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Original Research Article

A Case-control Study to Assess the Role of Vitamin D in Pre-hypertension and its Association with Cellular Senescence

Farhat Ali[1], Pronoy Kumar Biswas[2]

[1]Assistant Professor, Department of Physiology, Shri Ramakrishna Institute of Medical Sciences and Sanaka Hospital, Durgapur, West Bengal, India

[2]Assistant Professor, Department of Physiology, Shri Ramakrishna Institute of Medical Sciences and Sanaka Hospital, Durgapur, West Bengal, India

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Corresponding author: Dr Pronoy Kumar Biswas

Conflict of interest: Nil

Abstract

Aim: To explore the link between Vitamin D and cellular senescence measured with the enzyme telomerase in pre-HTN.

Methodology: After obtaining the institute ethics committee clearance from Shri Ramakrishna Institute of Medical Sciences and Sanaka Hospital, volunteers were recruited from medical students. Inclusion criteria for the pre-hypertensive group (pre-HTN) (n = 50) were both genders between 18 and 25 years of age with SBP between 120 and 139 mmHg and DBP between 80 and 89 mmHg in apparently healthy individuals. The controls (n = 50) population were healthy individuals with 18–25 years of age with SBP between 100 and 119 mmHg and DBP between 60 and 79 mmHg. The volunteers were asked to not participate in heavy exercises, not drink alcohol and coffee 1 day before the data collection. Baseline, anthropometric parameters were recorded before recording of the BP by sphygmomanometer as per standard protocol. Then, 5 ml of blood was collected, allowed to clot, and subjected to centrifugation to separate the serum. Serum was stored at –80°C for processing of Vitamin D and telomerase levels as per the instructions provided in the commercially available kits.

Results: The study population included 100 apparently healthy individuals. 50 were pre-hypertensive with the age of 20.65±1.56and the age of controls was 19.98±1.25. Among 50 in each group, 28 males, 22 females in pre-HTN group and 26 males, 24 females in the control group. A significant difference was not found between-group differences in height and waist-hip ratio. Significantly low levels of Vitamin D ($P < 0.001$) and high telomerase ($P < 0.001$) were seen in pre-HTN group when compared to controls. Low levels of Vitamin D have no correlation with BMI, waist-hip ratio, DBP, and MAP. However, significant correlation was seen with HR, SBP, PP, RPP, and telomerase. High telomerase levels have correlation with waist-hip ratio, SBP, DBP, MAP, and RPP but no significant correlation was seen with BMI, HR, and PP.

Conclusion: From this study, it can be concluded that reduced Vitamin D levels in pre-HTN may cause derangements of cardiovascular homeostatic mechanism, enhance the speed of cellular senescence measured by telomerase.

Keywords: Telomerase, Hypertension, Vitamin D.

Introduction

Hypertension is a common health problem, one of the leading costs to the health care system, and a significant cause of mortality and morbidity worldwide [1]. Hypertension is also one of the most common and influential risk factors of cardiovascular disease including myocardial infarction, cerebral stroke, congestive heart failure, peripheral vascular disorders and kidney disease [2]. It has been estimated that eliminating high blood pressure would reduce the occurrence of stroke by 35% and heart attacks by 18% [3, 4]. To reduce the burden of hypertension, a multicomponent lifestyle intervention that includes weight loss, increased physical activity, restricted sodium and alcohol consumption, and adherence to a Dietary Approach to Stop Hypertension like diet with plenty of fruits, vegetables, and low-fat dairy items and little saturated fat is needed [5]. Moreover, improved vitamin D status has been proposed as an easily modifiable risk factor [6].

According to many reports in India, there is 70–100% deficiency of Vitamin D, playing pivotal role in a higher risk of development of bone diseases and several non-communicable diseases like cancer, metabolic diseases, and cardiovascular pathologies, [7] playing role as a major cause for higher health-care burden on Indian health-care system [8]. At a cellular level, vitamin D exerts antihypertensive effects through improved endothelial function [9-11], reduced production of pro-inflammatory cytokines [12], reduced activity of the renin-angiotensin-aldosterone system and reduced levels of parathyroid hormone [13-15].

