Original Article

Serum Osteoprotegerin in Relation to Metabolic Status, Severity, and Estimated Risk of Subsequent Coronary Heart Disease

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Abstract

Background: Osteoprotegerin (OPG), a key factor in bone remodeling and vascular calcification, has been suggested to be associated with cardiovascular events. This study sought to assess the relationship between plasma OPG, anthropometric, metabolic status, severity and extent of coronary artery calcification, and the two-year recurrence risk of coronary event in patients with coronary heart disease (CHD).

Methods: A total of 155 consecutive patients with symptoms suggestive of CHD were enrolled in this cross-sectional study. Blood samples were taken for laboratory tests. Coronary angiography and cardiac CT scan were performed to assess the severity and extent of involved vessels. Two-year risk of subsequent CHD was estimated based on the computational Framingham risk prediction model.

Results: OPG level was in direct linear association with age ($\beta = 0.38$, $p < 0.001$), waist to hip ratio ($\beta = 0.17$, $p < 0.05$), hs-CRP ($\beta = 0.17$, $p < 0.05$), systolic and diastolic blood pressure ($\beta = 0.17$, $p < 0.05$; $\beta = 0.23$, $p < 0.01$), and HbA1c ($\beta = 0.17$, $p < 0.05$). After age-sex adjustment, only HbA1c ($\beta = 0.15$, $p < 0.05$) was a significant indicator of serum OPG. OPG showed significant linear association with the coronary calcium score (CCS), and the number of involved vessels even after adjustment for age, sex, diabetes, blood pressure, and markers of bone-calcium metabolism ($\beta = 0.27$, $P < 0.05$; $\beta = 29$, $P < 0.01$). There is a significant positive association between two-year risk of subsequent CHD and serum OPG in females ($\beta = 0.45$, $P < 0.01$) but not in males.

Conclusion: Increased OPG is independently associated with the severity and extent of CHD. This study also proposes OPG as a potential marker in predicting the risk of subsequent CHD, in females.

Keywords: Biological markers, blood, cardiac imaging, coronary heart diseases, osteoprotegerin

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Introduction

Coronary heart disease (CHD) is the major cause of death globally.[1,2] Diabetes is associated with more than three-fold increase in risk of CHD and contributes to CHD morbidity and mortality to a great extent.[3–5] Osteoprotegerin (OPG) is a soluble tumor necrosis factor (TNF) receptor-like molecule that is expressed and secreted from a variety of tissues, including bone (osteoblasts) and vasculature cells (endothelial and vascular smooth muscle). OPG binds as a decoy receptor on the receptor activator for NF-kB ligand (RANKL) and prevents osteoclastogenesis.[1,6–8] Thereby, it acts as a key factor in bone remodeling and vascular calcification.[7–10]

The precise role of OPG in the vascular system is still unclear. Although experimental studies on animal models suggest a protective role for OPG against vascular calcification, clinical studies have indicated that increased OPG is positively associated with vascular calcification and cardiovascular events independent of the conventional risk factors.[1,7–11]

Serum OPG is proposed to be substantially interrelated with age, state of glycemic control, hypertension and CHD mortality.[7,9–13] Recent investigations on the pathogenesis of atherosclerosis suggest the calcifying role of calcium and insulin like growth factor 1 receptor (IGF1-R) to be modulated by serum OPG. It has also been suggested that moderate calcium levels increase OPG and IGF1-R expression and block vascular calcification. Nevertheless, high levels of exogenous calcium inhibit IGF1-R expression; leading to enhanced calcification.[14]

Epidemiological research in the Framingham Heart Study devised statistical models using multivariable CHD risk appraisal to predict the risk of CHD over an extended period. This method is now a widely used clinical tool for guiding the delivery of preventive medicine.[4,15,16]

In the present study, we assessed the relationship between plasma OPG, metabolic parameters, and the severity and extent of coronary artery calcification in patients with symptoms suggestive of CHD. We also compared these risk factors among CHD patients with and without history of diabetes. The distinctive point of this study is considering the hidden influence of bone metabolism markers on serum OPG – CHD association. We aimed to evaluate whether serum OPG – CHD association is modulated through the markers of bone metabolism and whether serum OPG can warn the recurrence of CHD in a specific population.[15]

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Patients and Methods

Participants

In this cross sectional study, patients with symptoms suggestive of CHD were recruited consecutively from the cardiology clinic of Vali-Asr hospital, Tehran University of Medical Sciences (TUMS), between January 2011 and June 2012. Patients with prior diagnosis of chronic kidney disease, declined e-GFR (< 60 ml/min), and ejection fraction < 40 were excluded.

