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The Effect of Vitamin D Supplementation on the Progression of Benign Prostatic Hyperplasia: A Randomized Controlled Trial

Running Head: Vitamin D Supplementation & the Progression of BPH

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Abstract

Background and aims: Inflammation and proliferation are the cause of benign prostatic hyperplasia (BPH) and are the key components of its mechanism of action. In this study we sought to determine the role of 25-hydroxyvitamin D in BPH, because of its anti-inflammatory activities, and its effect on prostate volume and BPH symptoms.

Methods: This randomized clinical trial (RCT) was conducted on 108 participants >50 years of age who had either asymptomatic or mild BPH symptoms according to the International Prostate Symptom Score (IPSS) questionnaire. Patients were randomly divided into two groups, intervention and control. The intervention group received 50000 units of vitamin D3 and the control group received a placebo every two weeks for six months. Prostate ultrasound, routine clinical examinations, toucher rectal (TR), and laboratory tests were performed for all patients. After six months, the patients underwent another ultrasound evaluation, measurement of prostate-specific antigen (PSA) levels and completed the IPSS. Results of the evaluations before and after the intervention were compared between the groups using the chi-square, t-test, and logistic regression analysis. Repeated measure analysis was used to evaluate the effect of vitamin D intervention on the changes in the IPSS score.

Results: The mean age of the participants was 56 ± 9 years. In the control group, the mean prostate volume was higher compared to the intervention group ($p<0.001$). The control group had a higher mean PSA level than the intervention group ($p<0.001$). Although the IPSS score decreased over time in both groups, analysis of variance showed that the amount of change or decrease in IPSS score in the intervention group was significantly more than the control group ($p<0.001$).

Conclusions: The results of our study support the effect of vitamin D in reducing prostate volume and PSA levels, and in improving BPH symptoms. Further studies are needed to confirm these findings to verify the use of vitamin D as a treatment for BPH.

Keywords: Vitamin D, Benign Prostate Hypertrophy, Inflammation, Proliferation

Introduction

Benign prostatic hyperplasia (BPH) is the most common cause of urinary incontinence in a substantial number of older men (1, 2). BPH is more common in North America and Europe than in Asia and China. Differences in global prevalence indicate the importance of environmental factors such as dietary habits in the development of BPH (3). BPH is a health problem that, in addition to medical costs for diagnosis and treatment, has indirect costs that include work absences for these people and reductions in their quality of life (4). Despite the tremendous impact on public health, the cause, pathophysiology, and necessary interventions to treat BPH are not entirely clear and there are no therapeutic or preventive guidelines (2). Dietary and nutritional factors may affect the onset and symptoms of BPH through various mechanisms (5).

Older men have decreased serum testosterone levels along with increased levels of estrogen, prolactin, luteinizing hormone (LH), and follicle-stimulating hormone (FSH). Estrogen increases the number of androgen receptors in the prostate and inhibits androgen metabolism (6). Androgens increase prostate cell proliferation and inhibit planned cell death, which leads to an increase in prostate volume (2, 7).

BPH is associated with chronic inflammation and increased expressions of pro-inflammatory cytokines such as interleukin-15 (8) in stromal cells, interleukin-17 in T cells (9), interferon-

gamma in basal cells and stromal cells, and IL-8 cells in growth factor-derived epithelial cells (10).

The active form of vitamin D, 1,25(OH)2D, binds to the vitamin D receptor (VDR) and performs a variety of biological activities such as calcium and bone metabolism, proliferation, and cell differentiation into different immune cells for immune system activity. VDR is expressed in different types of cells, including prostate and urogenital cells (11-14). As a result, VDR agonists have been introduced to treat BPH. Calcitriol analogs that have anti-proliferative properties can be effective treatments for BPH (15). Vitamin D analogs are an important therapeutic target for lower urinary tract symptoms due to their ability to reduce growth factor signaling (16, 17). In general, researchers have suggested the use of vitamin D and analogs in combination with 5α-reductase and α-blockers (18).

