

ASSOCIATION BETWEEN 25(OH)D LEVELS AND PRIMARY ARTERIAL HYPERTENSION

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Communicated by Arvids Barševskis

This study seeks to determine whether patients from a family physician's practice have an association between 25(OH)D levels and primary arterial hypertension (AH). The study included a total of 1068 patients who were tested for vitamin D status. Data from their outpatient medical records were analysed: sex, age, body mass index, glomerular filtration rate, and the history of AH. Primary arterial hypertension was diagnosed in 63% of the patients. The mean vitamin D level in the study population was 25 ng/ml, and the largest group, or 36%, was found to have vitamin D deficiency (20–29 ng/ml). Odds ratio (OR) for hypertension was not inversely correlated with higher vitamin D levels. No statistically significant increase was observed in OR in a multi-factor analysis. The relevant hypertension ORs were 1.8 (0.4–7.5), 1.1 (0.3–4.5), 1.7 (0.4–7.2) and 0.7 (0.1–4.8) 30 ng/ml to 45 ng/ml, 20 to 29 ng/ml, 10 to 19 ng/ml, and < 10 ng/ml compared to the group of ≥ 45 ng/ml. No association was found between 25(OH)D levels and the primary arterial hypertension in study participants.

Keywords: arterial hypertension, vitamin D, family physician's practice.

INTRODUCTION

The results of numerous studies highlight the key role of vitamin D in regulating calcium and phosphorus metabolism, thus ensuring healthy bone tissue metabolism. However, the biological role of vitamin D is not limited to the regulation of bone metabolism. In recent years, there is growing evidence for the involvement of vitamin D in other processes, such as the regulation of male sexual function (Požarskis and Lejnicks, 2019), as well as in the pathogenesis of cardiovascular diseases (CVD) (Vázquez *et al.*, 1990; Moyano *et al.*, 2012; Ortega *et al.*, 2016; Požarskis and Lejnicks, 2019). CVD involve a wide range of cardiovascular diseases, which are a major cause of death and disability worldwide (Bhandari *et al.*, 2011). The main risk factors for CVD are insufficient physical activity, tobacco and alcohol abuse, psycho-emotional stress and, according to the latest data, one of the risk factors is vitamin D deficiency

(Vázquez, *et al.*, 1990; Moyano *et al.*, 2012; Ortega *et al.*, 2016; Sanz *et al.*, 2020). The vitamin D receptor was first identified in cardiovascular tissues in low-salt chromatin preparations in normal rat hearts by Walters *et al.* in 1986. In humans, vitamin D receptors were also found in ventricular cardiomyocytes. The presence of these receptors was also established in endothelial cells of human arterioles and venules. This suggests that vitamin D is involved in the regulation of the cardiovascular system (Latic and Erben, 2020). Vitamin D deficiency may influence the pathophysiology of atherosclerosis through modulation of the inflammatory response by decreasing the expression of tumour necrosis factor alpha (TNF α), interleukin-6 (IL-6), interleukin-1 (IL-1), and interleukin-8 (IL-8) in isolated blood monocytes. Suppression of IL-6 leads to decreased synthesis of the acute-phase inflammatory C-reactive protein (CRP). CRP serum concentrations are associated with atherosclerosis and serve as a predictor of cardiovascular

