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Original Article

Association of body mass index with serum calcium and phosphate levels

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ABSTRACT

Objective: It has been shown that several environmental and physiological factors can affect on the serum levels of calcium and phosphate. The objective of the present study was explored the relationship between serum calcium and phosphate levels with anthropometric and hematological markers.
Methods: 908 subjects were recruited from the Mashhad stroke and heart atherosclerosis disorder (MASHHAD) program. Anthropometric parameters, liver/kidney function tests (e.g., Urea nitrogen, creatinine, urea and uric acid, creatinine, AST, ALT) were determined in all participants. Serum concentrations of calcium and phosphate were measured using Autoanalyzer BT3000P (Pars Azmoon kit, Tehran, Iran). SPSS software was used for statistical analyses.
Results: We observed that obese subjects had a lower level of serum calcium ($p<0.05$). Moreover, a relationship was detected between serum phosphate level and different menopausal status ($p<0.05$). Serum calcium and phosphate did not change by increasing age in the population. Additionally, there was a correlation between lymphocyte count with serum phosphate level ($p<0.05$). No statistically different were detected for the levels of calcium/phosphate with respect to smoking status, physical activity, lipid profile, liver and renal function markers.
Conclusion: We found an association between serum calcium and BMI as well as with serum phosphate and menopausal status.

1. Introduction

Calcium is the fifth common element and the abundant cation of our bodies [1]. The serum calcium for adults is 4.5–6.5 mg/dl (1.12–1.62 mmol/l) and the recommended daily allowance (RDA) in adults is 1000–1200 mg/dl (249.5–299.4 mmol/l). Calcium is necessary for bone health, appropriate neurological synapses, cardiac function and coagulation [2,3]. Phosphorus is another important element, which is present in bones and connective tissues. Serum phosphate in adults is 0.81 mg/dl (0.26 mmol/l) to 1.45 mg/dl (0.46 mmol/l) [4]. It's RDA for adults is 700 mg/dl (225.8 mmol/l) [3].

Osteoporosis is an important health problem, with an increasing prevalence [5]. Osteoporosis is often associated with low levels of calcium and phosphorus [6]. Several factors have been reported to influence the serum levels of calcium and phosphate, including environmental and physiological factors such as diet, and particularly vitamin D content, age, gender, quality of life, menopausal

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status, social-economic status, adiposity, smoking habit, sun exposure, skin color, physical activity, hypertension and malabsorption or chronic diarrhea [7–15]. Other studies have shown a correlation between age and calcium/phosphorus absorption [7,14–17]. It has been reported that calcium absorption in osteoporotic patients is less than healthy people [7,18]. Several studies have evaluated the association of dietary intake with serum calcium and phosphate levels, although the value of having a diet high in calcium and phosphorus on increasing the serum levels of these factors is still controversial [7,8,18–22]. Therefore, this study aimed to investigate the relationship between serum calcium and phosphate levels with demographic, anthropometric, and liver/kidney function in 908 subjects, who were recruited from the Mashhad stroke and heart atherosclerosis disorder (MASHAD) study.

2. Materials and methods

2.1. Population

In the present study, 908 subjects were recruited as part of the Mashhad stroke and heart atherosclerosis disorder (MASHAD) program. Exclusion criteria were pregnancy and lactation, established cardiovascular disease or diabetes, consumption of dietary supplements, thyroid disorders, and cancers. The protocol was approved by the Ethics Committee of Mashhad University of Medical Science (MUMS).

2.2. Demographic parameters

Information on demographic and clinical variables (such as age, sex and osteoporosis), and individuals health condition (having hypothyroidism or hyperthyroidism, rheumatoid arthritis, diabetes, cancer, osteoporosis, smoking, etc.), as well as information related to menopausal status, physical activity, education, and income were collected through interviews by completing the general information questionnaires, as described previously [23].

Postmenopausal women had passed the last menstrual period. Premenopausal women who were in reproductive period of their life before final menstrual period [24].