An association between vitamin D and the prostate was first established by the observation of an epidemiologic correlation between an increase in incidence and mortality from prostate cancer in patients with vitamin D deficiency (19). However, a meta-analysis concluded that 25OHD is directly correlated with prostate cancer incidence but inversely with prostate cancer mortality (20) since, vitamin D increases absorption of calcium and phosphorus from the gastrointestinal tract (21). The prostate was then identified by the expression of 1-hydroxylase and VDR as a site for vitamin D synthesis (22). It has been observed that serum 25-OH D levels are inversely associated with overall prostate volume and enlarged prostate gland ($\geqslant$40 gram), especially in men with benign prostatic disease (23, 24). Also, another study declared that there is a relationship between the presence of vitamin D deficiency and prostate growth-associated urinary symptoms. The patients with vitamin D deficiency had a significantly higher prostate

volume, aldosterone, PSA value, IPSS score, and a significantly lower maximum urinary flow versus free of vitamin D deficiency patients (25).

Increasing vitamin D intake from the diet or supplements reduces the prevalence of BPH. Vitamin D binds to specific receptors in the prostate and bladder, and reduces inflammation and prostate growth (14). It has been reported that a 6000 IU dose red day of Vitamin D analogs can reduce the prostate volume in BPH patients (17, 26).

Vitamin D has anti-proliferative and anti-inflammatory properties; therefore, it appears that supplementation with this vitamin may be a successful treatment for BPH symptoms. Vitamin D deficiency is widespread in Iran. There is an elevated incidence of BPH in men over the age of 50 years and a lack of medical treatments that reduce the severity of prostate symptoms. To date, no randomized clinical trial (RCT) studies have been performed in Iran due to the different VDR polymorphisms in this country. In addition, BPH imposes a tremendous cost on Iran's health care system. Therefore, we conducted this RCT to investigate the effect of vitamin D as a supplement on BPH progression.

Materials and Methods

This RCT was conducted at Ziaeian Hospital, Tehran University of Medical Sciences, Tehran, Iran for 22 months, from April, 2018 to March, 2020. All men over the age of 50 years who referred to the Family Physician and Urology Clinics with asymptomatic BPH or mild BPH symptoms based on the International Prostate Symptom Score (IPSS) were eligible to enter the study.

This study was performed in accordance with the principles of the Declaration of Helsinki. Approval was granted by Ethics Committee of Tehran University of Medical Sciences (Ethical code: IR.TUMS.MEDICINE.REC.1397.257). The trial registered at Iranian Registry of Clinical Trials (code# IRCT20180922041089N3).

Inclusion and exclusion criteria

Patients were considered eligible for study entry if they met the following inclusion criteria: signed the approved informed consent; age over 50 years; BPH diagnosis via ultrasound; and asymptomatic or mild BPH based on IPSS questionnaire. The exclusion criteria were: body mass index (BMI) >35 or weight loss greater than 10% of body weight within the previous six months; histories of kidney, bladder or urinary tract stones; prostate or bladder cancer or prostate surgery; acute or chronic prostatitis or recurrent urinary tract infections; histories of acute urinary retention or recurrent catheters within the past three months; use of gonadotropin-releasing hormone (GnRH), anti-androgenic agonists, or any hormonal drugs within the previous six months, or the use of herbal or non-herbal medications for the control and treatment of BPH; histories of chronic diseases (cerebrovascular accident, hypertension, chronic obstructive pulmonary disease [COPD], liver disease, renal failure, cancer, etc.; use of any medications that affect vitamin D metabolism (corticosteroids, phenytoin, phenobarbital, or regular use of vitamin D supplements) over the past three months; use of diuretics such as Lasix (furosemide), or alcohol abuse.

Collected data

Questioners and all participant were blinded to randomization (double-blind). The following data were collected from the participants:

1. Demographic questionnaire (age, sex, education, marital status, weight, height, history of systemic disease, history of psychiatric illness, drug, and drug use).
2. IPSS questionnaire.
3. Routine and toucher rectal (TR) clinical examinations.
4. Prostate ultrasound.
5. Laboratory analyses: fasting blood sugar (FBS), blood urea nitrogen (BUN), and creatinine (Cr) (Biotecnica autoanalyzer, BT3000 model, Italy), complete blood count (CBC) (MEK6400 -Cell Counters from Nihon Kohden, Japan), prostate-specific antigen (PSA) using enzyme linked immunosorbent assay (ELISA) (Pishtaz Teb, Tehran, Iran), and urinalysis (U/A).
6. BMI. (The standing height was measured in centimeters without shoes with Seka's wall height gauge; weight was measured in kilograms with a Seka scale that had an accuracy of 500 grams. The patients had minimal clothing and no shoes or heavy accessories such as mobile phones and bags.)

