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Original Scientific Paper

SERUM PHOSPHORUS AS A RISK FACTOR FOR CARDIOVASCULAR MORBIDITY IN TYPE 2 DIABETES MELLITUS PATIENTS

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SUMMARY – Phosphorus plays an important role in many physiological functions. Numerous studies have linked phosphorus concentration to cardiovascular (CV) disease and mortality rates. In this study, we investigated the association between high-normal serum phosphorus levels and CV morbidity in patients with type 2 diabetes. Two groups of patients with type 2 diabetes were studied, comprising both males and females aged 18 years and older. One group (140 subjects) had no CV events, while the other group (140 subjects) had one or more CV events. In both groups, we collected fasting laboratory values, medical history, and examination findings. Serum phosphorus levels were statistically significantly higher in patients with CV events and had statistically significant but relatively modest discriminatory properties (AUC 0.591; 95% confidence interval (CI) 0.531–0.649; p=0.008). A univariate logistic regression model of serum phosphorus as a continuous variable revealed a statistically significant association with the presence of a cardiovascular event (odds ratio (OR) 7.1849, 95% CI 1.7728 - 29.1190; p=0.0057). For each 1 mmol/L increase in serum phosphorus, the probability of a CV event increased 7.4-fold. After a multivariable logistic regression model, serum phosphorus was associated with a CV event (p=0.0016, OR 16.6), regardless of age, sex, total cholesterol, and systolic blood pressure.

Keywords: *Serum phosphorus; Hyperphosphatemia; Cardiovascular morbidity; Type 2 diabetes mellitus*

Introduction

Phosphorus (P) has an important role in many physiological functions and basic processes, including bone growth and development, cell signaling, mitochondrial energy production and transmission, phospholipid membrane structure and function, acid-base status regulation, and platelet aggregation[1]. The kidneys almost entirely control the excretion of phosphorus, which is filtered in free form in the glomeruli and mainly reabsorbed in the proximal tubules. In addition to calcitriol and parathyroid hormone (PTH), fibroblast growth factor 23 (FGF-23) plays a major

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role in regulating elevated serum phosphorus concentrations. The association of cardiovascular disease and mortality with phosphorus concentration was first observed in patients with chronic renal disease undergoing hemodialysis[2-4]. Studies in animal uremia models have indicated a causal role of phosphorus from food products in vascular calcification, as well as in reducing calcification of the intima and vascular media layer after phosphorus binder therapy[5,6]. Kidney regulation of phosphorus concentration is altered in diabetes mellitus because elevated blood glucose concentrations depolarize the luminal side of the cell membrane, resulting in reduced phosphorus reabsorption, intracellular phosphorus deficiency, and hyperphosphaturia[7]. In the early stages of diabetes, a paradoxical metabolic imbalance of inorganic phosphorus occurs, which can cause a decrease in the concentration of high-energy phosphorus, resulting in tissue hypoxia[8]. These changes occur in cells and tissues in which glucose entry into cells is not affected by insulin (endothelial cells), especially in patients with poorly regulated type 2 diabetes[9]. According to some previous studies, serum phosphorus concentration, but not serum calcium concentration or the calcium-phosphorus product, is associated with cardiovascular mortality in patients with type 2 diabetes[10].

This study aimed to investigate whether high-normal serum phosphorus concentrations are associated with cardiovascular events in patients with type 2 diabetes mellitus.

Patients and Methods

Patients

This observational study examined 280 subjects with type 2 diabetes mellitus, both male and female, aged 18 years and older, with a diabetes history of 5 years and longer, on oral hypoglycemic agents and/or insulin therapy. There were 140 type 2 diabetes subjects who were hospitalized for a cardiovascular (CV) event at the Cardiology Clinic, University Hospital Center Sestre milosrdnice (Zagreb, Croatia). The control group consisted of 140 type 2 diabetes mellitus subjects with no cardiovascular event (according to medical history and examination findings) but at increased risk of a cardiovascular event who were treated at the Clinic for Internal Medicine, Department for Endocrinology, Diabetes and Metabolic Diseases "Mladen Sekso", Diabetes Day Clinic, University Hospital Center Sestre milosrdnice. The exclusion criteria were eGFR <45 mL/min/1.73 m^2, kidney transplantation, rhabdomyolysis, phosphorus-binding therapy, active vitamin D, artificial heart valve, active malignancy, active infection, liver cirrhosis, and albuminuria. The study protocol was approved by the Ethics Committee of the University Hospital Center Sestre milosrdnice and the Faculty of Medicine Ethics Committee at the University of Zagreb. Signed informed consent was obtained from each patient.

