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Original Article

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Does Serum 25-OH Vitamin D Affect the Development of Subclinical Atherosclerosis in Patients with Primary Hypertension?

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ABSTRACT

Background: Primary hypertension (HT) has a high mortality rate due to its atherosclerotic complications. Vitamin D (VD) deficiency has been reported to increase the development of non-dipper HT (ND-HT) and atherosclerosis, but there are also conflicting results. To our knowledge, there is no study investigating the relationship between VD and carotid artery intima–media thickness (C-IMT) in patients with HT. Therefore, we aimed to investigate the relationship between VD levels, ND-HT development, and C-IMT in patients with HT with no known cardiovascular disease (CVD).

Methods: This study was conducted in 60 (63% female) patients with HT. Ambulatory blood pressure monitoring was performed using a portable digital recording device. C-IMT was measured using B-mode ultrasonography. Vitamin D levels were measured using electrochemiluminescence immunoassay. Those with C-IMT ≥0.750 mm were assumed to have subclinical atherosclerosis, and serum 25-(OH) D3 (VD) levels <20 ng/mL were VD deficient.

Results: There was a significant negative linear relationship between VD levels and triglycerides (TG) and smoking, and a significant positive linear relationship between high-density lipoprotein cholesterol levels. Triglyceride levels were significantly ($P = .015$) higher in patients who were VD deficient compared to those who were VD sufficient. Patients with subclinical atherosclerosis were older ($P = .002$) than those without. Only patient age was positively associated with C-IMT ($P < .001$). There was no relationship between VD levels/VD deficiency and the presence of ND-HT and subclinical atherosclerosis.

Conclusion: Our findings suggest that VD deficiency does not facilitate the development of ND-HT and subclinical atherosclerosis.

Keywords: Primary hypertension, vitamin D, atherosclerosis, carotid artery intima–media thickness

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INTRODUCTION

Primary hypertension (HT) is a worldwide public health problem with high morbidity and mortality. According to 2017 data of the World Health Organization, approximately 200 million strokes and myocardial infarctions and 10.5 million deaths are due to HT complications every year.[1] Target organ damage in HT occurs due to atherosclerotic vascular damage, but the factors that influence the development of atherosclerosis remain unclear. To reduce morbidity and mortality rates in patients with HT, it is necessary to identify early signs of atherosclerosis and take preventive measures to determine all factors that cause it.

Recent data suggest that, in addition to bone–mineral metabolism, VD may play a role in blood pressure (BP) regulation and the development of atherosclerosis.[2-4] Vitamin D affects many organ and tissue systems through its receptors in many different cells, such as osteoblasts, colon cells, immune cells, myocytes, cardiomyocytes, pancreatic ß-cells, neurons, and vascular endothelial cells.[5] Studies in 1alpha-hydroxylase (−/−) or

VD receptor (−/−) knockout mice have shown that renin expression and plasma angiotensin 2 concentrations are significantly increased.[5,6] Thus, the renin–angiotensin–aldosterone system (RAAS) is suppressed, which may reduce BP by inhibiting sympathetic system activity and peripheral vasodilation and decreasing kidney sodium and water retention.[7] In addition to RAAS activation in VD deficiency, it has been reported that insulin secretion disturbance, insulin resistance, and cellular proliferation/differentiation disorder may facilitate the development of atherosclerosis.[8] Although it has been investigated in a few studies, it has been suggested that VD deficiency leads to a non-dipper (ND)-HT state with RAAS activation and may ultimately facilitate the development of atherosclerosis. Non-dipper-HT is defined as nocturnal BP values not decreasing by <10% compared with daytime values and is a condition that accelerates target organ damage.[9] However, the results of studies examining the relationship between VD levels and ND-HT development in the literature are contradictory. In addition to studies showing that VD levels do not affect the development of ND-HT,[10-13] reports indicate that VD deficiency increases the risk of developing ND-HT.[4,14,15]

It is essential to determine the presence of the atherosclerosis process at the earliest stages and to take precautions. The relationship between VD and carotid artery intima–media thickness (C-IMT), an early and non-invasive indicator of the development of atherosclerosis, is an issue where evidence-based data are limited. To our knowledge, there is no published study investigating the relationship between VD and C-IMT in a homogeneous group consisting only of patients with HT. The results of studies with heterogeneous patient groups including HT patients are quite contradictory. Although some of these studies concluded that the development of atherosclerosis was easier in cases of VD deficiency,[3,16,17] other studies found no relationship between VD levels and the development of atherosclerosis[18-21] and even concluded that atherosclerosis increased as VD levels increased.[17,22]

MAIN POINTS

- Primary hypertension (HT) has a high mortality rate due to its atherosclerotic complications, but the factors that influence the development of atherosclerosis are unclear.
- Although reports suggest that low vitamin D (VD) levels may be associated with an increased risk of non-dipper HT and atherosclerosis, there are also contradictory results.
- In our literature review, we found no studies examining the relationship between VD levels and carotid artery intima–media thickness, as a surrogate marker of subclinical atherosclerosis, in a homogeneous patient group consisting only of patients with HT.
- There was no relationship between VD levels and the development of non-dipper (ND)-HT and subclinical atherosclerosis.
- Our findings also suggest that VD deficiency (<20 ng/mL) does not facilitate the development of ND-HT and atherosclerosis.

