medical Sciences, University of Ha'il, Ha'il, Saudi Arabia

Abstract

Background and Objective: Vitamin D has been shown to improve muscle strength and bone health; consequently, be important for maintaining good balance. Possible risk factors related to postural stability in young adults still underdetermined. However, this study was designed to determine the effect of vitamin D status on postural stability. **Materials and Methods:** A cross-sectional study of 704 healthy young adult males were enrolled in this study. Dynamic balance was measured as overall stability index (OSI) using biodex balance system (BBS). Vitamin D deficiency was defined when its serum level $\leq$20 ng mL^{-1}. The effect size was measured for vitamin D, parathyroid hormone (PTH) and the interaction between vitamin D and PTH (VTD*PTH) with respect to the OSI values. Correlations between variables were examined according to the beta standardised coefficient (β) and the effect size was measured using the partial eta-squared (η^2) test. **Results:** About 95, 3.8 and 1.2% of individuals had deficient, insufficient and normal vitamin D levels, respectively. Vitamin D had no significant effect to OSI, but PTH exhibited a significant correlation with OSI (adjusted $\beta = 0.095$, $p = 0.038$). A significant effect size was observed between OSI and PTH (adjusted partial $\eta^2 = 0.012$, $p = 0.038$) and between OSI and VTD*PTH (adjusted partial $\eta^2 = 0.034$, $p<0.001$). **Conclusion:** A significant interaction of vitamin D deficiency and high PTH on postural stability is detected among healthy adult males.

Key words: Vitamin D, parathyroid hormone, overall stability index, postural stability

Citation: Osama A. Kensara, Omar F. Helal, Ehab M. Abd El-Kafy, Khloud J. Ghafouri, Mazen M. Ghaith, Fatmah J. Alsolami, Alaa H. Qadhi, Hamed R. Takruri, Abdelelah S. Jazar, Mohammed O. Ibrahim, Awfa Y. Alazzeh and Firas S. Azzeh, 2019. The combined effect of vitamin D deficiency and hyperparathyroidism on postural stability among healthy adult males. Pak. J. Biol. Sci., 22: 406-411.

Corresponding Author: Firas S. Azzeh, Department of Clinical Nutrition, Faculty of Applied Medical Sciences, Umm Al-Qura University, P.O. Box 7067, 21955 Makkah, Saudi Arabia Tel: +966-540833661 Fax: +966-125720000-4227

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

The relationship between the physiological level of vitamin D and postural stability is universally accepted[1]. It is well established that vitamin D is an important physiological determinant of bone mineralisation and muscle strength. Furthermore, a low level of vitamin D is associated with poor neuromuscular strength and poor physical performance, which predispose individuals to falls[2]. Several observational studies have reported associations between chronically low circulating levels of 25-hydroxyvitamin D_3 (25-(OH)-D3) and poor lower-extremity function[3], muscle strength[4], contraction speed and appendicular muscle mass[5]. Several lines of evidence also support the association of hypovitaminosis D with slower nerve conduction[6] and poorer executive functions[7].

Correspondingly, vitamin D supplementation was associated with better muscle performance and higher muscle mass in a group of elderly individuals after the vitamin D serum level was corrected to higher than 75 nmol L^{-1} (30 ng mL^{-1}) as compared to the control group[8]. Another previous study showed that the positive effect of vitamin D on fall prevention is even greater than its effect on muscle mass and muscle performance[9]. In particular, Bischoff-Ferrari *et al.*[9] demonstrated that fall prevention could be mediated by vitamin D supplementation when postural and dynamic balance are also modified. In this case, vitamin D plus calcium supplementation was associated with a 60% reduction in the rate of falls. Hence, the evidence reveals that vitamin D deficiency may be associated with impaired postural control.

Emerging evidence also suggests an inverse association between vitamin D status and motor imbalance[10], although the mechanisms underlying this relationship have not been completely elucidated. This might explain why hypovitaminosis D is accompanied by various poor health outcomes[10].

An independent relationship was additionally found between postural instability and parathyroid hormone (PTH) level in several previous cross-sectional studies[11,12]. In this regard, Sambrook *et al.*[11] demonstrated that serum baseline levels of 25-(OH)-D3 and PTH were significantly associated with falls among a large group of elderly individuals and that the effect of PTH continued to be a significant predictor of falls even after multivariate analysis adjusted for health status and postural stability independently of 25-(OH)-D3 level. Similar findings were also reported by Stein *et al.*[12], who demonstrated a direct positive association between PTH level and predisposition to falls in a group of ambulant nursing home and hostel residents.