Methods

Patients in both groups underwent a history and physical examination, fasting laboratory variables (complete blood count, serum calcium, serum phosphorus, blood urea nitrogen, serum creatinine, urate, albumin, C-reactive protein (CRP), hemoglobin A1c (HbA1c), total cholesterol, high-density lipoprotein (HDL), and low-density lipoprotein (LDL), and triglycerides), and analysis of a single urine sample (chemical urine analysis, urine sediment). Calcium values were corrected in subjects with hypoalbuminemia. Glomerular filtration was assessed using the Modification of Diet in Renal Disease (MDRD) and Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equations. Normal phosphorus values in the University Hospital Center Sestre milosrdnice Biochemical Laboratory are in the range of 0.79-1.42 mmol/L. Blood pressure was calculated as the average of two measurements, and hypertension was defined as systolic blood pressure (SBP) ≥ 140 mmHg, diastolic blood pressure (DBP) ≥ 90 mmHg, or the use of antihypertensive medication. Type 2 diabetes mellitus was defined as the use of insulin and/or oral hypoglycemic medications. The body mass index (BMI) was calculated as weight (in kilograms) divided by height (in meters) squared. Cardiovascular events were defined as fatal or non-fatal heart attacks, angina pectoris (stable and unstable), cardiac arrhythmias, ischemic cardiomyopathy, heart failure, and peripheral vascular disease. The association between serum phosphorus and CV events was investigated for phosphorus levels as a continuous variable and for stratified serum phosphorus concentrations in three categories: (s) P ≥1.3 mmol/L, (s) P 0.9-1.2 mmol/L, and (s) P ≤0.8 mmol/L. The risk for cardiovascular

disease was observed between these three categories. All biochemical and hematological parameters performed during hospitalization were determined at the Clinical Department of Chemistry. The laboratory analyses employed standard recommended biochemical methods, either automated or semi-automated, and were carried out according to the manufacturer's instructions, adhering to the principles of good laboratory practice.

Statistical analysis

The normality of the distribution of numerical variables was tested using the Shapiro-Wilk test. Normally distributed numerical variables were expressed as arithmetic mean ± standard deviation (SD), and non-normally distributed numerical variables were presented as median and interquartile range (IQR). Categorical variables were expressed as ratios and percentages. Normally distributed numerical variables were compared between groups using the t-test for independent samples (for two groups) or one-way analysis of variance (ANOVA; for three groups). Non-normally distributed variables were compared between groups using the Mann–Whitney U test (for two groups) or the Kruskal-Wallis ANOVA (for three groups). Categorical variables were compared between the groups using the χ^2 test. The upward or downward trend was tested using the Jonckheere-Terpstra trend test for numerical variables and the χ^2 trend test for categorical variables. The discriminant properties for distinguishing patients with and without cardiovascular events were analyzed using the Receiver Operating Characteristic (ROC) curve analysis. Logistic regression was used for multivariate analysis to examine the association between serum phosphorus and the presence of CV events. In most models, individual variables were tested simultaneously, and in the final model, variables were selected using the backward method (with the criterion for inclusion $p<0.05$ and exclusion $p>0.1$). The level of statistical significance was set at $p<0.05$. Bonferroni correction was used for multiple comparisons (for three groups). All analyses were performed using MedCalc Statistical Software version 19.1. (MedCalc Software BV, Ostend, Belgium).

Results

The mean value of serum phosphorus was 1.1±0.2 mmol/L and was statistically significantly different between subjects with (1.2±0.2 mmol/L) and without a cardiovascular event (1.1±0.2 mmol/L); p=0.005; Figure 1.