Therefore, we aimed to investigate whether there was a relationship between VD levels and 24-hour ambulatory BP monitoring (ABPM), dipper/non-dipper status, and C-IMT determined using ultrasonography (USG) in patients with HT without known CVD.

MATERIAL AND METHODS

Study Population

Sixty participants with primary HT were included in this prosp ective/cross-sectional study between July 2021 and July 2022 in Trakya University Hospital's Internal Medicine outpatient clinic. The Scientific Research Ethics Committee of Trakya University Faculty of Medicine approved this study (approval number: TUTF-BAEK 2021/343; date: July 21, 2021). Exclusion criteria included age under 18 or over 80 years, pregnancy, breastfeeding, having secondary HT, diabetes mellitus, chronic kidney disease (estimated glomerular filtration rate (eGFR) <60 mL/min 1.73 m^2), chronic liver disease, hematologic diseases, autoimmune diseases, endocrinologic diseases, malabsorption, inflammatory bowel disease, chronic infective/inflammatory diseases, malignancy history, long-term immobilization, and those who had previously been diagnosed as having HT but did not use regular drugs. Patients were additionally excluded if they had taken calcium (Ca^{+2}) or VD during the previous 3 months or were taking drugs that altered Ca^{+2} or VD levels. Each participant provided written informed consent.

Clinical and Laboratory Examinations

Demographic and clinical data were collected on the same day that 24-hour ABPM was measured. The information obtained included age, height, weight, waist circumference, duration of HT, medication use, and smoking. Body mass index (BMI) was calculated as weight divided by height squared (kg/m^2). The patient's serum levels of VD, triglycerides (TG), total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C), sodium (Na^+), potassium (K^+), Ca^{+2}, and phosphorus (P^-) were measured. Vitamin D levels were determined using electrochemiluminescence immunoassays. The sample was considered VD deficient if the serum 25 (OH) D3 level was <20 ng/mL.[23]

Ambulatory Blood Pressure Monitoring

Twenty-four-hour ABPM was done using a portable digital recording device (IEM Mobil-O-Graph NG, Germany). The cuff was placed around the patient's nondominant upper arm. Sleep and awake periods were assessed based on the information obtained from the patients. The device was set to measure BP (8 AM-12 AM) every 15 minutes during the day and every 30 minutes at night (12 AM-8 AM). The patients were instructed to keep track of their sleeping and waking hours and continue taking their daily medications while participating in physical activity. The method was considered reliable if >70% of the measurements were valid. The percentage of decrease in nighttime BP according to 24-hour mean arterial pressure (MAP) values was calculated using the formula "Nighttime BP decrease (%) =

(Daytime BP − Nighttime BP) × 100/Daytime BP." Patients were classified as dippers if the nighttime MAP declined by ≥10%. If the reduction was <10%, they were classified as ND-HT.[9]

Carotid Intima–Media Thickness Measurement

Atherosclerosis was assessed by measuring C-IMT using USG) in the Department of Radiology. Each patient underwent C-IMT measurements, B-mode USG, and duplex Doppler examinations using a linear transducer with a frequency range of 7.5-18 MHz. C-IMT was measured 1 cm proximal to the common carotid artery (CCA) bifurcation in the plaque-free area where the anterior and posterior walls could be seen. C-IMT– was calculated by measuring the distance between 2 bright lines, edge to edge. Both measurements were recorded as right and left C-IMT, and the average of the 2 measurements was recorded as C-IMT. Those with C-IMT ≥ 0.750 mm were assumed to have subclinical atherosclerosis.[24]

Statistical Analysis

Statistical analyses were performed using the Statistical Package for the Social Sciences 22.0 (IBM SPSS Corp.; Armonk, NY, USA) software program. Numeric variables are presented as mean standard deviation (SD), and categorical variables are reported as percentages and counts. For numerical variables, descriptive statistics were created. The normality of parametric data was tested using the Shapiro–Wilk test. For normally distributed continuous variables, the independent Student's *t*-test was used. When the data were not normally distributed, a nonparametric statistical test, Mann–Whitney *U*, was used to compare the mean values between the groups. Differences in categorical variables were assessed using the chi-square test. Pearson's correlation analysis was used to assess the relationship between normal distribution parameters, and Spearman's rho correlation was used for non-normal distribution parameters. The results were considered significant when *P*-values were less than .05.

RESULTS

The mean age of the patients was 52.0 ± 12 years, 63% were female (n = 38), the mean duration of HT was 93.2 ± 120 months, the mean BMI was 29.8 ± 4.8 kg/m^2, and 28.3% (n = 17) were smokers. Forty-seven (78%) patients were taking angiotensin-converting enzyme inhibitors or angiotensin receptor blockers (ACEi/ARB), 15 (25%) took calcium channel blockers (CCB), 19 (32%) took beta-blockers (BB), and 21 (35%) were taking diuretics. The mean VD levels were 17.8 ± 8.6 ng/mL, and VD deficiency was present in 40 (67%) patients. The mean systolic BP (SBP) decrease at nighttime was 8.8 ± 7.2 mm Hg, the mean diastolic BP (DBP) decrease at nighttime was 11.0 ± 8.6 mm Hg, and there were 31 patients (52%) with ND-HT. The mean C-IMT was 0.733 ± 0.189 mm, and 25 patients (41.7%) had subclinical atherosclerosis.