We calculated the sample size of 54 for each group based on a study by Caretta and colleagues (27), the two-mean test for a quantitative variables in two independent communities, and the 10% attrition risk. Considering the potential reasons of discontinue the study process that were not considered in exclusion criteria, data was collected from more participants than the calculated amount for the sample size. A total of 180 people assessed for eligibility that 132 people were eligible that eight of them tended to change their diet and physical activity styles; so, 124 men entered the study, from which 16 patients were excluded for the following: 7 received medications from their urologists due to exacerbation of urinary clinical symptoms; 2 had vitamin D levels above 100 ng/ml after the third month of the study; 4 people did not return

for the final follow up visit; and 3 patients return too late for final examinations. Therefore, we evaluated the results from 108 people at the end of the sampling period that there were 54 patients assigned to the intervention group and 54 to the control group (Figure 1)

Individuals who met the entry criteria were enrolled in the study after providing informed consent. We used simple random sampling to assign the participants to either the intervention or the control group. The lottery method was used to place 66 green papers and 66 red papers in a box. Green was intended for the intervention group and red for the control group. The questioner, blind to the color assignment, gave each patient a sheet of paper at random; so, there was equal chance of being selected in each of the intervention and control groups. A family physician's assistant, blinded to the group in which the participant was, performed complete clinical and ultrasound examinations, and collected the IPSS data.

Study participants were instructed to abstain from any medications or supplements other than the study medication and placebo for six months. All participants were randomly assigned to the study and had a diet and physical activity routine in their lives.

At the beginning of the study, each person in the intervention group received 50 000 units of oral vitamin D3 supplementation (28) and participants in the control group received a placebo medication every two weeks for six months. The placebo and vitamin D supplement were made by the same company (Zahravi Pharmacutical Company, Tehran, Iran) and all their components except the presence or absence of vitamin D were exactly the same. Patients were excluded from the study if they did not follow this medication schedule for any reason (drug intolerance, etc.).

Three months after the study began, the patients were tested for vitamin D levels using ELISA (Pishtaz Teb, Tehran, Iran). The cutoff value for deficiency was considered as <12 ng/ml. Those

with serum levels of 25-hydroxy vitamin D more than 100 ng/ml were excluded from the study because this levels are toxic based on Harrison's Principles of İnternal Medicine (29). Also, three months after the start of the study, all patients were questioned and followed up for regular medication usage.

After the completion of the study medications, patients underwent an ultrasound evaluation, assessment of PSA levels using ELISA (Pishtaz Teb, Tehran, Iran) and completed the IPSS questionnaire. The results were compared with the results of the baseline assessment.

The IPSS is a seven-question tool used for screening, rapid diagnosis, follow-up, and recommendations for the management and treatment of BPH. The questions pertain to symptoms a person has experienced in the past month and include the feeling of emptying the bladder, frequent urination, stopping the flow of urine, urinary urgency, poor flow, and waking up at night to urinate. The IPSS can be used at various times to compare the progression and severity of symptoms over months and years. Each question has a score of 0 to 5 depending on the number of times each symptom is experienced, and a total symptom score of 0 to 35 (0–7 mild, 8–19 moderate, and 10–35 severe) (30).

Statistical analysis

SPSS software (version 23.0 for Windows; IBM SPSS Statistics, Armonk, NY, USA) was used for data analysis and to calculate the main and sub-objectives. The results were expressed for quantitative variables as mean and standard deviation (mean ± SD) and qualitative variables as

relative numbers and frequency. The chi-square, t-test, and logistic regression analysis were used for data analysis. P<0.05 indicated statistical significance.

Results

The study population consisted of 108 males (n=54 per group). The participants in the control group had a mean age of 56 ± 9 years and the mean age of the intervention group participants was 57 ± 9 years. The mean BMI in the intervention group was 27 ± 3 and in the control group, it was 27 ± 4. Table 1 lists the patients' qualitative demographic information.