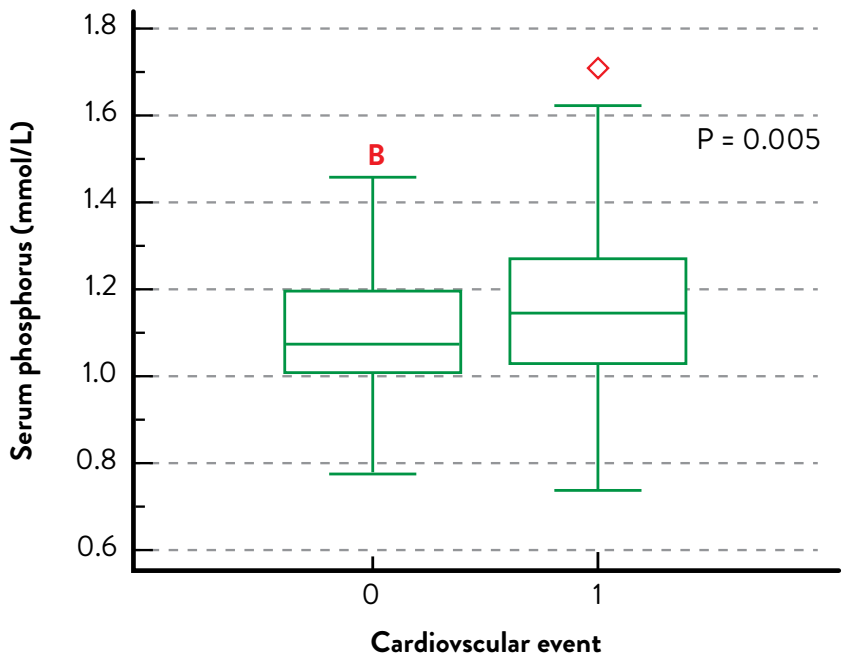

Fig 1. Serum phosphorus level in subjects without (0) and with (1) a cardiovascular event.

There were a total of 140 subjects with a CV event, of which acute coronary syndrome in 56 (40%), peripheral arterial disease in 21 (15%), cardiac decompensation in 5 (3.6%), rapid form of atrial fibrillation in 2 (1.4%), and stable angina pectoris in 56 (40%) subjects. The mean diabetes duration in the group with a CV event was 13.4±5.9 years, compared to 9.5±4.7 years in the group without a CV event. Another difference between the groups was the use of medication. A total of 61% of patients with a CV event were on statin therapy and 77% on ACE-I/ARB therapy compared to the non-CV event group, in which 42% of patients were on statin therapy and 58% on ACE-I/ARB agents. The characteristics of the subjects in relation to cardiovascular events are shown in Table 1.

Analysis of serum phosphorus concentrations in subjects categorized by cardiovascular events showed

Table 1. Baseline characteristics by cardiovascular event

Characteristic	**Total n=280**	**Non-CV event n=140**	**CV event n=140**	**p**
Age (years)	63.3±9.6	60.3±9.6	66.3±8.7	<0.001*
Male Female	162/280 (57.9%) 118/280 (42.1%)	69/140 (49.3%) 71/140 (50.7%)	93/140 (66.4%) 47/140 (33.6%)	0.004*
SBP (mmHg)	140 IQR (130-150)	140 IQR (130-154)	135 IQR (125-145)	0.006*
DBP (mmHg)	80 IQR (77-90)	84.5 IQR (80-90)	80 IQR (73-90)	<0.001*
Smoker Non-smoker	132/280 (47.1%) 148/280 (52.9%)	69/140 (49.3%) 71/140 (50.7%)	63/140 (45%) 77/140 (55%)	0.473
BMI (kg/m^2)	29.4 IQR (26-32.9)	30 IQR (27-33.7)	29.1 IQR (25.7-31.3)	0.009*
Insulin use (a) OHAs use (b) a + b	16/280 (5.7%) 213/280 (76.1%) 51/280 (18.2%)	6/140 (4.3%) 100/140 (71.4%) 34/140 (24.3%)	10/140 (7.1%) 113/140 (80.7%) 17/140 (12.1%)	0.024*
Hemoglobin (g/L)	141.4±13.8	143.7±14.1	139.1±13.2	0.005*
HbA1C (%)	7.2 IQR (6.3-8.4)	7.5 IQR (6.3-9)	7.1 IQR (6.3-7.8)	0.005*
MDRD eGFR**	79 IQR (67-89)	83 IQR (72-93)	77.5 IQR (64.8-85)	0.001*
CKD EPI eGFR	80 IQR (67-90)	85 IQR (72.5-93)	76 IQR (64-84.3)	<0.001*
Uric acid (mmol/L)	337 IQR (269.5-394)	311.5 IQR (257-364.3)	358.5 IQR (305.8-430.5)	<0.001*
Total cholesterol (mmol/L)	4.8 IQR (4-5.8)	5.1 IQR (4.4-6.1)	4.3 IQR (3.6-5.2)	<0.001*
HDL (mmol/L)	1.1 IQR (0.9-1.3)	1.1 IQR (0.9-1.3)	1 IQR (0.9-1.2)	0.187
LDL (mmol/L)	2.9 IQR (2.1-3.7)	3.2 IQR (2.5-3.9)	2.5 IQR (1.8-3.4)	<0.001*
TRG (mmol/L)	1.7 IQR (1.2-2.3)	1.8 IQR (1.3-2.3)	1.5 IQR (1.1-2.2)	0.081
Ca (mmol/L)	2.4 IQR (2.4-2.5)	2.4 IQR (2.4-2.5)	2.4 IQR (2.4-2.5)	0.781
P (mmol/L)	1.1±0.2	1.1±0.2	1.2±0.2	0.005*
Ca x P (mmol^2/l^2)	2.7±0.5	2.7±0.4	2.8±0.5	0.029*
CRP (mg/L)	1.8 IQR (1.1-3.2)	2.1 IQR (1.3-3.6)	1.6 IQR (1-2.8)	0.006*