Demographic, clinical, and laboratory parameters of patients with or without vitamin D deficiency

Vitamin D deficiency (< 20 ng/mL) and sufficiency (≥ 20 ng/mL) were identified in 40 and 20 patients, respectively. Except for TG, all demographic, clinical, and laboratory parameters were statistically similar in the 2 groups. The VD deficient (VDD) group had significantly higher TG levels compared with the VD sufficient (VDS) group (*P* = .015).

Based on the 24-hour ABPM assessment, no statistically significant difference existed between the groups. Twenty (50%) patients in the VDD group and 11 (45%) in the VDS group were ND-HT, and the 2 groups were statistically similar. The mean C-IMT was 0.726 ± 0.185 mm in the VDD group and 0.748 ± 0.201 mm in the VDS group, and there was no statistical difference between the 2 groups. There was also no difference in the presence of subclinical atherosclerosis in the VDD and VDS groups (38% and 50%, respectively). Table 1 includes data for the VDD and VDS groups, and Figure 1 shows the nighttime SBP dipping, DBP dipping, and C-IMT values.

Association of Vitamin D Levels with Demographic, Clinical, and Laboratory Parameters

Vitamin D levels were found to have a negative linear relationship with both TG (*r* = −0.419, *P* < .001) and smoking (*r* = −0.271, *P* = .035). In contrast, a positive linear relationship was discovered between VD levels and HDL-C (*r* = 0.303, *P* = .019). However, no significant correlation was found between VD levels and other data. No significant relationship was observed between VD, C-IMT, and subclinical atherosclerosis. Table 2 displays the relationships between VD and other data at *P* < .200.

Demographic, Clinical, and Laboratory Data of Patients With or Without Non-dipper Hypertension

Twenty-nine patients (48%) were D-HT, and 31 (52%) were ND-HT in the present study.

In the dipper group, the mean age was 50.4 ± 12.6 years, the mean duration of HT was 99 ± 42 months, the mean BMI was 29.5 ± 4.9 kg/m^2, 59% of the patients were female (n = 17), and 28.3% (n = 10) were smokers. Twenty-three (79%) patients were using ACEi/ARB, 8 (23%) patients used CCB, 9 (31%) patients used BB, and 9 (31%) patients were using diuretics. The mean VD levels were 18.7 ± 9.6 ng/mL, and VD deficiency was present in 20 (69%) patients. The mean SBP decrease at night was 13.57 ± 4.05 mm Hg, and the mean DBP decrease at night was 17.53 ± 6.32 mm Hg. In this group, the mean C-IMT thickness was 0.732 ± 0.121 mm, and 13 patients (45%) had subclinical atherosclerosis.

In the non-dipper group, the mean age was 53.5 ± 11.1 years, the mean duration of HT was 97 ± 96.7 months, the mean BMI was 29.5 ± 4.9 kg/m^2, 68% of the patients were female (n = 21), and 23% (n = 7) were smokers. Twenty-four (77%) patients were using ACEi/ARB, 7 (28%) patients used CCB, 10 (32%) patients used BB, and 12 (39%) patients were using diuretics. The mean VD levels were 17.1 ± 7.7 ng/mL, and VD deficiency was present in 20 (65%) patients. The mean SBP decrease at night

Table 1. Demographic, Clinical, and Laboratory Data of Patients with Vitamin D Deficiency or Sufficiency

Data	VD < 20 ng/mL (n = 40)	VD ≥ 20 ng/mL (n = 20)	*P*	Data	VD < 20 ng/mL (*n* = 40)	VD ≥ 20 ng/mL (*n* = 20)	*P*
Female n (%)	24 (60)	14 (70)	.606	VD ± SD (ng/mL)	13.7 ± 4.6	27.2 ± 7.1	**<.001**
Age ± SD (years)	51 ± 13	54 ± 8	.309	24-h-SBP ± SD (mm Hg)	124.5 ± 14.0	126.0 ± 12.1	.572
HT duration ± (months)	86 ± 123	108 ± 114	.497	D-SBP ± SD (mm Hg)	127.0 ± 13.8	127.9 ± 12.4	.812
BMI ± (kg/m^2)	30 ± 5	28 ± 3	.415	N-SBP ± SD (mm Hg)	116.2 ± 15.5	118.4 ± 13.1	.419
W.Circum. ± (cm)	102 ± 12	98 ± 10	.350	SBP dipping (%)	8.5 ± 7.3	7.2 ± 7.0	.518
Smoking, n (%)	14 (35)	3 (15)	.188	24-h-DBP ± SD (mm Hg)	77.0 ± 9.2	79.0 ± 9.2	.425
ACEi/ARB, n (%)	31 (78)	16 (80)	.965	D-DBP ± SD (mm Hg)	79.3 ± 9.6	79.7 ± 9.5	.879
BB, n (%)	12 (30)	7 (35)	.772	N-DBP ± SD (mm Hg)	7.0 ± 9.9	72.7 ± 1.8	.345
CCB, n (%)	11 (28)	4 (20)	.752	DBP Dipping (%)	11.7 ± 9.2	9.7 ± 7.3	.390
Diuretics, n (%)	15 (38)	6 (30)	.774	24-h-MAP ± SD (mm Hg)	98.8 ± 10.5	100.5 ± 9.7	.546
TG ± (mg/dL)	182 ± 119	116 ± 50	**.015**	D-MAP ± SD (mm Hg)	101.2 ± 10.6	102.3 ± 10.0	.701
LDL-C ± (mg/dL)	133 ± 29	133 ± 42	.940	N-MAP ± SD (mm Hg)	91.0 ± 11.4	93.7 ± 11.1	.402
HDL-C ± (mg/dL)	52 ± 19	55 ± 12	.167	MAP dipping (%)	9.9 ± 7.8	8.4 ± 7.0	.464
TC ± (mg/dL)	207 ± 41	202 ± 46	.682	ND-HT n (%)	20 (50)	11 (45)	.715
Na^+ ± (mmol/L)	140 ± 2	140 ± 2	.933	24-hour MPP ± SD (mm Hg)	47.40 ± 10.35	47.05 ± 8.67	.897
Ca^{+2} ± (mg/dL)	9.0 ± 0.4	9.1±0.2	.111	Daytime MPP ± SD (mm Hg)	47.65 ± 10.12	47.30 ± 8.76	.808
P ± (mg/dL)	3.5 ± 0.5	3.6±0.5	.605	Nighttime MPP ± SD (mm Hg)	46.12 ± 11.36	45.75 ± 9.05	.748
C-IMT ± (mm)	0.726 ± 0.185	0.748 ± 0.201	.634	MPP dipping (%)	3.01 ± 9.29	3.09 ± 11.54	.995
S.Atherosc. n (%)	15 (38)	10 (50)	.355				