However, to our knowledge, no detailed research has attempted to explain the physiological effect of vitamin D and PTH levels on postural stability among healthy young adults. Accordingly, the aim of the current study was to investigate the effect of vitamin D, PTH and both on dynamic balance in this group of individuals.

MATERIALS AND METHODS

Participants: Seven hundred and four young adult men aged 18-35 years old were participated in this study from November, 2013 to April, 2014. Inclusion criteria for participants were young adults males aged 18-35 years, apparently healthy, normoglycemic, non-smokers and had normotension. Exclusion criteria were any individual who taking any drugs known to affect bone metabolism or to interfere with balance stability or vitamin D supplementation within the last six months.

The study was approved by the Ethics Committee of Umm Al-Qura University (approval number ASE-21-2013). Before starting the experimental work, all eligible participants gave written informed consent.

Study design: In this observational cross-sectional study, the postural stability of all participants was evaluated in the laboratory and blood samples were taken. Participants were categorized into 3 different groups according to their vitamin D serum level. These groups corresponded[13] to (i) Deficient: $\leq$20 ng mL^{-1}, (ii) Insufficient: >20-29 ng mL^{-1} and (iii) Normal: $\geq$30 ng mL^{-1}.

The overall stability index (OSI), which is an indicator of dynamic balance, was measured for all participants using a Biodex balance system (BBS-SD; model 950-441, Jan-2013, Shirley, NY). The system was operated according to the Biodex balance falls risk assessment. The stability level of the BBS device was fixed at 8 and the other testing conditions were previously described by Azzeh *et al.*[14]. The reference OSI value for the selected age group is 0.8-2.3.

Blood analysis: Blood test were performed as previously published by Kensarah *et al.*[15] research. Briefly, blood samples were collected from all participants after an overnight fast and serum was separated within 30 min of collection. The samples were then stored at -20°C until analysis. Serum concentrations of both 25-(OH)-D3 and PTH were measured using a fully automated Elecsys instrument (Hitachi-Roche-Elecsys, Indianapolis, USA). Vitamin D and PTH tests were validated and the inter-assay coefficients of variation were 1.59 and 0.91%, respectively. All other biochemical parameters,

including serum alkaline phosphatase (ALP), calcium (Ca) and phosphorus (P) were measured with commercially available enzymatic and turbidimetric kits using a Dimension instrument (Siemens Dimension RXL, Erlangen, Germany). All analysis were performed at the clinical biochemistry laboratory of Al-Noor Specialty Hospital (Makkah, Saudi Arabia).

Statistical analysis: All statistical analysis were conducted in SPSS version 20. The Kolmogorov-Smirnov normality test was used to determine the normality of distribution. Continuous variables with non-normal distribution were transformed to normal distribution, then statistical analysis were performed. Comparisons between groups were performed based on the one-way Analysis of Variance (ANOVA) test. Relationships between dependant and independent variables were examined according to the beta standardised coefficient (β) determined by linear regression tests. A $p<0.05$ was considered statistically significant. The effect size was measured using the partial eta-squared (η^2) test by Univariate General Linear Model and was determined for vitamin D, PTH and the interaction between vitamin D and PTH (VTD*PTH) with respect to the OSI values. The effect sizes were interpreted to the criteria of Draper[16] where partial $\eta^2<0.01$= negligible, 0.01-0.05 = small, 0.06-0.13 = medium and >0.14 = large, respectively.

RESULTS

Characteristics of participants: Table 1 shows the characteristics of participants according to their vitamin D status. Overall, the average age was 19.9±0.09 years and the average BMI was 25.58±0.33 kg m^{-2}. The average values of PTH, Ca, P and ALP of the whole sample were within the normal value; only the mean vitamin D was below than the normal range (11.8±0.29 ng mL^{-1}). About 95% (n= 669) of individuals had deficient vitamin D levels and 3.8% (n= 27) had insufficient vitamin D levels. Only 1.2% (n= 8) had a normal vitamin D level.