2.3. Anthropometric and biochemical measurements

Anthropometric parameters (e.g., height, body weight, body mass index (BMI), waist circumference (WC), and hip circumference (HC) were assessed in all the subjected as previously described [25,26]. Urea nitrogen, creatinine, urea and uric acid, creatinine, aspartate transaminase (AST), alanine transaminase (ALT) were measured as described previously [27,28]. Fat % <25% and <30% as considered normal in men and women, respectively [29].

2.4. Physical activity

Physical activity level (PAL) of our subjects was evaluated as the total energy required within 24 h as a ratio of the basal metabolic rate for a 24-h period. PAL was studied using James and Schofield questionnaire about the human required energy. Questions were sorted based upon the activity time: time spent on activities during work (including housework), during non-working time (like watching TV) and in bed (bed rest and sleep) and then scored.

2.5. Measurement of calcium and phosphate

Serum calcium and phosphate levels were measured using Pars Azmoon kit (Tehran, Iran), on an Autoanalyzer BT3000. Briefly, calcium measurement was done through photometric method, in which calcium in an alkaline environment with Cresolphthalein Complexone forms a purple complex. A photometric method was used for measurement of phosphate. In this method, the ammonium molybdate and sulfuric acid and the sample phosphorus react together and the colored combination of phosphorus molybdate is produced. Intensity of the color generated is proportional to the amount of calcium or phosphate in the sample.

2.6. Statistical analysis

Data was analyzed using SPSS V.16 software (SPSS® Inc., Chicago, IL, USA). Descriptive statistics, including mean ± standard deviation (SD) or median and interquartile range were used for normal or non-normal distribution's data, respectively. T-student, Mann-Whitney or U Chi-square test were used for normally, not normally distributed variables or categorical parameters, respectively. All the data were analyses two-sided and statistical significance was set at $P < 0.05$ [30].

3. Results

3.1. Characteristics of the population

The demographic and clinical characteristics of population are shown in Table 1. This study included 51% women with mean age of 47.9 years who 41.3% and 58.7% were menopause and menstrual women, respectively. These data showed that the levels of BMI, Fat %, serum lipid profile, uric acid, creatinine, ALT, calcium and phosphate were significantly different between men and women ($p^{*}0.001$). Serum calcium and phosphate levels were 9.19 ± 1.02 mg/dl and 4.20 ± 0.80 mg/dl, respectively in total population. The results showed there were significant higher in serum levels of Ca (9.28 ± 0.92 vs. 9.11 ± 1.10 mg/dl, $p < 0.05$) and phosphate (4.27 ± 0.72 vs. 4.12 ± 0.88 mg/dl, $p < 0.01$) in females compared to males (Table 1).

3.2. Association of age, physical activity and smoking status with serum calcium and phosphate in male and female

The association of age, physical activity and smoking status with serum calcium and phosphate were assessed in Table 2. First, in order to determine whether there was any relationship between age and serum calcium and phosphate, we divided the population based on age and determined the levels of serum calcium and phosphorus. As shown in Table 2, serum levels of Ca change by increasing age in women ($p < 0.05$). This analysis revealed that Ca/P and phosphate level in non-smoker and current-smoker group were higher than other groups, respectively in male ($p < 0.05$) (Table 2).

3.3. Correlation of physiological parameters with serum Ca, P and Ca/P ratio

We also assessed the correlation between the physiological parameters with serum Ca, phosphate and ca/p ratio (Table 3). In total population, our data showed a significant correlation between BMI, cholesterol and osteoporosis with Ca and also it is observed a significant correlation between osteoporosis, menopausal status and creatinine with phosphate level and Ca/P, respectively ($p^{*}0.05$).

We further investigated serum level of Ca, phosphate and Ca/P ratio with physiological parameters in total population and pre/and post-menopausal women, respectively (Tables 4 and 5). Interestingly, we demonstrated that obese people had a lower level of calcium ($p^{*}0.05$). Conversely, no statistically significant different were detected for the levels of calcium with respect to menopausal

Table 1
Demographic and clinical characteristics of population.