*statistically significant at p<0.05; **mL/min/1.73 m^2; CV = cardiovascular; SBP = systolic blood pressure; DBP = diastolic blood pressure; BMI = body mass index; OHAs = oral hypoglycemic agents; HbA1c = hemoglobin A1c; MDRD = Modification of Diet in Renal Disease; CKD-EPI = Chronic Kidney Disease Epidemiology Collaboration; eGFR = estimated glomerular filtration rate; HDL = high-density lipoprotein; LDL = low-density lipoprotein; TRG = triglycerides; Ca = calcium; P = phosphorus; Ca x P = calcium – phosphorus product; CRP = C-reactive protein

no statistically significant differences in serum phosphorus levels between the individual categories of cardiovascular events (p=0.606). Serum phosphorus showed statistically significant but relatively weak discriminant properties for distinguishing subjects with and without CV events (AUC 0.591; 95% CI 0.531-0.649; p=0.008). The optimal cut-off value for discrimination of subjects with CV events was >1.24 mmol/L, with a sensitivity of 24.3% and a specificity of 82.9% (Figure 2).

Analyzing serum phosphorus concentrations by categories (category 1 (s) P ≤ 0.8 mmol/L, category 2 (s) P 0.9-1.2 mmol/L, category 3 (s) P ≥ 1.3 mmol/L), seven subjects had (s) P ≤ 0-8 mmol/L, 226 subjects had (s) P 0.9 - 1.2 mmol/L, and 47 subjects had (s) P ≥1.3 mmol/L. In category 1, three subjects were in the group with a cardiovascular event (42.86%), and four subjects were in the group without a cardiovascular event (57.1%). In category 2, 105 subjects had a cardiovascular event (46.46%), while 121 had no cardiovascular event (53.53%). In category 3, of the 47 subjects, 32 (68.09%) were in the group with a cardiovascular event, while the remaining 15 (31.91%) were in the group without a cardiovascular event. Higher serum phosphorus levels, (s) p ≥1.3 mmol/L, were statistically significantly associated with cardiovascular events (p=0.024), female gender (p=0.007), lower hemoglobin (p=0.007), a lower estimated glomerular filtration rate (eGFR) according to the MDRD equation (p=0.042) and CKD-EPI equations (p=0.030), lower albumin (p=0.028) and higher calcium–phosphorus (Ca x P) product (p<0.001). The relationship between the characteristics of the subjects and the serum phosphorus concentration, categorized by group, is shown in Table 2.

In a model adjusted for standard cardiovascular risk factors (age, sex, total cholesterol, LDL cholesterol, HDL cholesterol, body mass index, smoking, and systolic blood pressure), serum phosphorus still had statistically significant predictive properties (p=0.0016, odds ratio (OR) 16.6), regardless of age, sex, total cholesterol, and systolic blood pressure (Table 3).