events. Vitamin D deficiency was shown to accelerate progression of coronary artery disease in swine by enhancing nuclear factor- κ B (NF- κ B) activation, indirectly supporting the anti-inflammatory role of vitamin D (Latic and Erben, 2020). In humans, vitamin D deficiency can be linked to vascular dysfunction, arterial stiffness, and left ventricular (LV) hypertrophy. A lack of vitamin D receptors leads to increased LV mass and increased levels of atrial natriuretic peptide along with imbalance of homeostasis, cardiac metalloproteases and fibroblasts. In turn, this promotes the formation of a fibrotic extracellular matrix and leads to LV dilation and impaired electromechanical coupling (de la Guía-Galipienso *et al.*, 2021). Type 2 diabetes mellitus (T2DM) is a risk factor for coronary heart disease. A meta-analysis designed to examine the relationship between vitamin D status or its supplementation and the incidence of T2DM showed that subjects with serum levels of the vitamin > 25 ng/ml, compared to those with levels < 14 ng/ml, had a 43% lower risk of developing type 2 diabetes and that a daily dose of vitamin D supplements above 500 IU, compared to one of < 200 IU, reduced this risk by 13%. Vitamin D deficiency has also been observed in patients with cardiac failure, myocardial infarction (MI), stroke, and peripheral arterial disease (de la Guía-Galipienso *et al.*, 2021). The most significant risk factors for arterial hypertension are: overweight, sedentary lifestyle, excessive salt intake, smoking, and excessive alcohol consumption (Mancia *et al.*, 2013). A large number of studies have been conducted to determine the role of vitamin D in the development of hypertension, and a clear association was found in most studies: low 25(OH)D levels were associated with an increased risk of arterial hypertension (AH) (Carbone *et al.*, 2014). In a cross-sectional study conducted in the United States with 12 644 participants, an inverse relationship between 25(OH)D levels and blood pressure after adjusting for age, sex, ethnicity, and physical activity was shown (Latic and Erben, 2020). Endothelial dysfunction contributes to the development of hypertension. Experimental animal studies support a role for vitamin D in regulating endothelial function. One of the proposed mechanisms of action is renin-angiotensin-aldosterone (RAAS) activation. It was reported that global vitamin D receptor knock-out mice have higher blood pressure and develop cardiac hypertrophy due to increased renin expression and subsequent activation of the RAAS. (Latic and Erben, 2020). Latvia is a country with a low number of sunshine days per year, and low average levels of vitamin D in the population. The incidence rate of arterial hypertension in Latvia is high: on average, one in every two people living in Latvia has high blood pressure. Data from the cross-sectional epidemiological study of risk factors for CVD in the Latvian population show that 45.4% of the population has arterial hypertension. The incidence increases rapidly after the age of 45, though arterial hypertension is also often observed in young people (adolescents) (Centre for Disease Prevention and Control, 2018). To date, no studies have been conducted in Latvia on the correlation of vitamin D with the risk of developing arterial hypertension. Thus, there is an interest in studying and determining the association between 25(OH) D levels and the risks of ar-

terial hypertension in a particular family physician's practice in Latvia. The working hypothesis was: a decrease in vitamin D levels increases the odds of developing arterial hypertension.

The study seeks to determine whether patients from a family physician's practice have an association between 25(OH)D levels and primary arterial hypertension.

MATERIALS AND METHODS

The type of study was a case-control study. The study was conducted from 10 October, 2018 to 10 April, 2019. The site of study was Associate Professor A. Požarskis' Family Physician's Practice, 46 Cietokšņa Street, Daugavpils, Latvia. Patient recruitment took place from 10 October, 2018 to 28 February, 2019; exposure, follow-up, and data collection were carried out from 1 March 2019 to 10 April 2019. Patients who had reached the age of 18 were voluntarily enrolled in the study. All patients over the age of 18, who had seen a physician for various health problems, were offered to determine their blood vitamin D levels. A total of 1068 patients agreed to participate in the study. None of the participants of the study took additional vitamin D. In order to determine the level of vitamin D in the blood, patients had to have a blood test at a laboratory, and the results of the blood tests were included in the study. In addition, the following data from the patients' outpatient medical records were analysed: gender, age, body mass index (BMI), glomerular filtration rate (GFR), and the history of AH. Vitamin D levels were determined at a certified laboratory by the electrochemiluminescence method using a Cobas 8000 analyser (Roche Diagnostics) and the Roche Diagnostics Vitamin D total assay. The following values are considered as reference intervals for vitamin D: optimal 25(OH)D level: 45–55 ng/ml, sufficient 25(OH)D level: > 30 ng/ml, 25(OH)D deficiency: 20–29 ng/ml, 25(OH)D deficiency (moderate): 10–19 ng/ml, 25(OH)D deficiency (severe): < 10 ng/ml, vitamin D intoxication: > 150 ng/ml (Rasa *et al.*, 2011). In conducting the study, we used the currently available hypertension classification system recommended by the European Society of Hypertension and the European Society of Cardiology (Mancia *et al.*, 2013), where the optimal blood pressure (BP) level is $< 120/80$; normal BP: 120–129/80–84; high normal BP: 130–139/85–89; stage 1 hypertension: 140–159/90–99; stage 2 hypertension: 160–179/100–109; and stage 3 hypertension: $> 180/110$. Obesity was diagnosed based on the modern World Health Organization (WHO) classification principles depending on the body mass index. Normal weight corresponds to a BMI of 18.5–24.99 kg/m², overweight — to 25–29.99 kg/m², obesity — to 30–39.99 kg/m², and severe obesity — to over 40 kg/m² (WHO, 2000; Chan *et al.*, 2012). The study group included patients with arterial hypertension, while the control group involved patients without arterial hypertension.