		Total	Male	Female	P-value
Age (year)	35–44	385	193 (50.1%)	192 (49.9%)	0.742
	45–54	336	160 (47.6%)	176 (52.4%)	
	55–64	169	80 (47.3%)	89 (52.7%)	
BMI (mg/kg^2)		27.73±4.58[1]	26.43 ± 4.18	28.97 ± 4.60	0.001
Fat (%)		32.90 ± 12.90	24.60 ± 12.65	40.77 ± 6.72	0.001
Serum Cholesterol (mg/dl)		187 (163–214)	180 (158–210)	192 (167–219)	<0.001
Weight (Kg)		73.06 ± 12.96	75.78 ± 13.47	70.47 ± 11.90	0.001
Height (m)		1.62 ± .09	1.69 ± .06	1.55 ± .05	0.001
Waist circumference (Cm)		94.64 ± 12.03	93.52 ± 10.95	95.71 ± 12.90	0.006
Hip circumference (Cm)		103.61 ± 8.84	101.29 ± 7.59	105.81 ± 9.37	0.001
Mid upper arm circumference (Cm)		30.27 ± 3.54	29.96 ± 3.53	30.57 ± 3.52	0.010
Serum Uric Acid (mg/dl)		4.82 ± 1.51	5.35 ± 1.56	4.32 ± 1.28	0.001
Serum Creatinine (mg/dl)		.88 ± .26	.92 ± .21	.83 ± .29	0.001
Serum AST (IU/l)		22.61 ± 11.84	23.11 ± 10.45	22.09 ± 13.14	0.338
Serum ALT (IU/l)		18.14 ± 12.93	19.75 ± 12.17	16.43 ± 13.51	0.004
Serum calcium (mg/dl)		9.19 ± 1.02	9.11 ± 1.10	9.28 ± .92	0.012
Serum phosphate (mg/dl)		4.20 ± .80	4.1295 ± .88	4.27 ± .72	0.006
Serum Ca/P		2.24 ± .41	2.26 ± .41	2.22 ± .41	0.181
Physical activity level		1.55 (1.35–1.72)	1.36 (1.21–1.58)	1.64 (1.53–1.76)	0.001
Smoking status	Non Smoker	613 (67.4%)	258 (58%)	355 (76.5%)	<0.001
	Ex-Smoker	95 (10.5%)	68 (15.3%)	27 (5.8%)	
	Current Smoker	201 (22.1%)	119 (26.7%)	82 (17.7%)	
Osteoporosis	Yes	90	10 (11.1%)	80 (88.9%)	0.001
	No	780	423 (54.2%)	357 (45.8%)	

P value: male versus female.
Values expressed as mean ± SD for normally distributed data, and median and interquartile range for non-normally distributed data. BMI, body mass index; AST; aspartate transaminase, ALT; alanine transaminase, Ca; calcium, P; phosphate.

Table 2
Serum Ca, P level and Ca/p ratio in different groups of age, Physical activity level and smoking status according to gender.