The value in bold indicates statistical significance.
ACEi/ARB, angiotensin-converting enzyme inhibitor or angiotensin receptor blocker; BB, beta-blocker; BMI, body mass index; Ca+2, calcium; CCB, calcium channel blocker; C-IMT, mean carotid intima–media thickness; BMI, body mass index; D, daytime; DBP, diastolic blood pressure; HDL-C, high-density lipoprotein cholesterol; HT, hypertension; LDL-C, low-density lipoprotein cholesterol; N, nighttime; TC, total cholesterol; Na+, sodium; MAP, mean arterial pressure; MPP, mean pulse pressure; P, phosphor; SBP, systolic blood pressure; DBP, diastolic blood pressure; 24-h, average daytime; TC, total cholesterol; TG, triglycerides; VD, vitamin D; W. Circum., Waist Circumference; SD, standart deviation; S.Atherosc., Subclinical Atherosclerosis.

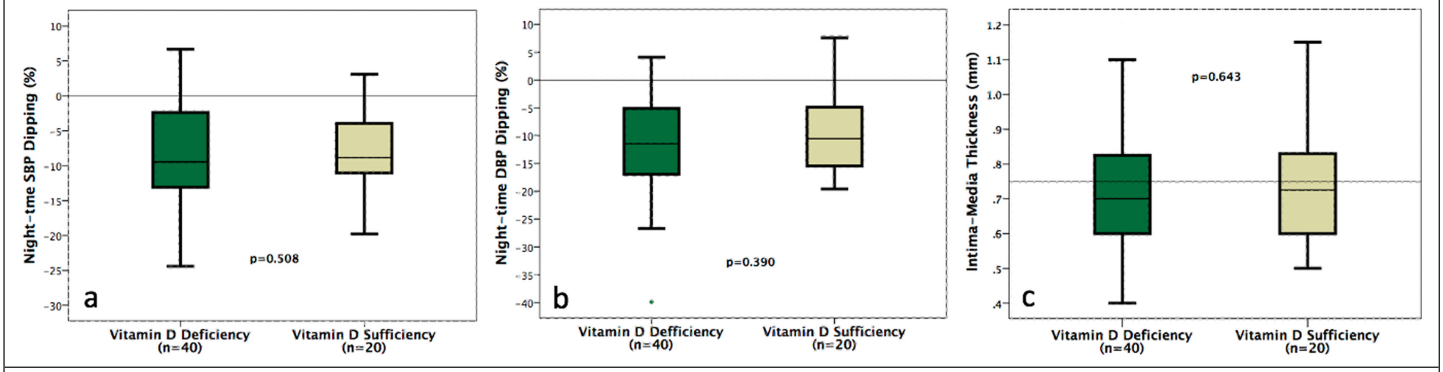

Figure 1. Nighttime systolic, nighttime diastolic blood pressure dipping and carotid artery intima–media thickness values of the hypertensive patients with vitamin D deficient or sufficient. A) nighttime systolic blood pressure dipping (in mm Hg) of the study groups, B) nighttime diastolic blood pressure dipping (in mm Hg) of the study groups, and C) carotid artery intima–media thickness (in mm) of the study groups.

was 2.86 ± 5.33 mm Hg, and the mean DBP decrease at night was 4.91 ± 5.42 mm Hg. In this group, the mean C-IMT thickness was 0.735 ± 0.175 mm and 12 (39%) patients had subclinical atherosclerosis.

No statistical differences were found between patients with D-HT and ND-HT in terms of demographic, clinical, and laboratory findings. The VD and C-IMT levels of the D-HT and ND-HT groups are shown in Figure 2.