After obtaining written informed consent, characteristics such as age, gender, smoking status, and medication were gathered through detailed history taking. The study protocol was approved by research ethics committee of TUMS according to the principles of the Declaration of Helsinki.

Measurements and Laboratory Investigations

The patients' weight, height, and waist and hip circumference were measured precisely. The systolic and diastolic blood pressures were taken twice with at least 10 minutes interval. Body mass index (BMI) was calculated as weight (Kg) divided by height squared (m^2). Homeostasis Model Assessment of Insulin Resistance (HOMA-IR) was calculated as (Fasting plasma glucose (mg/dL) × Fasting insulin (U/l) / 405).[17] Diabetes mellitus was diagnosed according to the criteria of the American Diabetes Association, 2011.[18]

After 12-hour overnight fasting, venous blood samples were collected from all participants at baseline. Fasting plasma glucose (FPG) and fasting insulin levels were assessed by glucose oxidase method and radioimmunoassay (Immunotech, Prague, Czech Republic), respectively. Lipid profile, including total cholesterol, triglyceride, LDL-C and HDL-C, was measured by direct enzymatic method (Parsazmun, Karaj, Iran). HbA1c was measured using High Performance Liquid Chromatography (HPLC). Plasma concentrations of highly sensitive C-reactive protein (hs-CRP) were determined using available commercial kits (Diagnostics Biochem Inc, Canada). Osteoprotegerin was measured using Enzyme Linked Immunosorbent Assay (Biomedica Medizinprodukte GmbH & Co KG). Intra-assay and inter-assay CV for OPG measurements were 4% and 8%, respectively. The glomerular filtration rate (GFR) was estimated from the Modification of Diet in Renal Disease (MDRD) equation; as per the following: e-GFR = 175.0 × (serum creatinine^1.154) × (age ^0.203) × 0.742 (if female).[2]

Assessing the Severity and the Recurrence Risk of CHD

All patients underwent cardiac CT scanning to evaluate the extent of coronary artery calcification. Coronary Calcium Scoring (CCS) was determined non-invasively using multi-detector computed tomography (MDCT). Cardiac MDCT provides direct visualization of the coronary artery lumen and wall and is widely regarded as a screening method in subjects with suspected CHD.[19,20] Using cardiac MDCT, calcified plaques can be quantified without contrast administration.[20–22] CCS of zero has high negative predictive value for excluding obstructive CHD. CCS > 400 is highly suggestive of at least one significant coronary vessel stenosis. However, results should be interpreted in the context of other factors, including patients' age, gender, symptoms, risk factors, and the number of involved vessels. Despite that, coronary angiography remains the gold standard when cardiac CT is inconclusive or suggestive of significant CHD.[11,19]

Diagnostic coronary angiography was performed by experienced cardiologists. The data were re-evaluated by a single experienced cardiologist blinded to the patients' clinical information or CT scan results. The inter-rater agreement was found to be substantial (kappa coefficient = 0.74, P < 0.001). Obstructive CHD was diagnosed when there was ≥ 50% stenosis in one or more major coronary arteries.[22,23] Patients were categorized into 4 groups according to the angiographic findings: from no significant stenosis to having one, two, or three vessels disease.

Gensini score was calculated based on the number of stenotic segments of coronary artery, the degree of lumen stenosis, and

Table1. Demographics of the study population with respect to their diabetes status

	All Patients (n = 155)	CHD patients with diabetes (n = 103)	CHD patients without diabetes (n = 52)	Sig
Male (%)	72.3%	67.3%	74.75%	0.32
Age (years)	56.8 ± 11.2	56.6 ± 10.9	56.9 ± 12.0	0.87
BMI (Kg/m ^{2})	26.1 ± 3.9	27.0 ± 4.7	25.66 ± 3.3	0.04
Waist (cm)	87.9 ± 11.1	90.3 ± 12.4	86.6 ± 10.2	0.04
Waist-Hip Ratio	0.99 ± 0.1	1.02 ± 0.1	0.97 ± 0.1	0.003
Medication (%)				
ACEI	61.3 %	65.4 %	59.2 %	0.45
Beta blocker	63.2 %	65.4 %	62.1 %	0.69
Diuretic	5.8 %	3.9 %	6.8 %	0.47
ARB	12.9 %	21.2 %	8.7 %	0.04
Nitrate	38.7 %	38.5 %	38.8 %	0.96
Aspirin	69.7 %	69.2 %	69.9 %	0.93
Statins	64.5 %	69.2 %	62.1 %	0.38
Symptoms (%)				
Palpitation	11.2 %	5.8 %	14.0 %	0.12
Typical chest pain	23.0 %	23.1 %	23.0 %	0.99
Atypical chest pain	13.2 %	9.6 %	15.2 %	0.34
Exertion chest pain	13.3 %	9.6 %	15.2 %	0.44
Dyspnea on exertion	53.6 %	65.4 %	47.5 %	0.03
No Symptom	11.8 %	7.6 %	13.6 %	0.27