	Variables	Serum Ca (mg/dl)	Serum Phosphate (mg/dl)	Serum Ca/P	Serum Ca (mg/dl)	Serum Phosphate (mg/dl)	Serum Ca/P
		(Male)			(Female)		
Age (year)	35–44	9.13 ± 1.20	4.14 ± 1.11	2.28 ± 0.45	9.25 ± 0.94 ab	4.22 ± 0.71	2.24 ± 0.41
	45–54	9.08 ± 1.01	4.08 ± 0.73	2.25 ± 0.38	9.19 ± 1.01a	4.29 ± 0.73	2.19 ± 0.41
	55–64	9.00 ± 1.05	4.13 ± 0.59	2.21 ± 0.37	9.50 ± 0.65b	4.36 ± 0.68	2.23 ± 0.39
Physical activity level	Sedentary (1–1.39)	9.17 ± 0.89	4.14 ± 1.07	2.27 ± 0.39	9.14 ± 1.32	4.25 ± 0.92	2.23 ± 0.52
	low active (1.4–1.59)	9.23 ± 1.12	4.05 ± 0.66	2.32 ± 0.41	9.23 ± 0.82	4.32 ± 0.72	1.19 ± 0.40
	active (1.6–1.89)	8.83 ± 1.71	4.17 ± 0.61	2.15 ± 0.51	9.29 ± 0.99	4.25 ± 0.70	2.24 ± 0.42
	Very active (1.9–2.5)	9.02 ± 0.80	4.05 ± 0.57	2.26 ± 0.32	9.39 ± 0.71	4.26 ± 0.71	2.25 ± 0.37
Smoking	Non Smoker	9.17 ± 0.92	4.02 ± 0.67a	2.31 ± 0.38a	9.30 ± 0.93	4.28 ± 0.71	2.22 ± 0.42
	Ex-Smoker	9.00 ± 1.16	4.14 ± 0.67ac	2.21 ± 0.41ac	9.10 ± 0.81	4.11 ± 0.88	2.29 ± 0.47
	Current Smoker	9.03 ± 1.40	4.30 ± 1.32c	2.18 ± 0.46c	9.23 ± 0.90	4.29 ± 0.70	2.19 ± 0.35
Fat %	<25%	9.12 ± 0.92	4.17 ± 1.02	2.24 ± 0.39	9.22 ± 0.81	4.48 ± 0.69	2.09 ± 0.30
	≥25%	9.13 ± 1.18	4.03 ± 0.72	2.29 ± 0.42	9.28 ± 0.93	4.27 ± 0.72	2.23 ± 0.41

Different letters showed significant association between groups; p.value < 0.05.
Values expressed as mean ± SD.

Table 3
Correlation of Physiological Parameters with Serum Ca, P and Ca/P ratio.

		Serum Ca (mg/dl)		Serum Phosphate (mg/dl)		Serum Ca/P	
		P	r	P Value	r	P Value	r
Total population	BMI (mg/kg^2)	0.029	−0.072	0.192	0.043	0.965	0.001
	Menopausal Status	0.215		0.026		0.770	
	Osteoporosis	0.004		0.048		0.082	
	Cholesterol (mg/dl)	0.003	0.098	0.100	0.055	0.887	0.005
	Serum Uric Acid (mg/dl)	0.476	0.024	0.894	−0.004	0.049	0.065
	Serum Creatinine (mg/dl)	0.747	0.013	0.451	−0.031	0.002	0.126
	Serum AST (mg/dl)	0.432	0.035	0.863	−0.008	0.096	0.075
	Serum ALT (mg/dl)	0.230	0.540	0.200	−0.058	0.188	0.060
Menopausal Women	BMI (mg/kg^2)	0.957	0.005	0.975	0.003	0.981	−0.002
	Osteoporosis	0.371	−0.080	0.344	−0.085	0.463	0.066
	Cholesterol (mg/dl)	0.366	0.078	0.447	0.066	0.647	−0.040
	Serum Uric Acid (mg/dl)	0.580	0.048	0.804	−0.022	0.340	−0.082
	Serum Creatinine (mg/dl)	0.800	0.026	0.494	0.071	0.830	0.023
	Serum AST (mg/dl)	0.564	0.072	0.857	0.023	0.803	−0.031
	Serum ALT (mg/dl)	0.823	0.028	0.895	0.17	0.603	−0.065

Table 4
Association of serum Ca, P level and Ca/p ratio with Physiological Parameters in total population.