Demographic, Clinical, and Laboratory Data of Patients With or Without Subclinical Atherosclerosis

Patients with subclinical atherosclerosis were older than patients without subclinical atherosclerosis (*P* = .02). The

Table 2. Multiple Relationships Between Vitamin D Levels and Other Data

Data	*r*	*P*
TG (mg/dL)	−0.419	**< .001**
HDL-C (mg/dL)	0.303	**.019**
Smoking	−0.272	**.035**
BMI (kg/m^2)	−0.238	.067
Age (years)	0.233	.073
Waist circumference	−0.202	.121
HT duration (months)	0.185	.158

Relationships with $P < .200$ are shown in the table. Values in bold indicate statistical significance.
BMI, body mass index; HDL-C, high-density lipoprotein cholesterol; HT, hypertension; TG, triglycerides; VD, vitamin D.

duration of HT was longer in patients with subclinical atherosclerosis than in patients without subclinical atherosclerosis, but it did not reach statistical significance ($P = .05$). Although the differences were not statistically significant, patients with subclinical atherosclerosis showed higher values of nighttime DBP (NT-DBP), 24-hour DBP (24-h DBP), 24-hour MAP (24-h MAP), and NT-MAP than those without the condition ($P < .100$ for all). Additionally, patients with subclinical atherosclerosis had lower BMI values compared with those without subclinical atherosclerosis ($P < .100$). There was no difference in VD levels between patients with and without subclinical atherosclerosis. The data of patients with or without subclinical atherosclerosis are shown in Table 3, and the VD levels of patients with or without subclinical atherosclerosis are shown in Figure 3.

Evaluation of the Determinants of the Increase in Carotid Artery Intima–Media Thickness

When the multiple correlations between C-IMT thickness and other data were analyzed, patient age was the only parameter with a statistically significant relationship with C-IMT ($r = 0.399$, $P < .001$). Although it did not reach statistical significance, a positive linear relationship was found between C-IMT thickness and N-DBP, HT duration, AD-DBP, N-MPP, AD-MAP, N-MAP, and D-MAP values ($P < .200$ for all). The data correlated with C-IMT at the $P < .200$ level and their correlation levels are shown in Table 4. The relationship between carotid-artery intima-media thickness and vitamin D values of the study group are shown in Figure 4.

DISCUSSION

The VD level was below 20 ng/mL in 67% of our patients with HT. This finding is crucial because it reveals that VD deficiency is highly prevalent in hypertensives. Data in the literature show that VD deficiency in patients with HT reaches up to 80%.[10] In our study group, we found no statistically significant relationship between VD levels or the presence of VD deficiency and any ABPM parameters or the presence of ND-HT. Studies investigating the effect of VD levels or VD deficiency on the development of ND-HT are limited in number, and results are contradictory. Similar to the results of our study, Zhang et al[11] found no difference in VD levels between those with dipper (D-HT) or ND-HT in their study group consisting of patients with HT who did or did not receive anti-hypertensive treatment. However, they reported a weak ($r = 0.135$) positive linear relationship between VD and nighttime DBP decrease. Larsen et al[12] found that after 20 weeks of treatment with 3000 IU/day VD cholecalciferol in 112 patients with HT with VD deficiency in Norway, there was a 3/1 mm Hg decrease in SBP/DBP values, but it was not statistically significant. Witham et al,[13] in a study group consisting of 159 patients with isolated systolic HT with an average age of 77 years and basal VD levels < 30 ng/mL, determined that VD treatment administered at a dose of 100 000 U every 3 months for 1 year had no significant effect on ABPM parameters, arterial stiffness, and endothelial function. McMullan et al[10] reported that 8-week VD treatment did not significantly affect ABPM data in patient groups consisting of VD-deficient and obese individuals with HT. On the other hand, there are reports in the literature that the VD deficiency facilitates ND-HT development. Gu et al[4] found that in patients with an average age of 60 years, VD deficiency was 25% more common in patients with ND-HT than in those with D-HT. Avunduk et al[14] observed no relationship

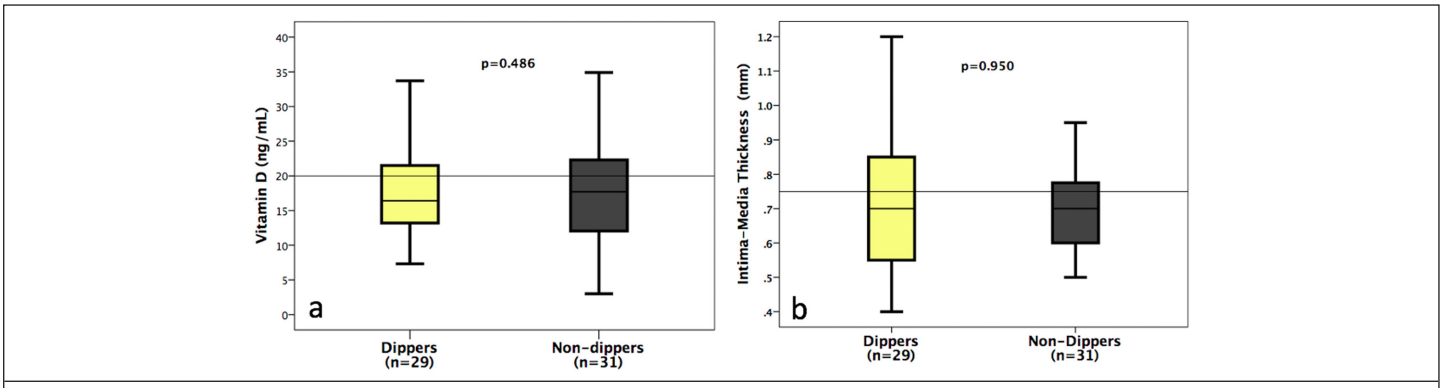

Figure 2. Vitamin D and carotid artery intima–media thickness values of the hypertensive patients with D-HT or ND-HT. A) vitamin D values (in ng/mL) of the study group and B) carotid artery intima–media thickness (in mm) of the study groups.