In the final logistic regression model, in which all investigated variables were taken backward by

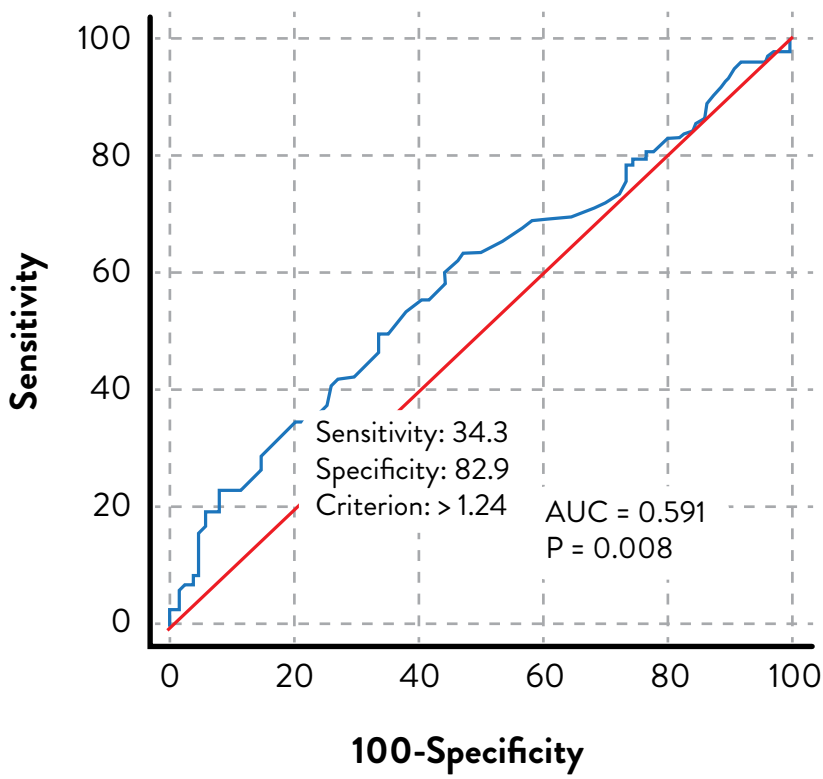

Fig. 2. ROC curve analyses to set an optimal serum phosphorus cut-off value for discriminating between subjects with and without a cardiovascular event.

Table 2. Baseline characteristics by serum phosphorus level in categories

Characteristic	P≤0.8 mmol/L n=7	P 0.9-1.2 mmol/L n=226	P≥1.3 mmol/L n=47	p
No CV event CV event	4/7 (57.1%) 3/7 (42.85%)	121/226 (53.53%) 105/226 (46.46%)	15/47 (31.91%) 32/47 (68.09%)	0.024*
Age (years)	64±11.4	62.7±9.7	65.7±9	0.145
Male Female	7/7 (100%) 0/7 (0%)	135/226 (59.7%) 91/226 (40.3%)	20/47 (42.6%) 27/47 (57.4%)	0.007* (1vs3)
SBP (mmHg)	140 IQR (137-152)	140 IQR (130-150)	132 IQR (130-145)	0.281
DBP (mmHg)	82 IQR (80 - 91)	80 IQR (77-90)	80 IQR (75.5-90)	0.714
Smoker Non-smoker	4/7 (57.1%) 3/7 (42.9%)	109/226 (48.2%) 117/226 (51.8%)	19/47 (40.4%) 28/47 (59.6%)	0.538
BMI (kg/m^2)	28.7 IQR (25.5-33.1)	29.6 IQR (26-32.8)	29.4 IQR (26.8-32.65)	0.970
Insulin use (a) OHAs use (b) a + b	1/7 (14.3%) 4/7 (57.1%) 2/7 (28.6%)	13/226 (5.8%) 170/226 (75.2%) 43/226 (19%)	2/47 (4.3%) 39/47 (82.9%) 6/47 (12.8%)	0.560
Hemoglobin (g/L)	151.3±15.6	142.1±13.4	136.6±14.5	0.007*
HbA1C (%)	7.8 IQR (6.8-8.3)	7.2 IQR (6.2-8.3)	7.2 IQR (6.6-8.8)	0.695
MDRD eGFR**	85 IQR (73.5-96)	80 IQR (70-91)	74 IQR (61-84.5)	0.042* (2vs3)
CKD EPI eGFR	84 IQR (72.5-90)	80 IQR (68.3-90)	72 IQR (61.5-85)	0.030* (2vs3)
Uric acid (mmol/L)	290 IQR (266-338)	337 IQR (267.3-392.5)	343 IQR (280-438.5)	0.133
Total cholesterol (mmol/L)	5 IQR (4.7-5.3)	4.8 IQR (4-5.8)	4.6 IQR (3.7-5.75)	0.552
HDL (mmol/L)	0.9 IQR (0.9-1.2)	1.1 IQR (0.9 - 1.3)	1.1 IQR (0.9-1.25)	0.472
LDL (mmol/L)	2.9 IQR (2.8-3.3)	3 IQR (2.2-3.7)	2.6 IQR (1.7-3.7)	0.456
TRG (mmol/L)	2.4 IQR (1.8-2.8)	1.7 IQR (1.2-2.2)	1.8 IQR (1.2-2.25)	0.255
Ca (mmol/L)	2.5 IQR (2.4-2.5)	2.4 IQR (2.4-2.5)	2.4 IQR (2.39-2.525)	0.635
Ca x P (mmol^2/l^2)	1.9±0.1	2.6±0.3	3.3±0.5	<0.001*
CRP (mg/L)	2.1 IQR (1.5-2.7)	1.8 IQR (1.1-3.3)	1.8 IQR (1.1-3)	0.951
Albumin (g/l)	44.5 IQR (43.2-46.1)	43.7 IQR (41.45.9)	42.4 IQR (38.5-45)	0.028* (2vs3)