There was a significant difference between the education status of the two groups (p=0.08). In the intervention group, 27.8% of the participants had a university education, whereas only 5.6% of the control group had a university education. This difference was taken into consideration for regression analysis as a confounder.

Vitamin D supplementation and prostate volume

The mean prostate volumes in vitamin D group before and after intervention were 39±20 and 37±21 cc, respectively; these volumes in the control group were 33±15 and 36±16 cc, respectively. In the control group, the mean prostate volume was 4.65 cc higher than the mean prostate volume in the intervention group (95% CI: 2.11–7.19, p<0.001). This difference was independent of the ultrasound assessment of the prostate volume before the intervention. At the study onset, the mean prostate volume of the control group was 0.97 cc more than the intervention group (p<0.001). Table 2 lists the differences in prostate volume after adjustments for age and diabetes, and according to the primary and secondary ultrasound results in the two groups.

The prostate volume in the intervention group was significantly smaller than the control group based on age ($p<0.001$) and diabetes ($p<0.001$).

Vitamin D supplementation and prostate-specific antigen (PSA) levels

The mean PSA levels in vitamin D group before and after intervention were 1.8±1.7 and 1.6±1.6 ng/ml, respectively; these volumes in the control group were 2.6±7 and 3±8 ng/ml, respectively. The mean PSA level was 0.45 ng/ml higher in the control group compared to the intervention group (95% CI: 0.21–0.70, p-value<0.001). This difference was independent of the mean PSA level before intervention, which was 1 ng/ml higher in the control group ($p<0.001$). Table 3 shows the changes in PSA levels by adjusting for age and diabetes in both groups.

The PSA in the intervention group was significantly lower than the control group based on age ($p<0.001$) and diabetes ($p<0.001$).

Vitamin D supplementation and International Prostate Symptom Score (IPSS)

The mean IPSS score in the intervention group at baseline (time 0) was 7 ± 6. Three months after the start of the study (time 1), the mean IPSS score was 5.6 ± 5.6, and at the end of the study (time 2), it was 5 ± 5.

In the control group, the mean IPSS at the beginning of the study (time 0) was 6 ± 4. Three months after the start of the study (time 1), the mean IPSS score was 6 ± 4 and at the end of the study (time 2), it was 5 ± 4.

Repeated measurement was used to evaluate the effect of vitamin D intervention on the changes in IPSS. Although the IPSS values decreased over time in both groups, the results of repeated analysis of variance showed that the amount of change or decrease in IPSS score in the

intervention group was significantly higher than the control group ($p<0.001$). The effect of supplementation on the IPSS score in terms of age and diabetes in both groups are presented in Table 4.

Repeated measurement was used to evaluate the effect of vitamin D supplementation on IPSS score changes with age and diabetes. The analysis of variance results showed that the amount of change or decrease in IPSS score in the intervention group was significantly higher than the control group in terms of age and diabetes ($p<0.001$).

Discussion

Studies of the effect of 25 (OH) vitamin D supplementation on improving BPH symptoms have increased since 2002. Vitamin D analogs were previously used to investigate BPH because of the possibility of tissue calcification with vitamin D supplementation (15). However, in recent years the mechanisms of vitamin D have been determined and it has been used as a supplement in studies. Nevertheless, few studies have been conducted worldwide to determine the effects of vitamin D supplementation instead of its analogs on BPH (24). The current study is the first RCT in Iran to assess the effects of vitamin D on BPH.

We observed that the intervention group had decreased prostate volume and lower PSA levels than the control group ($p<0.001$). These results were significant after adjustments for age and diabetes. The results indicated that vitamin D could be an effective treatment for improving the course of BPH because of its anti-inflammatory mechanisms, increased expression of apoptotic genes, and other mechanisms. The patients' clinical symptoms were assessed by the IPSS questionnaire at the beginning of the study, three months after the onset of the study, and at the

end of the study. The clinical symptoms improved in the intervention group. Participants in the intervention group had significantly lower mean IPSS scores ($p<0.001$).