Table 3. Demographic, Clinical, and Laboratory Data of Patients With and Without Subclinical Atherosclerosis

Data	C-IMT < 0.750 mm (n = 35)	C-IMT ≥ 0.750 mm (n = 25)	*P*	Data	C-IMT < 0.750 mm (n = 35)	C-IMT ≥ 0.750 mm (n = 25)	*P*
Female n (%)	24 (69)	14 (56)	.469	VD ± SD (ng/mL)	16.2 ± 7.6	20.1 ± 9.6	.170
Age ± SD (years)[#]	48.2 ± 12.0	57.4 ± 9.5	**.002**	24-h SBP ± SD (mm Hg)	122.9 ± 11.3	127.8 ± 15.5	.162
HT duration ± SD (months)	74 ± 108	119 ± 133	.050	D-SBP ± SD (mm Hg)	125.5 ± 11.6	129.7 ± 15.2	.413
BMI ± SD (kg/m^2)	31 ± 5	29 ± 4	.051	N-SBP ± SD (mm Hg)	114.9 ± 11.2	119.7 ± 18.5	.397
W.Circumfer. ± SD (cm)	101 ± 13	100 ± 9	.771	SBP dipping (%)	8.2 ± 6.8	7.8 ± 7.7	.830
Smoking, n (%)	10 (%29)	7 (%28)	.997	24-h-DBP ± SD (mm Hg)	75.8 ± 9.9	80.2 ± 7.5	.064
ACEi/ARB, n (%)	28 (%80)	19 (%76)	.958	D-DBP ± SD (mm Hg)	78.1 ± 10.3	81.2 ± 8.1	.216
BB, n (%)	10 (%29)	9 (%36)	.743	N-DBP ± SD (mm Hg)	68.7 ± 10.4	73.8 ± 9.4	.055
CCB, n (%)	10 (%29)	5 (%20)	.650	DBP dipping (%)	11.9 ± 9.1	9.8 ± 8.0	.351
Diuretics, n (%)	13 (37)	8 (32)	.891	24-h-MAP ± SD (mm Hg)	97.4 ± 10.2	102.0 ± 9.7	.079
TC ± SD (mg/dL)	206 ± 41	205 ± 45	.917	D-MAP ± SD (mm Hg)	99.9 ± 10.5	103.8 ± 9.8	.142
LDL-C ± SD (mg/dL)	134 ± 33	131 ± 35	.662	N-MAP ± SD (mm Hg)	89.8 ± 10.3	94.8 ± 12.2	.094
HDL-C ± SD (mg/dL)	54 ± 17	52 ± 17	.589	MAP dipping (%)	9.8 ± 7.4	8.7 ± 7.7	.351
TG ± SD (mg/dL)	157 ± 81	164 ± 135	.549	24-h-MPP ± SD (mm Hg)	47.2 ± 6.0	47.4 ± 13.5	.254
Na^+ ± SD (mmol/L)	140 ± 2	140 ± 2	.891	D-MPP ± SD (mm Hg)	47.3 ± 6.4	47.8 ± 13.0	.322
Ca^{+2} ± SD (mg/dL)	9.2 ± 0.3	9.1 ± 0.3	.295	N-MPP ± SD (mm Hg)	46.1 ± 6.1	45.8 ± 14.9	.177
P ± SD (mg/dL)	3.6 ± 0.5	3.6 ± 0.6	.959	MPP dipping (%)	10.9 ± 9.7	4.6 ± 1.4	.296
C-IMT ± SD (mm)	0.608 ± 0.082	0.909 ± 0.152	**< .001**	ND-HT n (%)	13 (52)	16 (46)	.631

Values in bold indicate statistical significance.
ACEi/ARB, angiotensin-converting enzyme inhibitor or angiotensin receptor blocker; beta-blocker; BMI, body mass index; Ca^{+2}, calcium; CCB, calcium channel blocker; C-IMT, mean carotid intima–media thickness; D, daytime; DBP, diastolic blood pressure; HD-HTi, non-dipper hypertension; HDL-C, high-density lipoprotein cholesterol; HT, hypertension; LDL-C, low-density lipoprotein cholesterol; N, nighttime; Na+, sodium; MAP, mean arterial pressure; MPP, mean pulse pressure; P, phosphor; SBP, systolic blood pressure; 24-h, average daytime; TC, total cholesterol; TG, triglycerides; VD, vitamin D; W.Circumfer., waist circumference; SD, standart deviation.

between VD levels and ABPM parameters in patients who were followed up for HT for at least a year, all of whom were using RAAS blockers. However, they reported that VD levels were lower in patients with ND-HT. Karadag et al,[15] in their study group of 73 patients with HT and 34 healthy individuals, found that the VD levels of patients with ND-HT were lower than those with D-HT and healthy controls. However, when regression analysis was performed, it was observed that there was no relationship between VD levels and the presence of ND-HT in this study.[15] Conflicting data in the literature regarding the relationships of VD levels with BP, ABPM parameters, and ND-HT may be related to the fact that the patient groups in these studies are not comparable. Differences in the ethnicity, age, sex, comorbid conditions accompanying HT, and medications used by the patients evaluated in the studies may have affected the relationship between VD and BP, which may have led to the contradictory results we presented above.