		Serum Ca (mg/dl)	Serum Phosphate (mg/dl)	Serum Ca/P
BMI (mg/kg^2)	Normal	9.32 ± 0.99*	4.18 ± 0.83	2.24 ± 0.41
	30<	9.15 ± 1.02	4.24 ± 0.71	2.25 ± 0.42
Menopausal Status	Postmenopausal	9.35 ± 0.91	4.37 ± 0.66*	2.18 ± 0.37
	Premenopausal	9.24 ± 0.92	4.22 ± 0.74	2.25 ± 0.43
Osteoporosis	Yes	9.40 ± 0.67	4.35 ± 0.60	2.19 ± 0.30
	No	9.17 ± 1.06	4.17 ± 0.83	2.25 ± 0.42
Serum Cholesterol (mg/dl)	Normal	9.11 ± 1.07	4.16 ± 0.68	2.23 ± 0.42
	High	9.33 ± 0.92	4.25 ± 0.96	2.26 ± 0.40
Serum Uric Acid (mg/dl)	Normal	9.18 ± 1.01	4.19 ± 0.82	2.24 ± 0.42
	Abnormal	9.37 ± 1.06	4.27 ± 0.56	2.22 ± 0.33
Serum Creatinine (mg/dl)	Normal	9.16 ± 1.08	4.20 ± 0.88	2.23 ± 0.43
	Abnormal	9.32 ± 0.92	4.15 ± 0.67	2.30 ± 0.45
Serum AST (mg/dl)	Normal	9.16 ± 1.08	4.20 ± 0.90	2.24 ± 0.43
	Abnormal	9.18 ± 1.21	4.19 ± 0.72	2.23 ± 0.41
Serum ALT (mg/dl)	Normal	9.22 ± 1.18	4.20 ± 0.63	2.23 ± 0.38
	Abnormal	9.12 ± 1.04	4.19 ± 0.99	2.25 ± 0.45

Values expressed as mean ± SD. T-student test was used.
*p < 0.05.

Table 5
Association of serum Ca, P level and Ca/p ratio with Physiological Parameters in women.

		Post menopause			Pre menopause (mensturation)		
		Ca/P	P (mg/dl)	Ca (mg/dl)	Ca/P	P (mg/dl)	Ca/P (mg/dl)
BMI (mg/kg^2)	Normal	9.33 ± 1.06	4.34 ± 0.66	2.19 ± 0.38	9.17 ± 0.91	4.20 ± 0.72	2.24 ± 0.43
	30<	9.38 ± 0.65	4.41 ± 0.66	2.17 ± 0.36	9.36 ± 0.94	4.25 ± 0.77	2.26 ± 0.43
Osteoporosis	Yes	9.44 ± 0.63	4.44 ± 0.50	2.14 ± 0.25	9.38 ± 0.64	4.29 ± 0.72	2.23 ± 0.36
	No	9.27 ± 1.02	4.31 ± 0.73	2.20 ± 0.41	9.23 ± 0.96	4.21 ± 0.75	2.25 ± 0.45
Cholesterol (mg/dl)	Normal	9.21 ± 0.65	4.29 ± 0.63	2.13 ± 0.35	9.23 ± 1.01	4.24 ± 0.75	2.23 ± 0.43
	High	9.45 ± 1.01	4.43 ± 0.68	2.18 ± 0.38	9.26 ± 0.73	4.16 ± 0.70	2.28 ± 0.43
Serum Uric Acid (mg/dl)	Normal	9.33 ± 0.91	4.37 ± 0.67	2.17 ± 0.37	9.24 ± 0.92	4.22 ± 0.74	2.25 ± 0.43
	Abnormal	9.76 ± 0.58	4.40 ± 0.63	2.26 ± 0.38	8.85 ± 0.91	4.26 ± 0.19	2.01 ± 0.30
Serum Creatinine (mg/dl)	Normal	9.33 ± 1.05	4.47 ± 0.66	2.12 ± 0.37	9.27 ± 0.87	4.22 ± 0.74	2.25 ± 0.42
	Abnormal	9.58 ± 0.62	4.30 ± 0.73	2.29 ± 0.43	9.35 ± 1.02	4.08 ± 0.62	2.35 ± 0.52
Serum AST (mg/dl)	Normal	9.28 ± 1.11	4.38 ± 0.64	2.15 ± 0.38	9.16 ± 0.91	4.21 ± 0.74	2.23 ± 0.42
	Abnormal	9.25 ± 0.21	4.30 ± 0.28	2.15 ± 0.19	9.53 ± 0.37	4.66 ± 0.80	2.07 ± 0.28
Serum ALT (mg/dl)	Normal	9.33 ± 1.55	4.53 ± 0.43	2.07 ± 0.37	9.14 ± 1.28	4.30 ± 0.86	2.18 ± 0.37
	Abnormal	9.27 ± 0.72	4.34 ± 0.77	2.19 ± 0.37	9.16 ± 0.75	4.18 ± 0.67	2.25 ± 0.44