*statistically significant at p<0.05; **mL/min/1.73 m^2; CV = cardiovascular; SBP = systolic blood pressure; DBP = diastolic blood pressure; BMI = body mass index; OHAs = oral hypoglycemic agents; HbA1c = hemoglobin A1c; MDRD = Modification of Diet in Renal Disease; CKD-EPI = Chronic Kidney Disease Epidemiology Collaboration; eGFR = estimated glomerular filtration rate; HDL = high-density lipoprotein; LDL = low-density lipoprotein; TRG = triglycerides; Ca = calcium; P = phosphorus; Ca x P = calcium–phosphorus product; CRP = C-reactive protein

selecting variables, serum phosphorus was statistically significantly associated with CV events (OR=14.4), regardless of gender, BMI, diabetic therapy, HbA1c, MDRD eGFR, total cholesterol, calcium, and albumin (Table 4).

Discussion

According to the results of this study, there is a statistically significant difference in the mean serum phosphorus level between subjects with and without a CV event. When serum phosphorus was observed as

Table 3. Adjusted logistic regression model for standard CV risk factors, analyzing serum phosphorus predictive properties for a CV event

Variable	OR; 95% CI	p
(s) P**	OR 16.5697 95% CI [2.9108-94.3235]	0.0016*
Age (years)	OR 1.0862 95% CI [1.0490-1.1248]	<0.0001*
Male gender	OR 2.6893 95% CI [1.4195-5.0949]	0.0024*
Total cholesterol**	OR 0.5103 95% CI [0.2677-0.9726]	0.0409*
LDL**	OR 1.2387 95% CI [0.6101- 2.5150]	0.5535
HDL**	OR 1.0888 95% CI [0.3964-2.9908]	0.8689
BMI (kg/m^2)	OR 0.9824 95% CI [0.9211-1.0479]	0.5902
Smoking	OR 1.0942 95% CI [0.5966-2.0067]	0.7711
SBP (mmHg)	OR 0.9772 95% CI [0.9606-0.9941]	0.0084*

*statistically significant at p<0.05; **mmol/L; 95% CI = 95% confidence interval; CV = cardiovascular; OR = odds ratio; P = phosphorus; LDL = low-density lipoprotein; HDL = high-density lipoprotein; BMI = body mass index; SBP = systolic blood pressure

Table 4. Fully adjusted logistic regression model analyzing serum phosphorus' predictive properties for CV events