In May 2003, the joint national commission (JNC 7), to emphasize the perception of close abnormal blood-pressure levels, introduced the term pre-hypertension (pre-HTN) which described as 120–139 mmHg of systolic blood pressure (SBP) of 80–89 mmHg of diastolic blood pressure (DBP) [16,17]. It helps such pre-HTN individuals could modify their lifestyle to postpone hypertension [18]. Reports have demonstrated that pre-HTN caused higher risk if cardiovascular morbidity independent to other risk factors [17,19]. Many original research articles, reviews, and meta-analyses reported the pre-HTN prevalence in varied groups and its relation with risks and benefits of treatment in cardiovascular diseases. The prevalence of pre-HTN in India ranges from 20 to 80% [20,21].

Observational studies consistently find an association between lower serum 25-hydroxyvitamin D (25(OH)D) concentrations and higher blood pressure levels [11]. However, there have been conflicting results from trials investigating vitamin D supplementation as an intervention to improve blood pressure [23]. Information on the importance of vitamin D in cellular senescence in individuals with pre-HTN was not available. Hence, in this study, we planned to explore the link between Vitamin D and cellular senescence measured with the enzyme telomerase in pre-HTN. Further, an attempt was made to describe the importance of vitamin D in blood pressure.

Materials and Methods

After obtaining the institute ethics committee clearance from Shri Ramakrishna Institute of Medical Sciences and Sanaka Hospital, volunteers were recruited from medical students. Inclusion criteria for the pre-hypertensive group (pre-HTN) (n =50) were both genders between 18 and 25 years of age with SBP between 120 and 139 mmHg and DBP between 80 and 89 mmHg in apparently healthy individuals. The controls (n = 50) population were healthy individuals with 18–25 years of age with SBP between 100 and 119 mmHg and DBP between 60 and 79 mmHg.

Individuals suffering from diabetes, hypertension, endocrine disorders, kidney diseases, and hypertensive patients already receiving medication were not considered to take part in this research.

The volunteers were asked to not participate in heavy exercises, not drink alcohol and coffee 1 day before the data collection. Baseline, anthropometric parameters were recorded before recording of the BP by sphygmomanometer as per standard protocol.[24] Then, 5 ml of blood was collected, allowed to clot, and subjected to centrifugation to separate the serum. Serum was stored at –80°C for processing of Vit-D and telomerase levels as per the instructions provided in the commercially available kits.

Statistical analysis was done to analyze the data. To study the between-group differences, independent t-test, to assess the correlation of vitamin D with telomerase and other parameters, Pearson's correlation coefficient analysis was applied.

Results

The study population included 100 apparently healthy individuals. 50 were pre-hypertensive with the age of 20.65±1.56and the age of controls was 19.98±1.25. Among 50 in each group, 28 males, 22 females in pre-HTN group and 26 males, 24 females in the control group. A significant difference was not found between-group differences in height and waist-hip ratio. However, pre-HTN group subject's BMI ($P < 0.001$) and weight ($P < 0.001$) was more compared to controls. In pre-HTN group, significantly higher HR ($P < 0.001$), SBP ($P < 0.001$), DBP ($P < 0.001$), MAP ($P < 0.001$), and RPP ($P < 0.001$) were seen when compared to controls. No significant difference was seen in PP but it was slightly high in pre-HTN group and negatively associated with Vitamin D.

Table 1: Comparison of anthropometric characteristics between pre-HTN and controls

Parameter	Pre-HTN (*n*=50)	Controls (*n*=50)	*P*-value
Age	20.65±1.56	19.98±1.25	0.465
Gender (male/female)	28/22	26/24	NA
Height (cm)	170.29±9.82	166.72±7.48	0.398
Weight (kg)	65.46±10.36	58.46±8.46	<0.001
BMI (k/m^2)	25.15±4.74	22.74±4.82	<0.001
Waist to hip ratio	0.92±0.10	0.87±0.08	0.487

Table 2: Comparison of cardiovascular parameters between pre-HTN and controls

Parameter	Pre-HTN (*n*=50)	Controls (*n*=50)	*P*-value
HR (BPM)	89.46±5.75	80.46±4.83	<0.001
SBP (mmHg)	126.15±4.86	114.2±4.57	<0.001
DBP (mmHg)	85.64±4.06	74.36±3.96	<0.001
PP (mmHg)	40.74±5.98	39.02±5.54	0.180
MAP (mmHg)	96.54±2.88	89.78±3.74	<0.001
RPP	10943.86±706.74	9084.72±492.85	<0.001

Significantly low levels of Vitamin D ($P < 0.001$) and high telomerase ($P < 0.001$) were seen in pre-HTN group when compared to controls. Low levels of Vitamin D have no correlation with BMI, waist-hip ratio, DBP, and MAP. However, significant correlation was seen with HR, SBP,

PP, RPP, and telomerase. High telomerase levels have correlation with waist-hip ratio, SBP, DBP, MAP, and RPP but no significant correlation was seen with BMI, HR, and PP.