Another aim of our study was to reveal whether there was a relationship between VD levels and the development of atherosclerosis, which is the most important cause of mortality in patients with HT. Therefore, detecting the development of atherosclerosis at the earliest stage before target organ damage occurs is essential. Although the patients included in our study had no known CVD, we found that 41% of the patients had subclinical atherosclerosis. In the present study, the ages of the patients with subclinical atherosclerosis were statistically significantly higher than those without subclinical atherosclerosis, and the HT durations, N-DBP, AD-DBP, AD-MAP, and N-MAP values were higher than in patients without subclinical atherosclerosis, at the limit of statistical significance. In the multiple relationship analysis of C-IMT and other data, only patient age was associated with C-IMT. We found no association between subclinical atherosclerosis and either VD levels or deficiency in our study population. Our study is the first to investigate the relationship between VD and C-IMT in patients with HT as a homogeneous group. When we evaluated the results of studies including patients with HT, similar to our results, Carnevale et al[18] found that there was no relationship between VD and C-IMT in their study group of 97 patients with HT and 62 with type 2 diabetes. They showed that C-IMT was positively related to patient age and negatively related to eGFR value. Reis et al[19] evaluated the relationship between VD and C-IMT in a patient group with an average age of 76 years: 65% were women, had

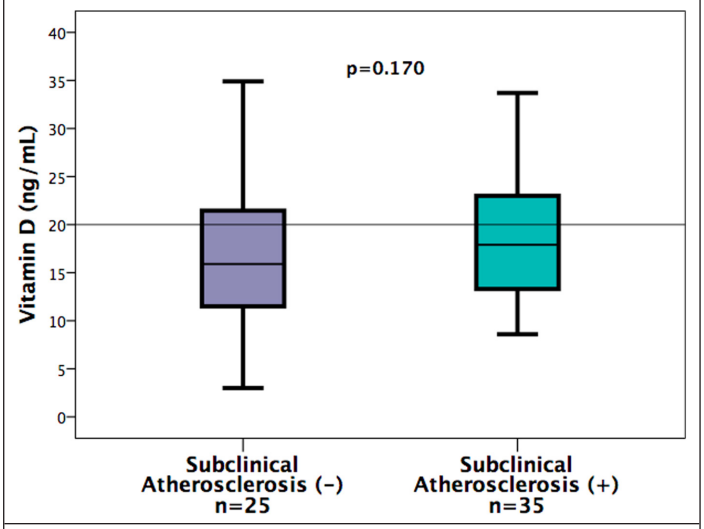

Figure 3. Vitamin D values of the hypertensive patients with no subclinical atherosclerosis or subclinical atherosclerosis.

Table 4. Relationships of Carotid Intima–Media Thickness with Other Demographic, Clinical, and Laboratory Data

Data	*r*	*P*
Age (years)	0.399	**< .001**
Nighttime DBP (mm Hg)	0.203	.120
HT duration (months)	0.198	.130
24-hours DBP (mm Hg)	0.195	.136
Nighttime MPP (mm Hg)	−0.190	.145
24-hours MAP (mm Hg)	0.185	.156
Nighttime MAP (mm Hg)	0.172	.189
Daytime MAP (mm Hg)	0.171	.192

Relationships with *P* > .200 are shown in the table. The value in bold indicates statistical significance.
DBP, diastolic blood pressure; HT, hypertension; MPP, mean pulse pressure; MAP, mean arterial pressure; SBP, systolic blood pressure.

no known CVD, 60% had HT, and 11% had diabetes mellitus. They found no relationship between CCA C-IMT and internal carotid artery IMT (ICA-IMT) values, which were determined by taking the average of the right and left carotid artery IMT values and VD levels. However, when they performed the same evaluation in the HT subgroup, they observed a negative linear relationship between ICA-IMT and VD. They interpreted this association as not being observed in any subgroup in the study but only in the HT subgroup: "Admittedly, we cannot rule out that this finding may be due to chance since we performed numerous subgroup analyses."[19] Lee et al[20] reported that there was no association between C-IMT values, which were determined by averaging the bilateral maximum right and left CCA measurements, and VD in their study of individuals aged ≥ 50 years with HT. Blondon et al[21] showed that there was no correlation between baseline VD levels and either C-IMT or ICA-IMT values in a group of patients with a mean age of 60 years; 40% were Caucasian, 25.8% were African-American, 21.3% were Hispanic, and 13.3% were Chinese, 40% had HT, 8% had DM, and 12% were smokers, and the study covered a patient recruitment period of 3 years. Another result of this study was that no correlation was shown between baseline VD levels and changes in C-IMT or ICA-IMT after a mean of 9.4 years in the same patient group. In contrast to our results, Kalkan et al[16] found a negative correlation between thoracic aortic IMT values and VD in their study with a patient recruitment period of 1 year. In a study conducted in China, a negative correlation was found between VD levels and C-IMT in women without known CVD.[17] In a meta-analysis conducted by Chen et al,[3] in which 16 434 subjects were evaluated, it was concluded that VD deficiency led to increased C-IMT, although the heterogeneity between studies (I^2 = 54%) was relatively high.