Vitamin D and parathyroid correlations with OSI: The correlations between OSI and other biochemical parameters are presented in Table 2. Only PTH exhibited a significant correlation with OSI and this remained significant after adjusting for age and BMI. Vitamin D showed a non-significant and weakly negative correlation with OSI. The estimated effect sizes of vitamin D, PTH and VTD*PTH on OSI values are presented in Table 3. The adjusted partial η^2 values did not show a significant effect between OSI and vitamin D. On the other hand, a significant effect size was observed between OSI and PTH (adjusted partial $\eta^2 = 0.012$, $p = 0.038$) and between OSI and VTD*PTH (adjusted partial $\eta^2 = 0.034$, $p<0.001$).

DISCUSSION

This study is one of the few studies that have investigated the effect of vitamin D deficiency on balance among young adults. This research findings show that high serum PTH levels were significantly associated with the results of the impaired balance test. The interaction of hyperparathyroidism and hypovitaminosis D had a significant correlation with poor balance stability.

This study was hypothesized to determine the possible negative effect of low vitamin D on balance stability through its role on muscles and the central nervous system. It is well known that 1,25-dihydroxyvitamin D3 (1,25-(OH)2D3), the active vitamin D metabolite, binds to a vitamin-D-specific nuclear receptor (VDR) in muscle tissue, leading to *de novo* protein synthesis and muscle cell growth[17-19]. Vitamin D may improve muscle strength and function as well as balance as a

Table 1: Characteristics of participants according to vitamin D status

	Vitamin D status				
Parameters	Deficient (n = 669)	Insufficient (n = 27)	Normal (n = 8)	Total (n = 704)	p-value
Age (years)	19.99±0.09	19.79±0.4	20.00±0.7	19.98±0.09	0.869
Weight (kg)	75.07±1.06	68.82±3.96	73.00±6.75	74.81±1.03	0.384
Height (cm)	170.82±0.31	169.04±1.42	172.75±2.77	170.77±0.31	0.497
BMI (kg m^{-2}) (normal; 18-25)	25.65±0.34	23.95±1.47	24.48±3.07	25.58±0.33	0.483
OSI (normal; 0.8-2.3)	0.91±0.02	0.98±0.19	1.03±0.37	0.91±0.02	0.941
Vitamin D (ng mL^{-1}) (normal; ≥30)	10.98±0.19[c]	23.09±0.65[b]	45.11±1.7[a]	11.81±0.29	<0.001
PTH (pg mL^{-1}) (normal; 14-65)	40.94±0.95	35.60±3.2	26.60±4.3	40.80±0.92	0.155
Ca (mg dL^{-1}) (normal; 8.8-10.8)	9.08±0.03	8.86±0.2	8.68±0.32	9.07±0.03	0.347
P (mg dL^{-1}) (normal; 4.5-5.5)	3.81±0.09	3.69±0.19	3.78±0.43	3.80±0.09	0.99
ALP (u L^{-1}) (normal; 60-170)	102.38±2.24	111.93±8.71	96.75±7.65	102.68±2.2	0.932

Values are expressed as mean and standard error of mean, p-values were determined by ANOVA test, BMI: Body mass index, OSI: Overall stability index, PTH: Parathyroid hormone, Ca: Calcium, P: Phosphorus, ALP: Alkaline phosphatase

Table 2: Relationship between OSI and other parameters (n = 704)

Independent variables	Unadjusted β (p-value)	Adjusted β (p-value)
Vitamin D	-0.026 (0.616)	-0.06 (0.189)
PTH	0.158 (0.002)	0.095 (0.038)
Ca	0.015 (0.77)	0.009 (0.852)
P	-0.038 (0.468)	-0.019 (0.679)
ALP	0.033 (0.533)	0.039 (0.386)

Beta standardized coefficient (β) was determined by a linear regression test, the dependent variable is OSI, OSI: Overall stability index, PTH: Parathyroid hormone, Ca: Calcium, P: Phosphorus, ALP: Alkaline phosphatase, Adjustment was performed for age and BMI

Table 3: Estimates of effect size for the main studied parameters on OSI

	Unadjusted		Adjusted	
Parameters	Partial η^2	p-value	Partial η^2	p-value
Vitamin D	0.001	0.616	0.005	0.189
PTH	0.025	0.002	0.012	0.038
VTD*PTH	0.019	0.007	0.034	<0.001

Partial eta squared (η^2) test was performed by Univariate General Linear Model Dependent variable is OSI, VTD*PTH: Interaction between vitamin D and parathyroid hormone, PTH: Parathyroid hormone, VTD: Vitamin D as 25-(OH)-D3 adjustment was performed for age and BMI

result of improved strength. Additionally, VDRs have been found in some parts of the brain, especially in the cortical, subcortical and spinal motor zones[20,21]. According to this evidence, vitamin D may also play a role in the cerebral processes involved in postural balance. Study results found that vitamin D alone had a non-significant effect on OSI, but PTH had a significant and negative outcome to OSI.