SPSS Statistics 20 software was used for data processing. Descriptive statistics were used to characterise the study sample: percentages in the case of non-parametric features

and means in the case of parametric variables, as well as the chi-square test and binomial test. The chi-square test, or Fisher's exact test (in cases where the number of respondents in the subgroups is small), was used to characterise the selection in the vitamin D level subgroups. The mean values of continuous variables were compared with ANOVA, after which the compliance of the data with a normal distribution was checked using the Kolmogorov–Smirnov test. One-factor and multi-factor binary logistic regression was used to examine the association of independent signs with arterial hypertension. The results were considered statistically reliable if the level of significance (p) was less than 0.05.

RESULTS

The study proposed to determine vitamin D levels in the blood in 2020 patients (all patients over the age of 18 who saw the family physician between 10 October 2018 and 28 February 2019). A total of 1068 patients agreed to take part. All patients who agreed to take part were examined for compliance with the study inclusion criteria; all were enrolled in the study, all were monitored, and the data of all 1068 patients were also analysed. Statistically significantly more women participated in the study: 797, or 74.6%, while the number of men was much smaller: 271, or 25.4% ($p < 0.001$, binomial test, Fig. 1). The mean age of the patients in the study sample was 54.1 years.

Of 1068 patients enrolled in the study, 673, or 63%, had primary arterial hypertension, which was statistically significantly higher than the number of patients without this diagnosis: 395, or 37% ($p < 0.001$, binomial test, Fig. 2). The mean vitamin 25(OH)D level in the study sample was 25 ng/ml. A detailed distribution of the study sample by 25(OH)D level is shown in Figure 3.

406 patients, or 38% were overweight. Normal body weight and obesity were found in the same number of patients: 320 people, or 30%. Only 22 patients, or 2.1%, were underweight. Distribution of the study sample by body mass index is shown in Figure 4.

Statistically significantly more patients with GFR > 60 ml/min were enrolled in the study, amounting to 1020 sub-

jects, or 95.5%, and 48 patients, or 4.5%, had reduced GFR < 60 ml/min ($p < 0.001$, binomial test).

The demographics of the study sample stratified by vitamin D levels are shown in Table 1. No statistically significant difference was found between the data and the different vitamin D levels. Relatively identical parameters were found within each level of vitamin D studied.

It was found that the prevalence of arterial hypertension among the patients in the study sample did not increase with each of the lowest vitamin D levels, as shown in Figure 5.

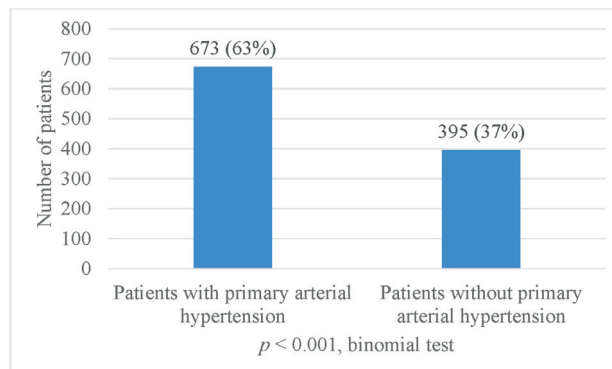


Fig. 2. Distribution of the study sample by presence of an arterial hypertension.