In several studies, the positive effects of vitamin D analogs on inhibition of cellular prostate growth and various mechanisms of apoptosis in prostate cells have been reported (31-33). In line with the results of our research, the findings of a study conducted by Cretta et al found an inverse relationship between 25-OH-vitamin D plasma levels and IPSS scores, while a significant positive linear correlation was present between IPSS and age, PSA level, and prostate volume (27).

Zhang et al. conducted a study in China to determine the prostate volume and changes in urine flow in men with vitamin D deficiency compared to men with normal vitamin D levels. Their case-control study enrolled 322 men, from which 231 (71.7%) had vitamin D deficiency. The group that was deficient in vitamin D had significantly higher prostate volume, aldosterone, PSA levels, and IPSS values. The maximum urine flow was significantly lower. Two-way logistic regression analysis showed that vitamin D deficiency was inversely related to BPH symptoms. This study suggested that vitamin D deficiency may be a marker of BPH (25).

An observational study examined the association of serum PSA and prostate volume with serum vitamin D levels. The results of this study showed that serum vitamin D levels were inversely related to total prostate volume and an enlarged prostate (23). In this study, the regulated logistic regression model for age, serum PSA and 5α-reductase inhibitors indicated that the prostate volume was inversely related to serum vitamin D levels, which was consistent with the results of the present study. Two limitations of the above-mentioned study (23) were the cross-sectional design and not taking into consideration the impact of diabetes on the results.

A clinical trial conducted by Safwat et al. in Egypt (34) enrolled 196 men per group. The intervention group received vitamin D supplementation. The PSA level in the intervention group was significantly lower than the control group (p=0.05). This result was consistent with our findings. However, in their study, prostate volume did not change with vitamin D supplementation. The above mentioned study is the only clinical trial of vitamin D and BPH. The present results also showed that PSA levels in the intervention group were significantly lower than the control group. However, participants in our study had asymptomatic or mild BPH symptoms whereas the study by Safwa et al. included 389 patients with simple BPH and moderate to severe symptoms, which could explain the difference between the results.

In addition to vitamin D, researches have focused on the associations with BPH and dietary patterns, and other macronutrients and micronutrients. In one study conducted by Kristal et al., patients who consumed a diet that contained high total fat and red meat consumption had increased risk for developing BPH. In contrast, a diet with proper amounts of protein and vegetables reduced the risk of BPH. The study also found no link between the use of antioxidants, vitamin E supplements, vitamin C, and selenium with BPH. However, the use of zinc and vitamin D supplements has been associated with a reduced risk of BPH (35).

Vitamin D has an inhibitory effect on the Rho A/Rho-associated protein kinase (ROCK) pathway, along with cyclooxygenase 2 expression and prostaglandin E2 production in BPH stromal cells (32, 36). An increased vitamin D intake from diet and supplements has been linked to a reduced prevalence of BPH (37). A study by Murphy et al, reported that the use of vitamin D analogs and supplements has shown that prostate volume decreases in BPH patients. Controlling for age, serum PSA, 5-α reductase inhibitors use, obesity and prostate cancer diagnosis, prostate volume was inversely associated with vitamin D using serum vitamin D as a continuous and

categorical variable. Logistic regression model also demonstrated an inverse association between vitamin D (continuous and categorical) and prostate volume ⩾40 grams (23). Para-clinical and clinical trial results indicated that vitamin D reduces BPH cells and the proliferation of prostate cells, PSA levels, and IPSS scores (38). Therefore, it is necessary to pay special attention to vitamin D intake in patients with BPH. There is a high prevalence of BPH. Therefore, in older individuals without BPH, vitamin D supplementation is suggested for prevention of BPH.

Limitations of this study included budget constraints that prevented further evaluations. Also, we included patients with asymptomatic or minimal symptoms of BPH in this study. This might affect the results because of the possibility that BPH was not progressive in these stages. Further studies should separately compare mild, moderate, and severe BPH. The trial registered at Iranian Registry of Clinical Trials (code# IRCT20180922041089N3).

Conclusion

In this study, we evaluated the effect of vitamin D supplementation on prostate volume, PSA levels, and patients' symptoms according to the IPSS questionnaire. We found that vitamin D supplementation significantly reduced prostate volume and serum PSA levels in the intervention group compared to the control group. This effect was independent of age and the presence or absence of diabetes. Both groups had a reduction in clinical symptoms after the end of the study. This reduction was significantly more in the intervention group and was independent of patients' age or diabetes. Therefore, vitamin D supplementation played a significant role in reducing the symptoms of BPH in patients.

