

CLINICAL STUDY

Serum osteocalcin concentrations in relation to glucose and lipid metabolism in Chinese individuals

Mi Zhou^{1,2}, Xiaojing Ma¹, Huating Li¹, Xiaoping Pan¹, Junling Tang¹, Yunchao Gao³, Xuhong Hou¹, Huijuan Lu¹, Yuqian Bao¹ and Weiping Jia¹

¹Shanghai Key Laboratory of Diabetes Mellitus, Department of Endocrinology and Metabolism, Shanghai Diabetes Institute, Shanghai Clinical Center for Diabetes, Shanghai Jiao Tong University Affiliated Sixth People's Hospital, 600 Yishan Road, Shanghai 200233, People's Republic of China, ²Department of Medicine, Medical School of Soochow University, Suzhou 215006, People's Republic of China and ³The Immunoassay Laboratory, Department of Nuclear Medicine, Shanghai Jiao Tong University Affiliated Sixth People's Hospital, Shanghai 200233, People's Republic of China

(Correspondence should be addressed to Y Bao; Email: byq522@163.com)

(M Zhou and X Ma contributed equally to this work)

Abstract

Objectives: Osteocalcin, a bone-derived protein, has recently been reported to affect energy metabolism. We investigated the relationship between serum osteocalcin and parameters of adiposity, glucose tolerance, and lipid profile in Chinese subjects.

Methods: Serum osteocalcin was measured by electrochemiluminescence immunoassay in 254 men (128 with newly diagnosed type 2 diabetes mellitus (T2DM) and 126 with normal glucose tolerance (NGT)), 66 premenopausal women (33 with T2DM and 33 with NGT) as well as 180 postmenopausal women (92 with T2DM and 88 with NGT). Their associations with parameters of adiposity, glucose tolerance, and lipid profile were examined.

Results: Serum osteocalcin concentrations in diabetic patients were significantly lower than those in NGT subjects after adjusted for age, gender, and body mass index ($P=0.003$). Postmenopausal women had higher osteocalcin concentrations than premenopausal women and men (both $P<0.001$). Multiple stepwise regression analysis showed that age, %fat, high-density lipoprotein cholesterol, fasting plasma glucose, and fasting serum insulin were independently associated with osteocalcin in men ($P<0.05$). Age and HbA1c were independently correlated with osteocalcin in postmenopausal women. Besides age and HbA1c, serum triglyceride was also an independent factor influencing osteocalcin in premenopausal women. In addition, osteocalcin was also positively associated with homeostasis model assessment of β -cell function. Furthermore, multiple logistic regression analysis demonstrated that osteocalcin was independently associated with T2DM.

Conclusions: Serum osteocalcin was closely associated with not only fat and glucose metabolism but also with lipid metabolism.

European Journal of Endocrinology 161 723–729

Introduction

Recent studies have verified that adipose tissue could regulate bone remodeling through the adipokine leptin by acting on osteoblasts (1, 2). In turn, bone modulates energy metabolism in a feedback loop. The novel function for the skeleton unraveled its importance as an endocrine organ. A bone-derived protein, osteocalcin, has raised much attention as a hormone regulating glucose metabolism and fat mass. Osteocalcin is an osteoblast-specific secreted molecule. It has been observed as a constituent of the bone extracellular matrix. It is also secreted in the general circulation and has some other features of a hormone. Animal experiments and cell-based assay suggested that osteocalcin can increase β -cell proliferation, insulin secretion, and insulin sensitivity by increasing

adiponectin gene expression (3). Lee *et al.* showed that osteocalcin knockout mice (*Ocn*^{-/-}) displayed higher blood glucose level and lower serum insulin level than wild-type mice (3). Furthermore, osteocalcin treatment weakened the development of obesity and diabetes in wild-type mice (4).

Although these animal-based studies are certainly of interest, the clinical relevance of osteocalcin has seldom been explored so far. Recently, Fernandez-Real *et al.* demonstrated that serum osteocalcin was associated with insulin sensitivity and insulin secretion in nondiabetic subjects (5). Kanazawa *et al.* showed correlations of serum osteocalcin with blood glucose and fat mass in Japanese patients with type 2 diabetes mellitus (T2DM) (6). However, the associations of serum osteocalcin with obesity, glucose, and lipid metabolism remained unknown in the Chinese population.

The purpose of this study was to investigate the role of osteocalcin in glucose tolerance, lipid profile, insulin secretion, and sensitivity in Chinese individuals.