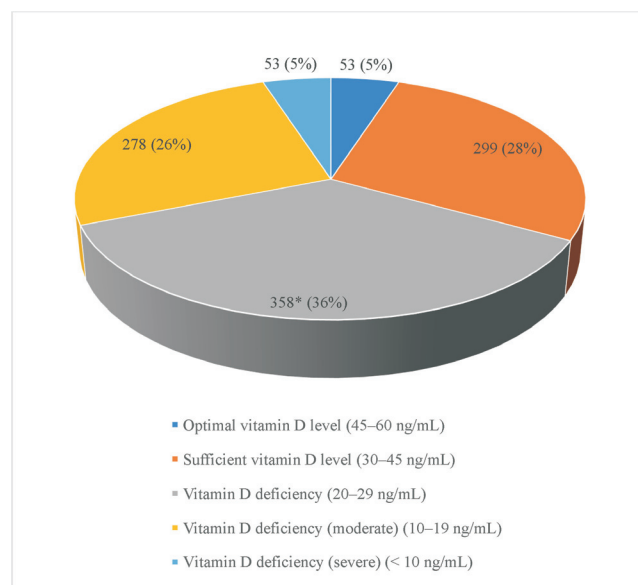


Fig. 3. Distribution of the study sample by 25(OH)D level [N (%)].

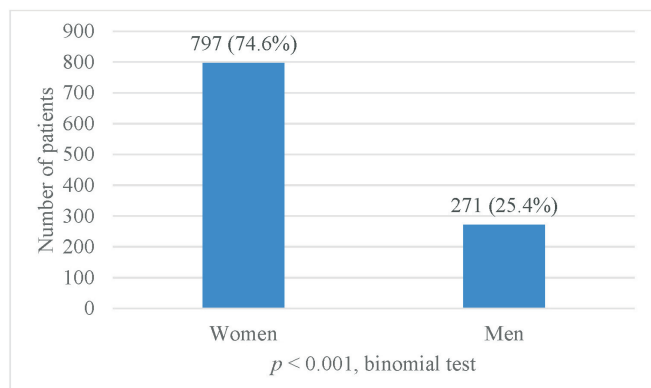


Fig. 1. Distribution of the study sample by sex.

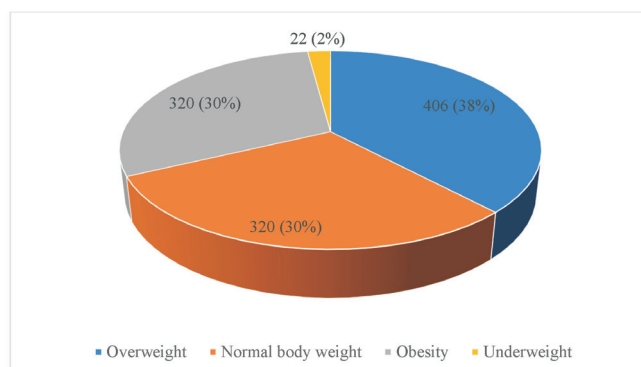


Fig. 4. Distribution of the study sample by body mass index.

Table 1. Study sample demographics stratified by 25 (OH) D levels (%)

	Optimal vitamin D level	Sufficient vitamin D level	Vitamin D deficiency	Vitamin D deficiency (moderate)	Vitamin D deficiency (severe)	<i>p</i>
Age (mean value)	54.2	55.3	52.4	54.7	56.9	0.78
Sex						
Women (%)	92.9	71.6	78.4	66.7	85.7	0.15
Men (%)	7.1	28.4	21.6	33.3	14.3	
BMI						
Underweight (%)	7.1	5.4	0	1.4	0	0.07
Normal body weight (%)	21.4	27.0	34.0	31.9	21.4	
Overweight (%)	57.1	47.3	32.0	29.0	42.9	
Obesity (%)	14.3	20.3	34.0	37.7	35.7	
Renal filtration						
GFR > 60 ml/min (%)	92.9	97.3	96.9	92.8	92.9	0.42
GFR < 60 ml/min (%)	7.1	2.7	3.1	7.2	7.1	
Primary arterial hypertension						
Yes (%)	57.1	66.2	58.8	68.1	64.3	0.73
No (%)	42.9	33.8	41.2	31.9	35.7	