Table 3: Comparison of Vitamin D and telomerase levels between pre-HTN and controls

Parameter	Pre-HTN (*n*=50)	Controls (*n*=50)	*P*-value
Vitamin D (ng/ml)	16.28±4.28	21.02±6.28	0.051
Telomerase (IU/ml)	36.88±17.92	7.04±5.91	<0.001

Discussion

Vitamin D deficiency has recently emerged as a public health problem, affecting almost 50% of the population worldwide [25]. In addition to the reduced exposition to sunlight [26], also genetic and environmental factors have been suggested as a cause of this pandemic, such as pollution, diet, sedentary life style and stress [27]. Moreover, vitamin D is no longer considered as only a pivotal mediator of calcium metabolism and skeletal health, but it also regulates several cell functions, including differentiation and metabolism. This aspect may explain the reason why hypovitaminosis D has been proved to be an independent risk factor for overall mortality in various cohort analyses[28], whereas vitamin D supplementation significantly reduced mortality [29].

Although the effects of vitamin D on blood pressure have been known for several decades, some physiological aspects on the modulation of vascular cells and the vascular tone still remain to be clarified. Martins *et al* [30] showed an increased prevalence of hypertension associated with low serum 25 (OH) vitamin D levels in 15088 subjects from this cohort. Forman *et al* [31] firstly reported an increased risk of incident hypertension in 1811 subjects selected from these two matched cohorts at 4-year follow-up (pooled RR = 3.18, 95%CI: 1.39-7.29, $P < 0.05$). In addition, the investigators extended this risk prediction, as a surrogate, to the overall study population including 38388 man from HPFS (adjusted RR = 2.31, 95%CI: 2.03-2.63, $P < 0.05$) and 77531 women from the NHS 2 (adjusted RR = 1.57, 95%CI: 1.44-1.72, $P < 0.05$). Afterwards, the same authors also designed a prospective nested case-control study including 1484 normotensive women from the NHS 2 that confirmed the previous results (inter-quartile OR = 1.66, 95%CI: 1.11-2.48, *P* value for trend = 0.01)[32].

This study volunteers had no statistically significant variations in their age, height, and waist-hip ratio. Weight and body mass index were significantly high in the pre-HTN group when compare to age-matched and gender-matched controls. HR, SBP, DBP, PP, MAP, RPP, and telomerase levels were high, Vitamin D levels were low in the pre-HTN group. Low Vitamin D levels were negatively correlated with telomerase, HR, SBP, and PP and it was independent of age, gender, BMI, and hip-waist ratio.

Earlier reports have shown that higher Vitamin D is related to longer telomere length, which underscores the conceivably advantageous impacts of this hormone on cell senescence and age-related conditions [33]. In this study, cellular senescence was assessed using telomerase. This enzyme attempts to inhibit the process of telomere shortening [34]. Since the cell telomere loss appears to result from cell division just to a fractional degree, different components, particularly oxidative stress, were attested to assume a job in the expanded rate for shortening of telomeres [35]. The exact mechanism by which lower Vit-D levels are associated with this cellular senesce is hypothesized dependent on the perceptions recommends that the degrees of the

telomerase may really be related to oxidative stress, with higher oxidative stress prompting higher telomerase levels. Cells of nearly complex organism may not have an ability to divide. This marvel was depicted by Hayflick in 1961 [36]. The quantity of potential divisions – around fifty – was named the "Hayflick Limit" and at times is called cell senescence. Just because it was valued that cells could be mortal (typical cells) or unfading (tumor cells). The results of our study showed an association between Vit-D and BP is consistent with previously conducted studies [37,38] and stretch out the relationship to pre-HTN, a prior stage when essential anticipation is powerful.

Conclusion

From this study, it can be concluded that reduced Vitamin D levels in pre-HTN may cause derangements of cardiovascular homeostatic mechanism, enhance the speed of cellular senescence measured by telomerase.