Variable	OR; 95% CI	p
P (mmol/L)	OR 14.3673 95% CI [1.4285-144.4995]	0.0236*
Male gender	OR 8.7834 95% CI [3.3415-23.0876]	<0.0001*
BMI (kg/m^2)	OR 0.9047 95% CI [0.8322-0.9834]	0.0186*
Insulin+OHAs use	OR 0.3113 95% CI [0.1119-0.8657]	0.0253*
HbA1c (%)	OR 0.6057 95% CI [0.4585-0.8002]	0.0004*
MDRD eGFR (mL/min/1.73 m^2)	OR 0.9634 95% CI [0.9383-0.9892]	0.0057*
Uric acid (mmol/L)	OR 1.0050 95% CI [0.9999-1.0101]	0.0544
Total cholesterol (mmol/L)	OR 0.7149 95% CI [0.5198-0.9832]	0.0390*
Ca (mmol/L)	OR 4133.2681 95% CI[61.4354-278079.1631]	0.0001*
Albumin (g/L)	OR 0.5141 95% CI [0.4298-0.6149]	<0.0001 *

Total p < 0.0001; *statistically significant at p<0.05; CV = cardiovascular; BMI = body mass index; OHAs = oral hypoglycemic agents; HbA1c = hemoglobin A1c; MDRD = Modification of Diet in Renal Disease; eGFR = estimated glomerular filtration rate; Ca = calcium; P = phosphorus

a continuous variable, the results showed statistically significant but relatively weak discriminatory properties for distinguishing patients with and without CV events. ROC analysis indicated that the optimal cut-off value for discriminating patients with a CV event > 1.24 mmol/L had a low sensitivity of 24.3% and a specificity of 82.9%. When serum phosphorus concentration was investigated as a categorical variable, an association was observed between elevated phosphorus concentrations in the high-normal range and a CV event. It remains unclear whether elevated phosphorus concentrations are the primary cause or a concomitant occurrence of metabolic complications associated with adverse cardiovascular events, as multiple molecules have been identified in the complex process of calcification of the cardiovascular system[11]. Elevated serum phosphorus and FGF-23 values and decreased calcium and vitamin D values[12-14] were measured in chronic kidney disease (CKD) stages 1-2 of diabetic patients with diabetic nephropathy. Several previous studies have confirmed that patients with diabetic nephropathy exhibit lower phosphaturia and higher serum phosphorus concentrations than patients without diabetic nephropathy, accompanied by concomitant increases in FGF-23 concentrations and lower vitamin D concentrations[15–17].

Using a series of multivariate logistic regression models, we examined the association between serum phosphorus levels and the presence of CV events. In the model adjusted for standard cardiovascular risk factors (age, sex, total cholesterol, LDL, HDL, body mass index, smoking, and systolic blood pressure), serum phosphorus showed statistically significant predictive properties, regardless of age, sex, total cholesterol, smoking, and systolic blood pressure. According to previous research, high-normal serum phosphorus

concentrations have also been associated with coronary artery calcification in young, healthy men[18] and were predictors of adverse cardiovascular events in the Framingham study[19]. According to a study by Chonchol *et al.*, in a group of patients with type 2 diabetes with normal or near-normal renal function, higher serum phosphorus concentrations were associated with cardiovascular mortality independently of other important factors[10].

According to the results of this study, higher serum phosphorus levels were statistically significantly associated with female gender when serum phosphorus concentration was investigated as a categorical variable. Only a few previous studies showed that known risk factors for hyperphosphatemia include female gender, although the reasons for this association remain unclear[20]. One study suggests that estrogen may act directly to suppress sodium-dependent phosphorus absorption in the renal proximal tubules by inducing phosphaturia and a decrease in serum phosphorus levels. The study indicates that menopausal and estrogen-deficient women are at increased risk of hyperphosphatemia[21].

In this study, no statistically significant association was found between BMI and serum phosphorus concentration as a categorical variable. Several years ago, studies observed an inverse relationship between serum phosphorus concentration and BMI, blood pressure, and plasma glucose concentration[22-25]. Given the known association of phosphorus from food with organ calcification in patients with renal failure, as well as the growing cognition that phosphorus can have a detrimental effect on health even in people with normal renal function, studies on the impact of diet in diabetic patients are needed, given the specific dietary regime required in that population.