Data represented as mean ± SD or proportion.
Abbreviations: BMI, body mass index; ACEI: Angiotensin-converting enzyme inhibitor; ARB: Angiotensin receptor blocker.

Table2. Clinical and laboratory characteristics of the study population with respect to their diabetes status

	All Patients (n=155)	CHD patients with diabetes (n=103)	CHD patients without diabetes (n=52)	Sig
Systolic BP (mmHg)	124.2 ± 18.6	127.5 ± 18.6	122.6 ± 18.5	0.12
Diastolic BP (mmHg)	76.5 ± 7.9	77.2 ± 7.9	76.13 ± 7.9	0.41
MAP (mmHg)	92.4 ± 10.7	94.0 ± 10.7	91.6 ± 93.9	0.19
Total Cholesterol (mg/dL)	181.1 ± 52.4	190.6 ± 107.3	176.3 ± 45.8	0.13
TG (mg/dL)	144.0 (109.2–180.0)	140.0 (110.0–175.0)	159.5 (110.0–231.5)	0.05
LDL-c (mg/dL)	103.0 ± 3.9	107.5 ± 44.6	100.6 ± 35.2	0.33
HDL-c (mg/dL)	42.7 ± 10.9	43.5 ± 12.8	42.4 ± 9.9	0.56
FBS (mg/dL)	113.5 ± 41.7	153.0 ± 51.9	94 ± 10.9	< 0.001
HBA1c (%)	5.9 ± 1.5	7.6 ± 1.4	5.04 ± 0.6	< 0.001
HOMA-IR	1.9 (1.3, 4.1)	2.9 (1.9, 4.4)	1.6 (1.1, 3.5)	0.01
Hs-CRP (mg/L)	21.0 (7.8–53.7)	20.5 (12.5–55.7)	22.0 (6.4–53.0)	0.72
OPG (pmol/L)	3.2 ± 1.2	3.6 ± 1.3	3.04 ± 1.2	0.01
PTH (pg/mL)	20.0 (16.8–24.0)	22.0 (18.0–27.0)	17.7 (15.8–20.0)	< 0.001
Calcium (mg/dL)	9.0 ± 0.5	9.0 ± 0.5	9.0 ± 0.4	0.9
25 (OH) Vit D (nmol/L)	32.0 (23.0, 67.0)	35.0 (20.0, 75.0)	32.0 (23.0, 64.0)	0.55
Creatinine (mg/dL)	1.0 ± 0.1	1.0 ± 0.1	1.0 ± 0.1	0.75
e-GFR (mL/min)	75.9 ± 14.7	74.5 ± 14.7	76.7 ± 14.7	0.40
LV EF (%)	46.8 ± 10.1	47.6 ± 7.7	46.4 ± 11.1	0.41
CCS	133.0 (33.0, 491.0)	170.5 (31.0, 316.0)	111.0 (32.0, 514.0)	0.30
Gensini score	31.0 (8.0, 55.0)	46.0 (20.0, 80.0)	22.5 (7.0, 52.5)	0.01
Total Framingham Point Score	12.9 ± 4.8	15.7 ± 4.3	11.5 ± 4.5	< 0.001
2-yr risk of subsequent CHD (%)	7.1 ± 3.6	8.5 ± 4.4	6.5 ± 3.1	< 0.001

Data represented as mean ± SD or median (25^{th}, 75^{th} centile).
Abbreviations: BP: blood pressure; MAP: Mean arterial pressure; TG: triglycerides; HDL-c: high density lipoprotein cholesterol; LDL-c: low density lipoprotein cholesterol; Hb A1c: haemoglobin A1c; HOMA-IR: homeostasis model assessment of insulin resistance index; hs-CRP: high sensitivity CRP; OPG: osteoprotegerin; PTH: parathyroid hormone; e-GFR: estimated glomerular filtration rate; CCS: coronary calcium scoring; LVEF: left ventricular ejection fraction.