References

1. Mittal, B.V., Singh, A.K. Hypertension in the developing world: Challenges and opportunities. Am. J. Kidney Dis. 2010, 55, 590–598.
2. Holick, M.F. 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.
3. Colley, R.C., Garriguet, D., Janssen, I., Craig, C.L., Clarke, J., Trembla, M.S. Physical activity of Canadian adults: Accelerometer results from the 2007 to 2009 Canadian health measures survey. Health Rep. 2011, *22*, 7–14.
4. Warburton, D.E., Charlesworth, S., Ivey, A., Nettlefold, L., Bredin, S.S. A systematic review of the evidence for Canada's physical activity guidelines for adults. Int. J. Behav. Nutr. Phys. Act. 2010, 7, 39.
5. Elmer, P.J., Obarzanek, E., Vollmer, W.M., Simons-Morton, D., Stevens, V.J., Young, D.R., Lin, P.H., Champagne, C., Harsha, D.W., Svetkey, L.P., *et al*. Effects of Comprehensive Lifestyle Modification on Diet, Weight, Physical Fitness, and Blood Pressure Control: 18-Month Results of a Randomized Trial. Ann. Intern. Med. 2006, 144, 485–495.
6. Hosseinpanah F., Yarjanli M, Sheikholeslami F, Heibatollahi M, Eskandary P.S., Azizi F. Associations between vitamin d and cardiovascular outcomes. Tehran lipid and glucose study. Atherosclerosis 2011, 218, 238–242.
7. Ritu G, Gupta A. Vitamin D deficiency in India: Prevalence, causalities and interventions. Nutrients 2014,6:729-75.
8. Basit S. Vitamin D in health and disease: A literature review. Br J Biomed Sci 2013,70:161-72.
9. Sugden, J.A., Davies, J.I., Witham, M.D., Morris, A.D., Struthers, A.D. Vitamin D improves endothelial function in patients with type 2 diabetes mellitus and low vitamin D levels. Diabet. Med. 2008, 25, 320–325.
10. Tarcin, O., Yavuz, D.G., Ozben, B., Telli, A., Ogunc, A.V., Yuksel, M., Toprak, A., Yazici, D., Sancak, S., Deyneli, O., *et al*. Effect of vitamin D deficiency and replacement on endothelial function in asymptomatic subjects. J. Clin. Endocrinol. Metab. 2009, 94, 4023–4030.
11. Vaidya, A., Forman, J.P. Vitamin D and hypertension: Current evidence and future directions. Hypertension 2010, 56, 774–779.
12. Schleithoff, S.S., Zittermann, A., Tenderich, G., Berthold, H.K., Stehle, P., Koerfer, R. Vitamin D supplementation improves cytokine profiles in patients