Methods

Subjects

This study was part of the epidemiological survey for diabetes and metabolic syndrome in six communities from May 2007 to August 2008 in Shanghai, China. All participants were expected to complete a uniform questionnaire containing questions about the histories of present and past illness and medical therapy. In the present study, we selected 253 patients with newly diagnosed T2DM (128 men, 33 premenopausal women, and 92 postmenopausal women), and 247 normal glucose tolerance (NGT) subjects (126 men, 33 premenopausal women, and 88 postmenopausal women) matched for age, gender, and body mass index (BMI). The age range for men, premenopausal women, and postmenopausal women were 23–76, 23–53, and 44–76 years respectively. Diabetes was diagnosed according to the 1999 World Health Organization criteria (7). None of the subjects had taken any hypoglycemic medications. Complete physical examinations and routine biochemical analyses of blood were performed. The study was approved by the ethics committee of the hospital. Written informed consent was obtained from all participants.

Participants with the following conditions were excluded: hepatic or renal dysfunction, nutritional derangements, anemia, history of malignancy, hyperparathyroidism or thyroid dysfunction, pregnancy, and fracture within 1 year or taking medications known to influence bone and calcium metabolism, such as vitamin D, bisphosphonate, calcitonin, estrogen, tamoxifen, or corticosteroids.

Anthropometric evaluation

Participants arrived at the community service center at 0600 h following an overnight fasting of 10 h. A physical examination, including the measurement of height, weight, waist circumference, blood pressure (BP), and body fat, was performed for each subject. BMI was calculated as weight divided by squared height. Body fat percentage (%fat) was estimated by the TBF-410 Tanita Body Composition Analyzer (Tanita, Tokyo, Japan). Waist circumference was measured midway between the lowest rib and the superior border of iliac crest on midaxillary line. Hip circumference was measured around the buttocks at the level that yielded the maximum measurement. Waist-to-hip ratio (WHR) was also calculated as the ratio of the waist circumference to the hip circumference. BP value was the average of three time measurements using a sphygmomanometer with the interval of 2 min.

Biochemical analyses

Blood samples were collected after an overnight fast. Each participant received a standard 75 g oral glucose tolerance test (OGTT) (8). Fasting plasma glucose (FPG) and 2-h post-OGTT plasma glucose (2hPG) were assayed by glucose oxidase method. Serum insulin concentration was measured by RIA (Linco Research, St Charles, Missouri, USA). HbA1c was determined by HPLC (Bio-Rad Inc). Serum triglyceride (TG), total cholesterol (TC), high-density lipoprotein cholesterol (HDL-c), and low-density lipoprotein cholesterol (LDL-c) were measured by enzymatic procedures using an autoanalyzer (Hitachi 7600-020, automatic analyzer). Serum osteocalcin was measured by electrochemiluminescence immunoassay (Roche Diagnostics GmbH) with intra and interassay coefficients of variation of 1.2–4.0 and 1.7–6.5% respectively.

Homeostasis model assessment

Insulin sensitivity was estimated by homeostasis model assessment-insulin resistance (HOMA-IR) based on fasting glucose and insulin measurements as follows: $HOMA-IR = \text{fasting serum insulin (FINS)} (mU/l) \times \text{FPG} (mmol/l) / 22.5$. Basal insulin secretion was assessed by HOMA-%B, which was calculated as $(FINS (mU/l) \times 6 - 3.33) / (\text{FPG} (mmol/l) - 3.5)$ (9).

Statistical analyses

All analyses were performed with Statistical Package for Social Sciences version 13.0 (SPSS, Chicago, IL, USA). Data were expressed as mean \pm s.d. except for skewed variables, which were presented as median (interquartile range 25–75%). Clinical characteristics that followed a normal distribution were compared among the three groups using one-way ANOVA test, and those that were not normally distributed were compared with Kruskal–Wallis test. Unpaired Student's *t* test was used for comparisons between subjects with NGT and T2DM for normally distributed parameters in each group, while the Wilcoxon rank-sum test was applied for variables with a skewed distribution. Serum osteocalcin concentrations were compared for each group using an analysis of covariance. The Spearman correlation coefficients were calculated to assess the strength of the correlations of osteocalcin and parameters of adiposity, glucose, and lipid metabolism. Multiple stepwise regression analysis was performed to determine the associations between serum osteocalcin and metabolic parameters after adjusting for potential confounders. Multivariate logistic regression analysis was conducted using the existence of T2DM as a dependent variable. All reported *P* values were two-tailed, and *P* values < 0.05 were considered statistically significant.

Table 1 Demographic and clinical characteristics of the study subjects.