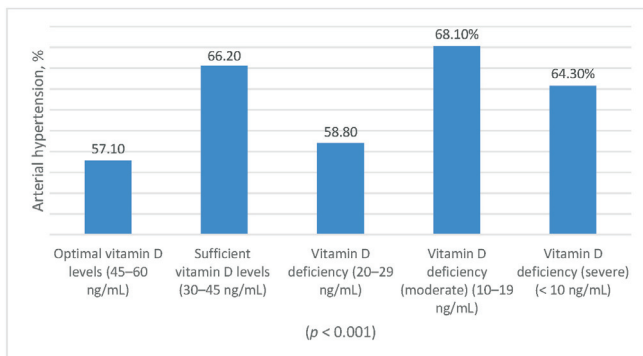


Fig. 5. Vitamin D levels and arterial hypertension.

The lowest number of patients with registered arterial hypertension was in the group with optimal vitamin D levels, i.e. 57.1%, followed by the group of patients with vitamin D deficiency (8.8%) and the patients with severe vitamin D deficiency (64.3%). Only then followed the group of patients with sufficient levels of vitamin D (66.2%) and, finally, the patients with moderate vitamin D deficiency, i.e. 68.1% ($p < 0.001$).

The results of the unadjusted and adjusted logistic regression analysis are listed in Table 2.

Odds ratio (OR) for hypertension was not inversely correlated with higher vitamin D levels. No statistically signifi-

Table 2. Single and multiple factor analysis of the relationship between vitamin D levels and other factors

	mOR	95% CI	<i>p</i>	cOR	95% CI	<i>p</i>
Age						
0-59	1			1		0.001
60+	11.7	5.7–24.1	< 0.001	10.9	4.7–25.0	
Sex						
Women	1			1		0.42
Men	1.2	0.7–2.1	0.59	1.3	0.7–2.8	
BMI						
Underweight	1			1		
Normal body weight	2.9	0.3–26.4	0.34	3.4	0.2–49.8	0.37
Overweight	9.7	1.1–86.4	0.04	12.8	0.9–186.4	0.06
Obesity	45.6	4.7–440.6	0.001	49.3	3.1–783.7	0.006
Renal filtration						
GFR 60 ml/min	1			1		0.56
GFR < 60 ml/min	6.7	0.9–52.8	0.07	2.1	0.2–27.9	
25(OH)D level						
Optimal vitamin D levels	1			1		
Sufficient vitamin D levels	1.5	0.5–4.7	0.52	1.8	0.4–7.5	0.41
Vitamin D deficiency	1.1	0.3–3.3	0.91	1.1	0.3–4.5	0.88
Vitamin D deficiency (moderate)	1.6	0.5–5.2	0.43	1.7	0.4–7.2	0.48
Vitamin D deficiency (severe)	1.4	0.3–6.2	0.70	0.7	0.1–4.8	0.48

mOR, marginal odds ratio; cOR, conditional odds ratio; CI, confidence interval; *p*, level of significance

cant increase was observed in OR in a multi-factor analysis adjusting for age, gender, BMI, and renal filtration covariates. The relevant hypertension ORs were 1.8 (0.4–7.5), 1.1 (0.3–4.5), 1.7 (0.4–7.2) and 0.7 (0.1–4.8) 30 ng/ml to 45 ng/ml, 20 to 29 ng/ml, 10 to 19 ng/ml, and < 10 ng/ml compared to the group of ≥ 45 ng/ml. Age and obesity were independent risk factors of elevated OR for arterial hypertension. In patients with reduced renal filtration, OR for arterial hypertension was 2.1 (0.2 to 27.9) compared to patients with GFR > 60 ml/min, which is not statistically significant.