with congestive heart failure: A double-blind, randomized, placebo-controlled trial. Am. J. Clin. Nutr. 2006, 83, 754–759Brickman, A., Nyby, M., von Hungen, K., Eggena, P., Tuck, M. Parathyroid hormone platelet calcium and blood pressure in normotensive subjects. Hypertension 1991, 18, 176–182.
13. Ullah, M.I., Uwaifo, G.I., Nicholas, W.C., Koch, C.A. Does vitamin D deficiency cause hypertension? Current evidence from clinical studies and potential mechanisms. Int. J. Endocrinol. 2010, 2010, 579640.
14. Forman, J.P., Williams, J.S., Fisher, N.D. Plasma 25-hydroxyvitamin D and regulation of the renin-angiotensin system in humans. Hypertension 2010, 55, 1283–1288.
15. Scragg, R., Sowers, M., Bell, C. Serum 25-hydroxyvitamin D, ethnicity, and blood pressure in the third national health and nutrition examination survey. Am. J. Hypertens. 2007, 20, 713–719.
16. Chobanian AV, Bakris GL, Black HR, Cushman WC, Green LA, Izzo JL, *et al*. The seventh report of the joint national committee on prevention, detection, evaluation, and treatment of high blood pressure: The JNC 7 report. JAMA 2003,289:2560-72.
17. Liszka HA, Mainous AG, King DE, Everett CJ, Egan BM. Prehypertension and cardiovascular morbidity. Ann Fam Med 2005,3:294-9.
18. Lewington S, Clarke R, Qizilbash N, Peto R, Collins R, Prospective Studies Collaboration. Age-specific relevance of usual blood pressure to vascular mortality: A meta-analysis of individual data for one million adults in 61 prospective studies. Lancet 2002,360:1903-13.
19. Huang Y, Wang S, Cai X, Mai W, Hu Y, Tang H, *et al*. Prehypertension and incidence of cardiovascular disease: A meta-analysis. BMC Med 2013,11:177.
20. Yadav S, Boddula R, Genitta G, Bhatia V, Bansal B, Kongara S, *et al*. Prevalence and risk factors of pre-hypertension and hypertension in an affluent North Indian population. Indian J Med Res 2008,128:712-20.
21. Asmathulla S, Rajagovindan D, Sathyapriya V, Pai B. Prevalence of prehypertension and its relationship to cardiovascular disease risk factors in Puducherry. Indian J PhysiolPharmacol 2011,55:343-50.
22. Fraser, A., Williams, D., Lawlor, D.A. Associations of serum 25-hydroxyvitamin D, parathyroid hormone and calcium with cardiovascular risk factors: Analysis of 3 NHANES cycles (2001–2006). PLoS ONE 2010, 5, e13882.
23. Wu, S.H., Ho, S.C., Zhong, L. Effects of vitamin D supplementation on blood pressure. South Med. J. 2010, 103, 729–737.
24. Chobanian AV, Bakris GL, Black HR, Cushman WC, Green LA, Izzo JL, *et al*. Seventh report of the joint national committee on prevention, detection, evaluation, and treatment of high blood pressure. Hypertension 2003,42:1206-52.
25. Holick MF. Vitamin D deficiency. N Engl J Med. 2007,357:266–281.
26. Lucas RM, Ponsonby AL, Dear K, Valery PC, Taylor B, van der Mei I, McMichael AJ, Pender MP, Chapman C, Coulthard A, *et al*. Vitamin D status: multifactorial contribution of environment, genes and other factors in healthy Australian adults across a latitude gradient. J Steroid BiochemMol Biol. 2013,136:300–308.
27. Holick MF. Environmental factors that influence the cutaneous production of vitamin D. Am J ClinNutr. 1995,61:638S–645S.
28. Pludowski P, Holick MF, Pilz S, Wagner CL, Hollis BW, Grant WB, Shoenfeld Y,

Lerchbaum E, Llewellyn DJ, Kienreich K, *et al*. Vitamin D effects on musculoskeletal health, immunity, autoimmunity, cardiovascular disease, cancer, fertility, pregnancy, dementia and mortality-a review of recent evidence. Autoimmun Rev. 2013,12:976–989.

29. Amer M, Qayyum R. Relationship between 25-hydroxyvitamin D and all-cause and cardiovascular disease mortality. Am J Med. 2013,126:509–514.
30. Martins D, Wolf M, Pan D, Zadshir A, Tareen N, Thadhani R, Felsenfeld A, Levine B, Mehrotra R, Norris K. Prevalence of cardiovascular risk factors and the serum levels of 25-hydroxyvitamin D in the United States: data from the Third National Health and Nutrition Examination Survey. Arch Intern Med. 2007,167:1159–1165.
31. Forman JP, Giovannucci E, Holmes MD, Bischoff-Ferrari HA, Tworoger SS, Willett WC, Curhan GC. Plasma 25-hydroxyvitamin D levels and risk of incident hypertension. Hypertension. 2007,49:1063–1069.
32. Forman JP, Curhan GC, Taylor EN. Plasma 25-hydroxyvitamin D levels and risk of incident hypertension among young women. Hypertension. 2008,52:828–832.
33. Richards JB, Valdes AM, Gardner JP, Paximadas D, Kimura M, Nessa A, *et al*. Higher serum Vitamin D concentrations are associated with longer leukocyte telomere length in women. Am J ClinNutr 2007,86:1420-5.
34. vonZglinicki T. Telomeres and replicative senescence: Is it only length that counts? Cancer Lett 2001,168:111-6.
35. VonZglinicki T. Oxidative stress shortens telomeres. Trends BiochemSci 2002,27:339-44.
36. Hayflick L. The limited in vitro lifetime of human diploid cell strains. Exp Cell Res 1965,37:614-36.
37. Forman JP, Curhan GC, Taylor EN. Plasma 25-hydroxyvitamin D levels and risk of incident hypertension among young women. Hypertension 2008,52:828-32.
38. Scragg R, Sowers M, Bell C. Serum 25-hydroxyvitamin D, ethnicity, and blood pressure in the third national health and nutrition examination survey. Am J Hypertens 2007,20:713-9.