the area involved. In brief, scores of 1, 2, 4, 8, 16, and 32 were assigned correspondingly to 25%, 50%, 75%, 90%, 99%, and 100% reduction in lumen diameter. This score multiplied by the vessel scores was used to assess the Gensini score as follows: LM ×5; the proximal segment of LAD ×2.5; the proximal segment of LCX × 2.5; the mid-segment of the LAD × 1.5; the RCA, the distal segment of the LAD, the posterolateral and the obtuse marginal artery × 1; other segments × 0.5.[24]

Two-year risk of CHD recurrence was estimated based on the computational model proposed by the Framingham Heart Study. Framingham subsequent coronary risk appraisal is a sex-specific risk prediction model, applicable to patients with at least one previous CHD or ischemic stroke who have survived the acute stage of that event. Input parameters include age, ratio of total cholesterol/HDL-C and diabetes status in both sexes, along with systolic blood pressure (SBP) and cigarette smoking status in females.[15, 16] Applicability of the Framingham risk score which is derived from the Framingham offspring cohort has been previously validated in Iranian populations.[25] Thus, we used the Framingham covariates and risk scoring system to predict the two-year risk of CHD.

Statistical Analysis

Statistical analysis was performed using SPSS software (v.16; Chicago, Illinois, USA). The normality of each variable distribution was determined by the Kolmogorov–Smirnov test. Normally distributed variables were expressed as mean ± standard deviation (SD). Median (25^{th}, 75^{th} centile) was used to describe variables distributed otherwise. Independent t-test was performed to evaluate difference in continuous variables across different groups. Chi-square (χ^2) or Fisher exact tests were employed to assess the difference between distributions of the categorical variables. Binary logistic regression analysis was performed to assess the association between serum OPG and diabetes.

The association of serum OPG with metabolic parameters and the Framingham risk score was determined using linear regression, considering serum OPG as a dependent variable (Table 3, 5). On the other hand, multiple linear regression models were designed to evaluate the association of serum OPG (as a determinant) and the severity of CHD (as an outcome). Stepwise adjustments were performed for confounding variables including age, sex, diabetes, blood pressure, and markers of bone calcium metabolism. Standardized coefficients were calculated. Standardized coefficients present the regression coefficients per each standard deviation change in the determinant. By this standardization, coefficients from different determinants, with different magnitudes of scales, can be compared in their effect size. Statistically significant level was considered to be P value < 0.05.

Table3. Linear regression coefficients of the study variables as determinants of serum OPG

	Unadjusted		Age-Sex adjusted	
	Beta	**PV**	**Beta**	**PV**
BMI	-0.02	0.82	0.03	0.71
Waist-Hip Ratio	0.17	0.02	0.14	0.05
Hs-CRP	0.17	0.02	0.1	0.18
Systolic BP	0.17	0.03	0.12	0.10
Diastolic BP	0.23	0.002	0.18	0.08
HbA1C	0.17	0.02	0.15	0.03
Total Cholesterol	-0.06	0.39	- 0.07	0.33
TG/ HDL-c	-0.14	0.06	-0.06	0.44
HOMA-IR	- 0.03	0.69	-0.02	0.78
PTH	-0.23	0.002	-0.26	< 0.001
Calcium	-0.17	0.03	-0.13	0.05
e-GFR	- 0.28	< 0.001	-0.15	0.09

Beta derived from linear regression analysis, before and after adjustment for age and sex.
BMI: Body Mass Index; Hs-CRP: high sensitivity CRP; BP: blood pressure; Hb A1c: haemoglobin A1c; TG: triglycerides; HDL-c: high density lipoprotein cholesterol; HOMA-IR: homeostasis model assessment of insulin resistance index; PTH: parathyroid hormone; e-GFR: estimated glomerular filtration rate.

Table 4. Linear regression coefficients of serum OPG in indicating the severity of coronary heart disease according to coronary calcium score (CCS), number of involved vessels, and Gensini score)

Serum OPG	**Coronary calcium score**		**Number of involved vessels**		**Gensini Score**	
	Beta	**PV**	**Beta**	**PV**	**Beta**	**PV**
Unadjusted	0.29	0.001	0.35	< 0.001	0.18	0.04
Model 1	0.20	0.03	0.27	0.003	0.11	0.22
Model 2	0.32	0.01	0.37	0.001	0.18	0.05
Model 3	0.30	0.004	0.36	0.001	0.21	0.02
Model 4	0.27	0.02	0.29	0.004	0.12	0.19

Beta, Standardized coefficient derived from linear regression analysis with stepwise adjustments.
Model 1: after adjustment for age and sex; Model 2: after adjustment for diabetes, systolic and diastolic BP; Model 3: after adjustment for markers of bone calcium metabolism (Calcium, Phosphorus, PTH, Vit D); Model 4: Model 1 plus Model 2 plus Model 3.