It is well established that serum PTH is an independent predictor of time to first fall among frail elderly individuals. Elevated PTH levels are linked to sarcopenia in older men and women and muscle function impairment and are also associated with the loss of hip flexor and knee extensor strength[22]. Hence, in many studies on elderly individuals, a clear relation has been demonstrated between vitamin D deficiency and postural instability[2,23,24]. However, in one observational study, low serum vitamin D levels did not predict new onset of disability or loss of muscle strength in older women[25]. In another study, low serum vitamin D levels were found to play a role in the development sarcopenia among elderly men and women[22]. In any case, vitamin D deficiency is only one factor contributing to muscle weakness in the elderly[25].

Furthermore, both vitamin D and PTH play key roles in Ca homeostasis. The pattern of PTH secretion and synthesis is higher in individuals with vitamin D deficiency than those with normal vitamin D levels. In the kidneys, PTH stimulates the hydroxylation of 25-(OH)-D3 to its active form, 1,25-(OH)2D3, which facilitates intestinal absorption of Ca[26]. High levels of PTH enhance osteoclast activity and the urinary excretion of P, thus negatively impacting bone density. Accordingly, the interrelationships among low dietary Ca, skeletal muscle wasting, primary or secondary hyperthyroidism, chronic kidney disease and inadequate vitamin D status may influence PTH concentration and these factors may independently or jointly contribute to high serum PTH concentrations[26]. In the current study, the effect sizes of PTH and vitamin D on OSI were small, yet these effects might be clinically important.

It is well known that vitamin D deficiency/insufficiency increases PTH serum levels. In the current study, the PTH serum level was normal among most participants, yet 7.1% of participants had hyperparathyroidism. Study results showed that the effect size of vitamin D on PTH was 0.06 ($p<0.001$). In different studies, the level of vitamin D shown to influence PTH has been inconsistent, varying from 25-75 nmol L^{-1} (10-30 ng mL^{-1})[27]. Differing definitions of vitamin D deficiency/insufficiency may at least partially explain the conflicting results of meta-analysis addressing vitamin D status and outcomes. Additionally, while vitamin D deficiency/insufficiency is highly prevalent, the magnitude of hypovitaminosis D may vary depending on diet, the studied population and regional and seasonal considerations[28]. Some research has pointed to the role of genes in circulating vitamin D levels and, accordingly, in differences in vitamin D levels among distinct populations[29,30]. In Saudi Arabia, the prevalence of hypovitaminosis D is widespread and severe[29]. In this case, genetic factors might play a role, as firstly reported by Sadat-Ali *et al.*[31], who found that individuals with the GG allele in three SNPs, VDR rs2228570, CYP2R1 rs10741657 and GC rs4588, had significantly lower levels of 25-(OH)-D3 compared to individuals with normal levels.

This study had several limitations. Only young adult men were enrolled into the study, so study results cannot be extrapolated to older individuals. Second, bone markers such as serum osteocalcin and urine N-telopeptides were not measured, which are influenced by vitamin D level. Third, BMI, OSI, vitamin D and PTH cut-off points have not been set for the Middle Eastern population; therefore, internationally adopted ranges are used in this work.

CONCLUSION

In summary, this study showed that balance control in healthy young adult males was affected by both low levels of serum 25-(OH)-D3 and high PTH levels. This relationship should be explored in larger and different populations by national campaigns to enhance the awareness about balance stability at young adults. The effectiveness of vitamin D supplementation on postural stability for young adults should be determined in further studies.

SIGNIFICANCE STATEMENT

This study discovered the potential combination of low vitamin D and high PTH in decreasing the postural stability in young adult males. This study will help the researchers to identify another possible mechanism affecting dynamic balance. Thus, a new idea on the use of vitamin D supplements to enhance postural stability in young adult males may be achieved.