Conflict of interest: The authors deny any conflict of interest in any terms or by any means during the study.

Data share statement

Data described in the manuscript will be made available upon request pending application and approval and the authors will make the data available to editors upon request either before or after publication for checking.

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Clinical Trial Registry number

The trial registered at Iranian Registry of Clinical Trials (code# IRCT20180922041089N3). URL: https://www.irct.ir/trial/35683

Contributors' Statement Page:

Dr. Abolfazl Zendehdel: conceptualized and designed the study, drafted the initial manuscript, and reviewed and revised the manuscript.

Dr. Shapour Mansoursamaei and Dr.Mohsen Ansari**:** Designed the data collection instruments, collected data, carried out the initial analyses, and reviewed and revised the manuscript.

Dr. Hossein Dialameh and Farnaz Khatami: Coordinated and supervised data collection, and critically reviewed the manuscript for important intellectual content.

References

1. National Institute of Diabetes D, Kidney Diseases. Prostate enlargement: Benign prostatic hyperplasia: National Institute of Diabetes and Digestive and Kidney Diseases, National Institutes of Health; 1998.
2. Das K, Buchholz N. Benign prostate hyperplasia and nutrition. Clinical nutrition ESPEN. 2019;33:5-11.
3. Galeone C, Pelucchi C, Talamini R, Negri E, Dal Maso L, Montella M, et al. Onion and garlic intake and the odds of benign prostatic hyperplasia. Urology. 2007;70(4):672-6.
4. Espinosa G. Nutrition and benign prostatic hyperplasia. Current opinion in urology. 2013;23(1):38-41.
5. Suzuki S, Platz EA, Kawachi I, Willett WC, Giovannucci E. Intakes of energy and macronutrients and the risk of benign prostatic hyperplasia. The American journal of clinical nutrition. 2002;75(4):689-97.
6. Kyprianou N, Tu H, Jacobs SC. Apoptotic versus proliferative activities in human benign prostatic hyperplasia. Human pathology. 1996;27(7):668-75.
7. Ficarra V, Rossanese M, Zazzara M, Giannarini G, Abbinante M, Bartoletti R, et al. The role of inflammation in lower urinary tract symptoms (LUTS) due to benign prostatic hyperplasia (BPH) and its potential impact on medical therapy. Current urology reports. 2014;15(12):463.
8. Handisurya A, Steiner GE, Stix U, Ecker RC, Pfaffeneder-Mantai S, Langer D, et al. Differential expression of interleukin-15, a pro-inflammatory cytokine and T-cell growth factor, and its receptor in human prostate. The Prostate. 2001;49(4):251-62.
9. Steiner GE, Stix U, Handisurya A, Willheim M, Haitel A, Reithmayr F, et al. Cytokine expression pattern in benign prostatic hyperplasia infiltrating T cells and impact of lymphocytic infiltration on cytokine mRNA profile in prostatic tissue. Laboratory investigation. 2003;83(8):1131-46.
10. Royuela M, De Miguel M, Ruiz A, Fraile B, Arenas M, Romo E, et al. Interferon-g and its functional receptors overexpression in benign prostatic hyperplasia and prostatic carcinoma: parallelism with c-myc and p53 expression. European cytokine network. 2000;11(1):119-27.
11. Peehl DM, Skowronski RJ, Leung GK, Wong ST, Stamey TA, Feldman D. Antiproliferative effects of 1, 25-dihydroxyvitamin D3 on primary cultures of human prostatic cells. Cancer Research. 1994;54(3):805-10.
12. Skowronski RJ, Peehl DM, Feldman D. Vitamin D and prostate cancer: 1, 25 dihydroxyvitamin D3 receptors and actions in human prostate cancer cell lines. Endocrinology. 1993;132(5):1952-60.
13. Kahokehr A, Vather R, Nixon A, Hill AG. Non-steroidal anti-inflammatory drugs for lower urinary tract symptoms in benign prostatic hyperplasia: systematic review and meta-analysis of randomized controlled trials. BJU international. 2013;111(2):304-11.
14. Adorini L, Penna G, Fibbi B, Maggi M. Vitamin D receptor agonists target static, dynamic, and inflammatory components of benign prostatic hyperplasia. Annals of the New York Academy of Sciences. 2010;1193(1):146-52.
15. Wu-Wong JR, Tian J, Goltzman D. Vitamin D analogs as therapeutic agents: a clinical study update. Current opinion in investigational drugs (London, England: 2000). 2004;5(3):320.
16. DeLuca HF. Overview of general physiologic features and functions of vitamin D. The American journal of clinical nutrition. 2004;80(6):1689S-96S.
17. Espinosa G, Esposito R, Kazzazi A, Djavan B. Vitamin D and benign prostatic hyperplasia--a review. The Canadian journal of urology. 2013;20(4):6820-5.
18. Tiwari A. Advances in the development of hormonal modulators for the treatment of benign prostatic hyperplasia. Expert Opinion on Investigational Drugs. 2007;16(9):1425-39.