On the other hand, there are reports in the literature suggesting that there may be a positive relationship between VD levels and the development of subclinical atherosclerosis. Lu et al[17] showed that a VD deficiency group had 37% (*P* = .001) less subclinical atherosclerosis C-IMT values of ≥0.800

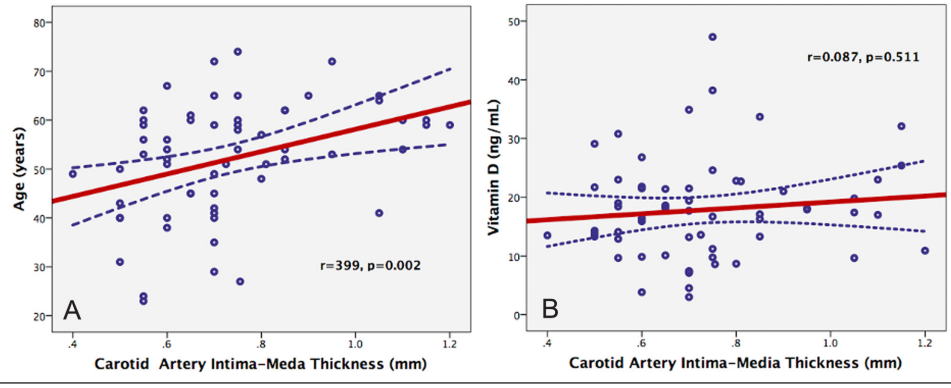

Figure 4. Relationship between carotid artery intima–media thickness and patients' age and vitamin D values. The relationship between carotid artery intima–media thickness (in mm) and patient age (in years) (A) and vitamin D values (in ng/mL) (B).

mm (by calculating the average IMT thickness of the right and left CCA) than a VD sufficient group. However, after adjusting for smoking, sex, age, DM, and BMI, this association disappeared in their study groups, 41% of whom had HT.[17] Deleskog et al[22] reported a positive linear relationship between VD levels and IMT values measured at the bifurcation, CC, ICA, and whole carotid tree (WCT) in their study group, 72% of whom had HT with high cardiovascular risk but without symptoms or signs of CVD. They demonstrated that this relationship disappeared after adjustment for current smoking, eGFR, high physical activity, latitude, and waist circumference, and only the positive relationship between VD and mean C-IMT persisted.[22] When the investigators analyzed the baseline data by dividing the patients into subgroups, they found a positive linear relationship between baseline VD levels and mean C-IMT only in patients with diabetes or statin users. In the evaluation in terms of progression 15-30 months later, when all patients were evaluated together, it was found that there was no correlation between baseline VD levels and IMT change. Interestingly, in the diabetic subgroup, a negative linear relationship was observed between baseline VD levels and the increase in mean WCT-IMT, mean C-IMT, and max C-IMT over 15-30 months, whereas a positive linear relationship was observed between baseline VD levels and the change in mean C-IMT in statin-free patients.[22]

In line with all these findings, we believe that the contradictions between the results of studies investigating the relationship between VD levels and the presence of C-IMT, or subclinical atherosclerosis, may be due to the co-morbidities, age, sex, and ethnicity differences of the patient populations. On the other hand, the IMT parameter used to evaluate the association with VD may also affect the results. Differences in the carotid artery segments were analyzed, unilateral or bilateral measurements were taken, and whether the average of the 2 sides or the result of the higher side was used as the IMT value may have affected the study results. Finally, the latitude of the region where the study was performed, the seasonal differences, the differences in the treatments administered to the patients, and whether the patients actively used VD during the study may have affected the relationship between VD and C-IMT, as well as the relationship between VD and ND-HT status, and caused different results in the studies.

The distinctive features of our study are that only patients with HT were examined, and individuals receiving VD or calcium treatment were excluded from the study. However, some limitations should be considered when evaluating our results. Although power analysis was performed during the planning of the study and the number of patients included was determined accordingly, the patient population was relatively small. Our patients live at a certain altitude and geographic region, and our study population reflects the data of patients from a single ethnicity. It should also be kept in mind that our study is an observational, cross-sectional study.

Our results show that approximately 70% of our patients with primary HT have VD deficiency, and subclinical atherosclerosis developed in 40% of patients with no known cardiovascular disease. On the other hand, our findings suggest no relationship between VD levels or VD deficiency status and the development of ND-HT, carotid artery intima–media thickening, or subclinical atherosclerosis.

Data Availability Statement: The data that support the findings of this study are available on request from the corresponding author.

Ethics Committee Approval: Ethics committee approval was received from the Scientific Research Ethics Committee of Trakya University Medical Faculty (approval number: TUTF-BAEK 2021/343; date: July 21, 2021).

Informed Consent: Written informed consent was obtained from the patients who agreed to take part in the study.

Author Contributions: Concept – A.Ü., E.B.; Design – A.Ü., E.B., D.K., N.S.; Supervision – A.Ü.; Resources – A.Ü., E.B.; Materials – A.Ü., E.B, A.A.; Data Collection and/or Processing – E.B., A.A.; Analysis and/or Interpretation – A.Ü., E.B., D.K., N.S.; Literature Search – A.Ü., E.B.; Writing Manuscript – A.Ü., E.B.; Critical Review – A.Ü., D.K.

Declaration of Interests: The authors have no conflict of interest to declare.

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