This study also suggested a possible inverse relationship between serum phosphorus concentration (as a categorical variable) and systolic blood pressure values, but it was not statistically significant. The group of subjects with higher normal serum phosphorus concentrations had lower systolic and diastolic blood pressure. According to one study, multivariate analysis indicated an inverse relationship between serum phosphorus concentration and systolic blood pressure values after adjustment for age, sex, and BMI[26]. When explaining the inverse mechanism between serum phosphorus concentration and blood pressure values, the fact that many prescription drugs contain significant amounts of phosphorus should certainly be considered, and this is often overlooked and has not been extensively investigated. Sherman *et al.* reviewed the 200 most commonly prescribed medications in patients undergoing dialysis. The phosphorus content ranged from 1.4 to 111.5 mg. According to the above study, the phosphorus content was inconsistent and varied depending on the dose of the drug, the form of the preparation (tablets or syrup), the original or generic formulation, and the manufacturer[27]. In this study, serum phosphorus levels were not associated with other well-known risk factors for cardiovascular disease (CVD) (age, hyperlipidemia, and smoking).

This study has several limitations. We were unable to investigate the effect of diet on serum phosphorus concentration, which may also be present in subjects with normal to slightly decreased renal function who were included in this study. The question of the influence of dietary habits on the relationship between serum phosphorus concentration and CVD was raised, given that subjects with diabetes who were presumed to have dietary habits different from those of the general population were included. In addition, only one value of serum phosphorus was observed in this study, and the concentration of serum phosphorus is variable and influenced by many factors. According to the results of a study by Chonchol *et al.*, more than half of the variability in serum phosphorus concentration was attributed to variability within a single subject in that study[10]. The clinical significance of repeated laboratory measurements in predicting cardiovascular morbidity in any population is likely to be greater. Future studies assessing the association between serum phosphorus concentrations and cardiovascular outcomes should use time-dependent models with repeated laboratory values to confirm these findings. Additionally, in this study, the concentrations of 25-hydroxyvitamin D, 1,25-dihydroxyvitamin D, or PTH were not measured. These measurements would enhance our understanding of the mechanisms by which serum phosphorus concentrations could be responsible for the increased risk of cardiovascular disease in individuals with diabetes.

In conclusion, the serum phosphorus level is statistically significantly higher in patients with CV events and shows statistically significant but relatively

modest discriminant properties for distinguishing between patients with and without CV events. Serum phosphorus may be an important adjunctive factor in assessing cardiovascular risk. It remains unclear whether serum phosphorus concentration is a biomarker, causative factor, or both. Moreover, understanding the basis for this association will likely yield new insights into the pathophysiological mechanisms of cardiovascular diseases.

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Sažetak

SERUMSKI FOSFOR KAO ČIMBENIK RIZIKA KARDIOVASKULARNOGA POBOLA U BOLESNIKA SA ŠEĆERNOM BOLEŠĆU TIPA 2

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Fosfor igra važnu ulogu u brojnim fiziološkim procesima. Mnoga istraživanja povezala su koncentraciju fosfora s povećanim rizikom za kardiovaskularna (KV) oboljenja i smrtnost. Ovo je istraživanje proučavalo povezanost visoko normalnih vrijednosti serumskog fosfora i kardiovaskularnog oboljenja u bolesnika s dijabetesom tipa 2. Proučavane su dvije skupine ispitanika s dijabetesom tipa 2 u dobi od 18 godina i više. Jedna skupina (140 ispitanika) nije imala KV događaj, a druga je skupina (140 ispitanika) imala jedan ili više KV događaja. Pokazalo se da je serumski fosfor statistički značajno viši u bolesnika s KV događajem i ima statistički značajna, ali relativno skromna diskriminacijska svojstva za razlučivanje bolesnika s i bez KV događaja (AUC 0.591; 95% C.I. 0.531 - 0.649; p=0.008). Univarijatni model logističke regresije serumskog fosfora kao kontinuirane varijable pokazao je statistički značajnu povezanost s prisutnošću kardiovaskularnog događaja (omjer vjerojatnosti (OR) = 7.1849, interval pouzdanosti od 95% (CI) 1.77728 - 29.1190; p=0.0057). Za svaki porast serumskog fosfora za 1 mmol/L, vjerojatnost KV događaja povećava se 7.4 puta. Nakon multivarijabilnog modela logističke regresije, serumski je fosfor i dalje bio povezan s KV događajem (p = 0.0016; OR 16.6), neovisno o dobi, spolu, ukupnom kolesterolu i sistoličkom krvnom tlaku.

Ključne riječi: *Serumski fosfor; Hiperfosfatemija; Kardiovaskularni morbiditet; Šećerna bolest tip 2*