REFERENCES

1. O'Donnell, S., D. Moher, K. Thomas, D.A. Hanley and A. Cranney, 2008. Systematic review of the benefits and harms of calcitriol and alfacalcidol for fractures and falls. J. Bone Miner. Metab., 26: 531-542.
2. Boersma, D., O. Demontiero, Z.M. Amiri, S. Hassan and H. Suarez *et al*., 2012. Vitamin D status in relation to postural stability in the elderly. J. Nutr. Health Aging, 16: 270-275.
3. Rebolledo, B.J., J.A. Bernard, B.C. Werner, A.K. Finlay and B.U. Nwachukwu *et al*., 2018. The association of vitamin D status in lower extremity muscle strains and core muscle injuries at the National Football League Combine. Arthroscopy: J. Arthroscopic Relat. Surg., 34: 1280-1285.
4. Annweiler, C., A.M. Schott-Petelaz, G. Berrut, R.W. Kressig, S. Bridenbaugh, F.R. Herrmann and O. Beauchet, 2009. Vitamin D deficiency-related quadriceps weakness: Results of the Epidémiologie de l'Ostéoporose Cohort. J. Am. Geriatr. Soc., 57: 368-369.
5. LeBoff, M.S., C.M. Donlon, S.H. Chou, N. Cook, J. Buring and J.E. Manson, 2019. Vitamin D and Omega-3 Fatty Acid Trial 2017: Addressing Effects on Muscle and Bone. In: Nutritional Influences on Bone Health: 10th International Symposium, Weaver, C.M., H. Bischoff-Ferrari, R.M. Daly and M.S. Wong (Eds.). Springer International Publishing, Switzerland, ISBN: 978-3-319-98463-6, pp: 11-24.
6. Annweiler, C., A.M. Schott, G. Berrut, V. Chauvire, D. Le Gall, M. Inzitari and O. Beauchet, 2010. Vitamin D and ageing: Neurological issues. Neuropsychobiology, 62: 139-150.
7. Annweiler, C., M. Montero-Odasso, D.J. Llewellyn, S. Richard-Devantoy, G. Duque and O. Beauchet, 2013. Meta-analysis of memory and executive dysfunctions in relation to vitamin D. J. Alzheimer's Dis., 37: 147-171.
8. Bischoff-Ferrari, H.A., B. Dawson-Hughes, H.B. Staehelin, J.E. Orav and A.E. Stuck *et al*., 2009. Fall prevention with supplemental and active forms of vitamin D: A meta-analysis of randomised controlled trials. Br. Med. J., Vol. 339. 10.1136/bmj.b3692.
9. Bischoff-Ferrari, H.A., M. Conzelmann, H.B. Stahelin, W. Dick and M.G. Carpenter *et al*., 2006. Is fall prevention by vitamin D mediated by a change in postural or dynamic balance? Osteoporosis Int., 17: 656-663.
10. Muscogiuri, G., B. Altieri, C. Annweiler, G. Balercia and H.B. Pal *et al*., 2017. Vitamin D and chronic diseases: The current state of the art. Arch. Toxicol., 91: 97-107.
11. Sambrook, P.N., J.S. Chen, L.M. March, I.D. Cameron and R.G. Cumming *et al*., 2004. Serum parathyroid hormone predicts time to fall independent of vitamin D status in a frail elderly population. J. Clin. Endocrinol. Metab., 89: 1572-1576.
12. Stein, M.S., J.D. Wark, S.C. Scherer, S.L. Walton and P. Chick *et al*., 1999. Falls relate to vitamin D and parathyroid hormone in an Australian nursing home and hostel. J. Am. Geriatr. Soc., 47: 1195-1201.
13. Holick, M.F., N.C. Binkley, H.A. Bischoff-Ferrari, C.M. Gordon and D.A. Hanley *et al*., 2011. Evaluation, treatment and prevention of vitamin D deficiency: An Endocrine Society clinical practice guideline. J. Clin. Endocrinol. Metab., 96: 1911-1930.
14. Azzeh, F.S., O.A. Kensara, O.F. Helal and E.M. Abd El-Kafy, 2017. Association of the body mass index with the overall stability index in young adult Saudi males. J. Taibah Univ. Med. Sci., 12: 157-163.
15. Kensarah, O., A. Jazar and F. Azzeh, 2015. Hypovitaminosis D in healthy toddlers and preschool children from Western Saudi Arabia. Int. J. Vitamin Nutr. Res., 85: 50-60.
16. Draper, S., 2019. Effect size. http://www.psy.gla.ac.uk/~steve/best/effect.html
17. Bischoff, H.A., M. Borchers, F. Gudat, U. Duermueller, R. Theiler, H.B. Stahelin and W. Dick, 2001. *In situ* detection of 1,25-dihydroxyvitamin D receptor in human skeletal muscle tissue. Histochem. J., 33: 19-24.
18. Endo, I., D. Inoue, T. Mitsui, Y. Umaki and M. Akaike *et al*., 2003. Deletion of vitamin D receptor gene in mice results in abnormal skeletal muscle development with deregulated expression of myoregulatory transcription factors. Endocrinology, 144: 5138-5144.
19. Gil, A., J. Plaza-Diaz and M.D. Mesa, 2018. Vitamin D: Classic and novel actions. Ann. Nutr. Metab., 72: 87-95.
20. Kiraly, S.J., M.A. Kiraly, R.D. Hawe and N. Makhani, 2006. Vitamin D as a neuroactive substance. Scient. World J., 6: 125-139.
21. Kalueff, A.V., S. Minasyan, T. Keisala, M. Kuuslahti, S. Miettinen and P. Tuohimaa, 2006. The vitamin D neuroendocrine system as a target for novel neurotropic drugs. CNS Neurol. Disord., 5: 363-371.
22. Visser, M., D.J.H. Deeg and P. Lips, 2003. Low vitamin D and high parathyroid hormone levels as determinants of loss of muscle strength and muscle mass (sarcopenia): The Longitudinal Aging Study Amsterdam. J. Clin. Endocrinol. Metab., 88: 5766-5772.
23. Bischoff-Ferrari, H.A., B. Dawson-Hughes, W.C. Willett, H.B. Staehelin, M.G. Bazemore, R.Y. Zee and J.B. Wong, 2004. Effect of vitamin D on falls: A meta-analysis. J. Am. Med. Assoc., 291: 1999-2006.