Values expressed as mean ± SD. T-student test was used.

status, education, smoking, activity, Fat %, uric acid, creatinine, AST and ALT (Table 4). Additionally, we found that the levels of phosphate were significantly different between different status of menopausal ($p < 0.05$) (Table 4). Moreover, serum level of Ca, phosphate and Ca/P ratio were presented in pre and post-menopausal women that we did not show significant association between the physiological parameters with serum Ca, phosphate and Ca/P ratio (Table 5).

4. Discussion

The results of current study showed that there was a significant association between BMI, cholesterol and osteoporosis with Ca. Moreover, a significant correlation was found between osteoporosis, menopausal status and creatinine with phosphate level and Ca/P. the results indicated that there was a significant lower in serum calcium levels among obese participants compared to the normal subjects (p<0.05).

There is growing body of data evaluating the association of age, sex and menopause with serum calcium and phosphorus levels [7,10,15–17,31–37], although they have reported conflicting results with respect to the effect on these parameters on these two elements. In particular, in a study on 7005 subjects, the effects of age, sex and menopause on serum calcium and phosphorus was investigated and showed the lower level of serum calcium in women than men [16]. Conversely, another study on 1113 subjects showed the higher serum level of calcium in men than women [37]. However, several other studies have illustrated no differences in calcium levels between men and women [10,32,38]. For example, in a study of the relationship between gender and serum calcium, albumin and phosphate levels in 1300 school children, 8–10 years old, no significant difference was detected between the levels of calcium [32]. Similarly, we also found no association between age with calcium and phosphorus levels in our population. About association of serum Ca and P levels with gender, we found that serum Ca and P levels were significantly higher in women than men, probably due to menopausal status and hormonal disorders on Ca metabolism. In agreement of our data, Kesteloot et al., on 9321 men and 421 women showed a similar finding [38].

Furthermore, we fund no relationship between the levels of Ca and P with smoking status and physical activity [3,9,39]. In addition, we assessed the association of smoking on serum calcium and phosphorus. Our data revealed no significant relationship between parameters, which is agreement with several other studies [9,39]. Brot's et al., evaluated the impact of smoking on vitamin D status and calcium metabolism, suggesting no relationship between the serum calcium levels in smokers and non-smokers [9]. Similarly, Haglin et al., in 2014 found a similar result, although they

found a link between smoking status of people and increased phosphorus levels, which can be explained by several factors, including increased phosphorus reabsorption in the renal tubules due to PTH; mobilization of phosphorus from bone via the blood, leading to increased bone loss in smokers as compared to non-smokers [39].

Moreover, our data showed obese subject had lower levels calcium levels. This observation is in line with several previous studies [14]. Additionally, we observed an association between phosphorus level and menopausal status. In line with our data, Qureshi et al., reported hypocalcemia in postmenopausal women [35]. Another study showed the higher level of phosphorus in women after menopause [3].

In conclusion, we suggest the association of serum calcium with BMI, indicating the obese subjects had a lower level of calcium. Further investigations are warranted to explore the clinical impact of biochemical parameters and hematological factors on serum calcium and phosphorus levels with respect to osteoporosis.

In this study, while achieving comprehensive information about the levels of serum calcium and phosphorus in Mashhad, which indicates inadequate intake of calcium in food, the correlation of both elements with some parameters such as serum parameters, physiological factors, physical status and gender was found, these findings show that the impact of these factors should also be considered in analysis of serum calcium, phosphorus and their metabolic functions.

Study limitations

This was a cross-sectional study, supporting more research of the association of Ca and P with BMI and biochemical factors in a prospective study. Moreover, the serum Ca levels should be corrected for albumin in future studies.

Conflicts of interest

The authors have no conflict of interest to disclose.

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