Results

Characteristics of the study population with respect to diabetes status

Of 177 patients enrolled in this study, 22 met our exclusion criteria. From a total of 155 patients, 98 (63.2%) underwent coronary angiography (67 non diabetic, 31 diabetic) due to their symptoms and clinical indications. Demographics of the study population are presented in Table 1. Table 2 represents the clinical and laboratory findings of CHD patients with respect to their diabetes status. BMI, waist circumference, waist to hip ratio, as well as mean fasting blood sugar, serum triglyceride, HbA1c, HOMA-IR, serum OPG, PTH, and the Gensini score were significantly higher in CHD patients with diabetes. In all, no significant sex difference was observed in serum OPG level.

Binary logistic regression analysis revealed a significant association between diabetes and serum OPG with the β coefficient of 0.38 (OR (95% CI): 1.46 (1.11 – 1.91) which remained significant after age and sex adjustment 0.42 (OR (95% CI): 1.52 (1.12 – 2.06), P < 0.01]. However, after adjusting for further variables, including markers of bone calcium metabolism (calcium, phosphorus, Vitamin D, PTH), this association lost its statistical significance (OR (95% CI): 1.36 (0.98 – 1.90), P > 0.05).

Association of serum OPG with demographic and metabolic variables

Serum OPG was positively associated with age, waist to hip ratio, systolic and diastolic BP, hs-CRP, HbA1c and inversely associated with e-GFR, PTH, and calcium. There was no significant association between serum lipid profile, HOMA-IR, Vitamin D, LVEF and OPG concentration. Since age was positively associated with serum OPG (P < 0.001), the age and sex adjusted β coefficients of each variable were determined. After age-sex adjustment, no significant association was found except for HbA1c and PTH (Table 3).

Table 4 shows that serum OPG was significantly associated with the severity of CHD in terms of the CCS, the number of involved

Table 5. Linear regression coefficients of individual components and total Framingham point score as determinants of serum OPG concentration

Framingham risk score predictors	Female				Male			
	Unadjusted		Adjusted*		Unadjusted		Adjusted*	
	Beta	PV	Beta	PV	Beta	PV	Beta	PV
Age Score	0.36	0.02	0.26	0.06	0.24	0.01	0.24	0.02
Diabetes	0.21	0.14	0.01	0.89	0.23	0.009	0.2	0.03
Total Cholesterol/ HDL Score	0.13	0.33	0.17	0.26	- 0.14	0.13	- 0.15	0.14
Systolic Blood Pressure Score ≠	0.25	0.04	0.23	0.05	- - -	- - -	- - -	- - -
Smoking ≠a	- - -	- - -	- - -	- - -	- - -	- - -	- - -	- - -
Total Framingham point Score	0.50	0.003	0.39	0.007	0.12	0.26	0.08	0.45
2 yr risk of subsequent CHD (%)	0.48	0.005	0.37	0.01	0.08	0.45	0.03	0.8

Beta derived from linear regression analysis before and after adjustments for markers of bone metabolism.
*: Adjusted for markers of bone metabolism (Calcium, phosphorus, PTH, vitamin D); ≠: SBP and smoking are risk predictors in females; a: none of the enrolled females report a history of smoking in our study.

vessels and the Gensini score. These associations remained statistically significant after adjustment for markers of bone calcium metabolisms, diabetes, systolic and diastolic BP (model 2, model 3). In the age and sex adjusted model, OPG was associated with CCS, and the number of involved vessels but not the Gensini score.

Risk Appraisal for Subsequent CHD

Table 5 demonstrates the association between serum OPG and individual components, total point score, and risk of subsequent CHD based on the computational Framingham risk prediction model. Compared with men, serum OPG showed a significant positive association with total Framingham point score and the 2-year risk of subsequent CHD in females. This association was attenuated but remained significant after adjusting for the confounding effect of bone-calcium metabolism markers.