DISCUSSION

A large number of studies have been conducted to determine the role of vitamin D status in the pathogenesis of hypertension. Carbone, with his working group (Carbone *et al.*, 2014), carried out a meta-analysis of 32 studies published between 2007 and 2013. These studies were focused on the relationship between 25(OH)D levels and the risk of developing AH. Most of these studies involved adults of different ages, who were divided into two groups: healthy participants and patients with AH. The total number of subjects was 145 486 (from 219 to 34 874 per study). It should be noted that most of the studies included in the meta-analysis, namely, 25 of 32, showed a clear correlation: low 25(OH)D values were associated with an increased risk of developing hypertension. However, in the other seven studies, this correlation was not found, like in our study. Similar to our study, it was not possible to determine the effect of 25(OH)D concentration on AH in two studies conducted in China and one study each in the United States, the Netherlands, Denmark, France, and Puerto Rico (Li *et al.*, 2002; Li *et al.*, 2004; Bhandari *et al.*, 2011; Caro *et al.*, 2012; Li *et al.*, 2012; Mateus-Hamdan *et al.*, 2013; Li, 2014). The population of our study consisted mostly of women, amounting to three quarters, and men were only one quarter, which could be explained by the fact that in this country, women take better care of their health, and see physicians more often; similar results in terms of sex are reflected in comparable studies in Puerto Rico and France (Caro *et al.*, 2012; Mateus-Hamdan *et al.*, 2013). The average age of our participants was 54.1 years. The other studies also involved patients over the age of 18, and the mean age of the patients was close to that of our study, such as the mean age of 41.5 years in the study group in Puerto Rico, 47 years in the study group in China, and 58.5 years in the study group in the United States (Bhandari *et al.*, 2011; Caro *et al.*, 2012; Li *et al.*, 2012). However, there were some studies that had enrolled comparatively much older patients, for example, Chan and his research team involved patients over 65 years of age (Chan *et al.*, 2012).

Of the patients enrolled in our study, 673 participants, or 63%, had documented hypertension, which is a relatively high rate compared to other studies, such as 34% of participants in China, 37% in France, 24% in the United States, and only 4% in Puerto Rico (Bhandari *et al.*, 2011; Caro *et al.*, 2012; Li *et al.*, 2012; Mateus-Hamdan *et al.*, 2013).

This may be explained by the fact that in our study, mostly patients who had visited a physician due to the pre-existing health conditions were enrolled, and arterial hypertension is one of the most common reasons for making an appointment with a family physician in our country. Unlike our study, in which a link between vitamin D status and blood pressure has not been found, there are studies that have found this association (Forman *et al.*, 2007; Scragg *et al.*, 2007). The results of the NHANES III study involving 12 644 people aged 20 years and older showed that systolic blood pressure was 3.0 mm Hg lower ($p < 0.001$) and diastolic blood pressure was 1.6 mm Hg lower ($p < 0.05$) in the higher vitamin D level group (25(OH)D ≥ 85.7 nmol/l or 34 ng/ml) compared to those with lower vitamin D status (25(OH)D ≤ 40.4 nmol/l or 16 ng/ml) adjusting for age, sex, ethnicity, and physical activity (Scragg *et al.*, 2007). The results of a prospective study conducted in the United States involving 51 529 men and 121 700 women showed that patients with vitamin D deficiency were 3.2-fold more likely to have hypertension than people with optimal vitamin D levels (Forman *et al.*, 2007). In another study in the United States, 2722 subjects aged 18 and over, 24% of whom had AH, were enrolled. All participants of the research study were divided into four groups according to vitamin D levels: 25(OH)D ≥ 40 ng/ml (Group 1), 30–39 ng/ml (Group 2), 15–29 ng/ml (Group 3), and < 15 ng/ml (Group 4). It was found that the decrease in 25(OH)D level increased the risk of developing hypertension ($p < 0.001$). In the first group, the disease was registered in only 20% of individuals with hypertension, and in the second, third and fourth groups — 27% (OR = 1.3), 41% (OR = 2.0), and 52% (OR = 2.7), respectively. Influencing factors, such as age, gender, race, and renal insufficiency were excluded from the statistical analysis (Bhandari *et al.*, 2011). However, similarly to our results, there are other studies that have not established the correlation between vitamin D levels and arterial hypertension. A study in the United States involving 559 women aged 24 to 44 found no link between vitamin D status and current blood pressure (Griffin *et al.*, 2011). A study in the Netherlands involving 1,205 participants aged 65 years and over also showed that blood pressure was not inversely correlated with vitamin D levels (Snijder *et al.*, 2007). Two studies conducted in China, and one study each in Denmark, France, and Puerto Rico, did not find this link as well (Caro *et al.*, 2012; Chan *et al.*, 2012; Li *et al.*, 2012; Skaaby *et al.*, 2012; Mateus-Hamdan *et al.*, 2013). The lack of association between blood pressure and vitamin D status in these studies may be related to the small study group and study design.