24. Mastaglia, S.R., M. Seijo, D. Muzio, J. Somoza, M. Nunez and B. Oliveri, 2011. Effect of vitamin D nutritional status on muscle function and strength in healthy women aged over sixty-five years. J. Nutr. Health Aging, 15: 349-354.
25. Verreault, R., R.D. Semba, S. Volpato, L. Ferrucci, L.P. Fried and J.M. Guralnik, 2002. Low serum vitamin D does not predict new disability or loss of muscle strength in older women. J. Am. Geriatr. Soc., 50: 912-917.
26. Hossein-Nezhad, A. and M.F. Holick, 2013. Vitamin D for health: A global perspective. Mayo Clinic Proc., 88: 720-755.
27. Gonzalez-Gross, M., J. Valtuena, C. Breidenassel, L.A. Moreno and M. Ferrari *et al*., 2012. Vitamin D status among adolescents in Europe: The healthy lifestyle in Europe by nutrition in adolescence study. Br. J. Nutr., 107: 755-764.
28. Halfon, M., O. Phan and D. Teta, 2015. Vitamin D: A review on its effects on muscle strength, the risk of fall and frailty. BioMed. Res. Int., Vol. 2015. 10.1155/2015/953241.
29. Hussain, A.N., A.H. Alkhenizan, M. El Shaker, H. Raef and A. Gabr, 2014. Increasing trends and significance of hypovitaminosis D: A population-based study in the Kingdom of Saudi Arabia. Arch. Osteoporosis, Vol. 9, No. 1. 10.1007/s11657-014-0190-3.
30. Pike, J.W., M.B. Meyer, N.A. Benkusky, S.M. Lee and H.S. John *et al*., 2016. Genomic determinants of vitamin D-regulated gene expression. Vitamins Hormones, 100: 21-44.
31. Sadat-Ali, M., H.A. Al-Turki, M.Q. Azam and A.H. Al-Elq, 2016. Genetic influence on circulating vitamin D among Saudi Arabians. Saudi Med. J., 37: 996-1001.