19. Hanchette CL, Schwartz GG. Geographic patterns of prostate cancer mortality. Evidence for a protective effect of ultraviolet radiation. Cancer. 1992;70(12):2861-9.
20. Xu Y, Shao X, Yao Y, Xu L, Chang L, Jiang Z, et al. Positive association between circulating 25-hydroxyvitamin D levels and prostate cancer risk: new findings from an updated meta-analysis. Journal of cancer research and clinical oncology. 2014;140(9):1465-77.
21. Wilson KM, Shui IM, Mucci LA, Giovannucci E. Calcium and phosphorus intake and prostate cancer risk: a 24-y follow-up study. The American journal of clinical nutrition. 2015;101(1):173-83.
22. Schwartz GG, editor Vitamin D in health and disease: Vitamin D and the epidemiology of prostate cancer. Seminars in dialysis; 2005: Wiley Online Library.
23. Murphy AB, Nyame YA, Batai K, Kalu R, Khan A, Gogana P, et al. Does prostate volume correlate with vitamin D deficiency among men undergoing prostate biopsy? Prostate cancer and prostatic diseases. 2017;20(1):55-60.
24. Haghsheno M-A, Mellström D, Behre C-J, Damber J-E, Johansson H, Karlsson M, et al. Low 25-OH vitamin D is associated with benign prostatic hyperplasia. The Journal of urology. 2013;190(2):608-14.
25. Zhang W, Zheng X, Wang Y, Xiao H. Vitamin D deficiency as a potential marker of benign prostatic hyperplasia. Urology. 2016;97:212-8.
26. Colli E, Rigatti P, Montorsi F, Artibani W, Petta S, Mondaini N, et al. BXL628, a novel vitamin D3 analog arrests prostate growth in patients with benign prostatic hyperplasia: a randomized clinical trial. European urology. 2006;49(1):82-6.
27. Caretta N, Vigili de Kreutzenberg S, Valente U, Guarneri G, Pizzol D, Ferlin A, et al. Hypovitaminosis D is associated with lower urinary tract symptoms and benign prostate hyperplasia in type 2 diabetes. Andrology. 2015;3(6):1062-7.
28. Zendedel A, Gholami M, Anbari K, Ghanadi K, Bachari EC, Azargon A. Effects of vitamin D intake on FEV1 and COPD exacerbation: a randomized clinical trial study. Global journal of health science. 2015;7(4):243.
29. Russell RM. Vitamin and trace mineral deficiency and excess. Harrison's principles of internal medicine. 2005.
30. Wong CK-h, Choi EP-H, Chan SW-H, Tsu JH-l, Fan C-w, Chu PS-k, et al. Use of the international prostate symptom score (IPSS) in Chinese male patients with benign prostatic hyperplasia. The Aging Male. 2017;20(4):241-9.
31. Crescioli C, Maggi M, Vannelli GB, Luconi M, Salerno R, Barni T, et al. Effect of a vitamin D3 analogue on keratinocyte growth factor-induced cell proliferation in benign prostate hyperplasia. The Journal of Clinical Endocrinology & Metabolism. 2000;85(7):2576-83.
32. Morelli A, Vignozzi L, Filippi S, Vannelli GB, Ambrosini S, Mancina R, et al. BXL-628, a vitamin D receptor agonist effective in benign prostatic hyperplasia treatment, prevents RhoA activation and inhibits RhoA/Rho kinase signaling in rat and human bladder. The Prostate. 2007;67(3):234-47.
33. Taniguchi K, Katagiri K, Kashiwagi H, Harada S, Sugimoto Y, Shimizu Y, et al. A novel nonsecosteroidal VDR agonist (CH5036249) exhibits efficacy in a spontaneous benign prostatic hyperplasia beagle model. The Journal of Steroid Biochemistry and Molecular Biology. 2010;121(1-2):204-7.
34. Safwat AS, Hasanain A, Shahat A, AbdelRazek M, Orabi H, Hamid SKA, et al. Cholecalciferol for the prophylaxis against recurrent urinary tract infection among patients with benign prostatic hyperplasia: a randomized, comparative study. World journal of urology. 2019;37(7):1347-52.
35. Kristal AR, Arnold KB, Schenk JM, Neuhouser ML, Goodman P, Penson DF, et al. Dietary patterns, supplement use, and the risk of symptomatic benign prostatic hyperplasia: results from the prostate cancer prevention trial. American journal of epidemiology. 2008;167(8):925-34.
36. Penna G, Fibbi B, Amuchastegui S, Corsiero E, Laverny G, Silvestrini E, et al. The vitamin D receptor agonist elocalcitol inhibits IL-8-dependent benign prostatic hyperplasia stromal cell