We have found that the chances of developing hypertension increase with age and higher BMI. Several worldwide studies and guidelines for the treatment of arterial hypertension have noted that age and high BMI are independent risk factors for hypertension (Hamano *et al.*, 2017; Mancia *et al.*, 2018).

In our study, there are several limitations, including the study design, which was case-control; for this reason, the

design could affect the outcome. As the data were collected from the patients at only one family physician's practice, the sample population cannot be considered representative for the whole Latvia. In the study, the effects of factors such as physical activity, smoking, menopause (in women), or intake of important nutrients (such as calcium, and magnesium) were not addressed. These factors should also be taken into account in future studies.

CONCLUSIONS

No association was found between 25(OH)D levels and arterial hypertension in study participants.

ETHICS

A permission to conduct this study was obtained from the Research Ethics Committee of the Institute of Experimental and Clinical Medicine, University of Latvia.

CONFLICT OF INTERESTS

The authors declare that there is no conflict of interests regarding the publication of this paper.

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Received 1 February 2022

Accepted in the final form 4 November 2022

SAISTĪBA STARP 25(OH)D LĪMENI UN ARTERIĀLĀS HIPERTENSIJAS ATTĪSTĪBAS RISKU

Pēdējos gados pieaug pierādījumu apjoms par D vitamīna lomu sirds un asinsvadu slimību patoģenēzē. Darba mērķis bija noteikt, vai ģimenes ārsta prakses pacientiem ir saistība starp 25(OH)D līmeņiem un arteriālās hipertensijas attīstības riskiem. Pētījumā iekļauti 1068 ģimenes ārsta prakses pacienti, kuriem bija noteikts D vitamīna līmenis asinīs. Papildus analizēti dati no pacienta ambulatorās kartes: dzimums, vecums, ķermeņa masas indekss (ĶMI), glomerulu filtrācijas ātrums, AH anamnēzē. Dati apstrādāti, izmantojot programmas *MS Excel* un *SPSS 20*. Vidējais pacientu vecums bija 54,1gads. 74,6% no respondentiem ir sievietes. Primāra arteriāla hipertensija konstatēta 63% pacientu. Vidējais D vitamīna līmenis pētījuma populācijā ir 25 ng/ml, lielākajai pacientu grupai jeb 36% ir konstatēta D vitamīna nepietiekamība (20–29 ng/ml), kas ir statistiski ticami atšķirīgi no pārējām grupām. OR (*odds ratio* – izredžu koeficients) hipertensijas gadījumā nebija apgriezti saistīts ar pieaugošajiem D vitamīna līmeņiem. Daudzfaktoru analizē, pielāgojot vecuma, dzimuma, ĶMI un nieru filtrācijas kovariantus, OR arī nebija statistiski nozīmīga pieauguma. Attiecīgie hipertensijas OR bija 1,8 (0,4–7,5), 1,1 (0,3–4,5), 1,7 (0,4–7,2) un 0,7 (0,1–4,8) 30 ng/ml līdz 45 ng/ml, 20–29 ng/ml, 10–19 ng/ml, un < 10 ng/ml salīdzinājumā ar grupu ≥ 45 ng/ml. Pētījumā netika konstatēta saistība starp D vitamīna līmeni un arteriālās hipertensijas attīstības risku.