Discussion

According to a number of studies, serum OPG level is elevated in subjects with CHD and in subjects with diabetes, independent of the conventional risk factors.[1,2,7–10,21]

Our finding of a higher serum OPG concentration in CHD patients with diabetes compared to the patients without diabetes indicates the cumulative pattern of increasing OPG in these groups. Meanwhile, we found a significant association between diabetes status and serum OPG concentration; this association remained significant after adjusting for age, sex and bone metabolism markers including calcium, phosphorus and vitamin D but it vanished when PTH was added to the regression.

Previous studies on asymptomatic diabetic patients have mainly demonstrated the association between serum OPG and the presence of CHD. They proposed an independent predictive value of serum OPG for significant CHD.[2,3,21] Besides, our findings are in line with those from other studies showing that serum OPG concentration was associated with the severity of angiographic findings in symptomatic CHD patients.[1, 26,27]

In our study, consistent with other reports, OPG levels increased with age[6,27] and were significantly associated with hs-CRP,[27] systolic[6,27] and diastolic BP, HbA1c,[6,27] diabetes, e-GFR as well as PTH and calcium. After age and sex adjustment, these associations disappeared except for the diabetes, HbA1c and PTH, suggesting that the status of age and sex does not provide justification for these links.

Clinical studies have revealed a strong positive association between serum OPG and advanced CHD.[1,5,21,26] Our study revealed a strong and independent association between CCS, the number of stenotic vessels and serum OPG, This remains significant after multiple adjustments for possible confounders, including age, sex, diabetes, systolic and diastolic blood pressure, and markers of bone calcium metabolism. The distinctive point of this study is considering the hidden influence of bone metabolism markers which was not assessed in the previous studies.

Association of serum OPG with severity of CHD did not vanish even when adjusted for the markers of bone metabolism. Thus, we propose an independent role for OPG in connection with the severity of CHD; a connection which is not modulated through the axis of bone metabolism.

We also found a significant association between the Gensini score and serum OPG, which remained significant after adjustment for diabetes, blood pressure and markers of bone calcium metabolism, but disappeared when adjusting for age and sex.

Yet, there are limited published studies to determine the value of this marker in predicting cardiovascular mortality. A recent study stated that a two-fold increase in serum OPG significantly predicted the combined end-point hospitalization of CHD, ischemic stroke and all-cause mortality.[12] The combination of OPG, Hs-CRP and traditional cardiovascular risk factors improves risk detection.[12] A 17-year prospective observational follow-up study declared that elevated serum OPG is strongly predictive of all cause mortality in diabetic patients independent of conventional cardiovascular risk factors.[28] Another study indicated serum OPG as a predictor of long term mortality and heart failure development in patients with Acute Coronary Syndrome (ACS).[29] Of note, the outcome in the aforementioned studies was all-cause mortality. They suggested further studies to indicate the association between OPG and cardiovascular mortality.[28,29] Altogether, the effect of serum OPG in predicting the risk of recurrent CHD was not assessed before.

The Framingham risk appraisal model includes age, gender,

total and HDL cholesterol, diabetes, systolic blood pressure and cigarette smoking to derive an estimated risk of recurring CHD within two years. As mentioned, risk factor scoring for men did not include SBP or cigarette smoking.[15] Using the computational Framingham risk prediction model, our data demonstrate a significant positive association between serum OPG and two year risk of recurrent CHD in females which was not confounded by the markers of bone calcium metabolism. Although serum OPG was associated with age and diabetes score in males, no significant association was detected between the Framingham risk score of subsequent CHD and serum OPG in this group. Thus, we propose the interaction of gender in OPG association and prediction of subsequent CHD. Only in females, increased serum OPG was the indicator of CHD recurrence.

With regard to the cross sectional nature of our study, the temporal effect of OPG on the recurrence risk of coronary event cannot be shown. Using the Framingham scoring system to estimate the prospective risk of recurrent CHD has also some limitations. First, important prognostic information, such as angiographic findings or cardiac imaging, is not included in this model. Second, the risk factors may be changed as a result of an interim myocardial infarction.[15,16] Future pathophysiological studies should unveil the axis which connects serum OPG levels with CHD. Moreover, translational researches would recognize the effect modifying role of gender in OPG – CHD association.

Serum OPG independently indicates the severity and extent of coronary artery involvement in CHD patients, as well as a higher risk of subsequent CHD in females. As a clinical implication, serum OPG is suggested as a potential marker in assessing the risk of subsequent CHD, specifically in females.

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Conflict of Interest: *The authors declare that there are no conflicts of interest.*

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