proliferation and inflammatory response by targeting the RhoA/Rho kinase and NF-kB pathways. The Prostate. 2009;69(5):480-93.
37. Vaughan CP, Tangpricha V, Motahar-Ford N, Goode PS, Burgio KL, Allman RM, et al. Vitamin D and incident urinary incontinence in older adults. European journal of clinical nutrition. 2016;70(9):987-9.
38. Nunes SBR, de Matos Oliveira F, Neves AF, Araujo GR, Marangoni K, Goulart LR, et al. Association of vitamin D receptor variants with clinical parameters in prostate cancer. Springerplus. 2016;5(1):364.

Table 1: Demographic characteristics of the participants.

Variable		Vitamin D group n (%)	Control group n (%)	P value
Educational level	Under Diploma	17 (31)	22 (41)	0.008
	Diploma	22 (41)	29 (54)	
	University	15 (28)	3 (5)	
Marital status	Single	2 (4)	0	0.9
	Married	51 (94)	54 (100)	
	Divorced	1 (2)	0	
Employment status	Employed	50 (91)	51 (95)	0.9
	Unemployed/retired	4 (7)	3 (5)	

Table 2: The effect of vitamin D supplementation on prostate volume in the intervention group compared with the control group (adjusted for age and diabetes).

	Coefficient	P-value	95% CI	
			Minimum	Maximum
Age	4.63	<0.001	2.08	7.18
Diabetes	4.66	<0.001	2.10	7.22

In the control group, the mean prostate volume was significantly higher than the mean prostate volume in the intervention group.

Table 3: The effect of vitamin D supplementation on prostate-specific antigen (PSA) levels in the intervention group compared with the control group (adjusted for age and diabetes).

	Coefficient	P-value	95% CI	
			Minimum	Maximum
Age	0.47	<0.001	0.22	0.72
Diabetes	0.47	<0.001	0.22	0.72

The mean PSA level was significantly higher in the control group compared to the intervention group.

Table 4: The effect of vitamin D supplementation on International Prostate Symptom Score (IPSS) in the intervention group compared with the control group (adjusted for age and diabetes).

Variable		Time 0	Time 1 (Month 3)	Time 2 (Month 6)	P-value
Age	Intervention group score	7 ± 6	6 ± 6	5 ± 5	<0.001
	Control group score	6 ± 4	6 ± 4	5± 4	
Diabetes	Intervention	7 ± 6	6 ± 6	4 ± 5	

	group score				
	Control group score	6 ± 4	6 ± 4	5 ± 4	<0.001

The mean IPSS score was significantly higher in the control group compared to the intervention group.

Figure 1: Participants Flowchart