Parameters	Men				Premenopausal women				Postmenopausal women									
	Total (n=254)		NGT (n=126)		T2DM (n=128)		Total (n=66)		NGT (n=33)		T2DM (n=33)		Total (n=180)		NGT (n=88)		T2DM (n=92)	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Age (years)	53.0±12.4		52.9±12.4		53.1±12.4		43.2±5.8*		43.9±5.0		42.6±6.5		61.5±7.2*†		61.8±7.0		61.2±7.4	
Height (cm)	168.7±6.5		168.5±6.4		168.9±6.6		157.2±9.6*		158.6±5.2		155.9±12.4		155.8±5.1*		156.3±4.8		155.3±5.3	
Weight (kg)	73.6±11.1		72.5±10.8		74.9±11.4		60.1±11.6*		60.7±10.5		59.5±12.8		61.5±9.0*		61.9±8.6		61.2±9.3	
BMI (kg/m ²)	25.9±3.5		25.5±3.1		26.3±3.9		24.2±3.8*		24.1±3.6		24.4±4.2		25.3±3.4		25.3±3.4		25.3±3.4	
W (cm)	88.5±9.2		87.0±8.4		90.0±9.6†		78.0±10.4*		77.5±9.8		78.6±11.2		83.5±8.5*†		83.2±8.7		83.9±8.3	
WHR	0.90±0.06		0.90±0.05		0.91±0.06†		0.84±0.06*		0.83±0.06		0.84±0.07		0.87±0.06*†		0.86±0.06		0.87±0.06	
%fat (%)	26.0±6.5		25.2±5.5		26.8±7.2§		33.4±8.4*		32.7±7.5		34.1±9.2		35.3±6.9*		35.0±7.0		35.6±6.8	
SBP (mmHg)	130 (120–140)		129 (116–140)		130 (120–140)		120 (110–130)*		115 (110–123)		120 (110–132)		130 (120–145)†		130 (120–140)		130 (121–150)	
DBP (mmHg)	80 (75–90)		80 (74–90)		81 (77–90)		80 (70–80)*		78 (70–80)		80 (70–86)		80 (74–89)*		80 (72–88)		80 (74–89)	
TC (mmol/l)	4.8±0.9		4.6±0.8		4.8±0.9§		4.5±0.9†		4.4±0.7		4.6±1.0		5.2±0.9*†		5.2±0.9		5.3±0.9	
TG (mmol/l)	1.7 (1.2–2.6)		1.6 (1.1–2.4)		1.8 (1.3–3.3)†		1.2 (0.8–2.0)*		1.0 (0.7–1.9)		1.3 (0.9–2.3)		1.7 (1.3–2.4)†		1.6 (1.1–2.1)		1.9 (1.3–2.9)†	
HDL-c (mmol/l)	1.1 (1.0–1.3)		1.1 (1.0–1.4)		1.1 (1.0–1.3)		1.3 (1.1–1.5)*		1.4 (1.3–1.6)		1.2 (1.1–1.4)		1.3 (1.1–1.5)*		1.4 (1.1–1.5)		1.3 (1.1–1.5)	
LDL-c (mmol/l)	3.1±0.8		3.0±0.7		3.1±0.8		2.8±0.7†		2.7±0.6		2.9±0.9		3.3±0.8*†		3.3±0.7		3.4±0.8	
FPG (mmol/l)	5.6 (5.1–7.1)		5.1 (4.8–5.4)		7.1 (6.1–8.3)†		5.7 (5.2–7.3)		5.2 (4.7–5.4)		7.3 (6.6–8.2)†		5.6 (5.1–7.1)		5.2 (4.6–5.5)		7.1 (6.1–7.7)†	
2hPG (mmol/l)	7.4 (5.6–12.9)		5.8 (4.8–6.6)		12.9 (11.4–15.7)†		6.5 (5.1–11.2)		5.2 (4.4–6.2)		11.2 (7.5–13.1)†		7.4 (6.1–12.2)		6.1 (5.2–6.7)		12.2 (11.3–14.4)†	
HbA1c (%)	5.8 (5.5–6.5)		5.6 (5.4–5.8)		6.5 (5.9–7.1)†		5.7 (5.4–6.3)		5.5 (5.1–5.7)		6.3 (5.7–6.8)†		6.0 (5.6–6.5)		5.7 (5.5–5.9)		6.5 (6.1–7.1)†	
FINS (mU/l)	17.5		20.0		16.4		19.2		19.2		19.6		20.0		22.7		18.6 (13.2–27.9)	
	(13.3–24.3)		(15.4–26.4)		(12.7–21.7)†		(14.4–24.5)		(14.6–24.5)		(13.4–25.1)		(14.7–24.4)*		(15.9–28.2)			
HOMA-IR	4.8 (3.5–6.6)		4.5 (3.3–6.0)		5.2 (3.6–7.8)†		5.5 (3.6–7.1)		4.2 (3.3–5.7)		6.0 (4.4–7.6)§		5.4 (3.7–7.4)¶		5.1 (3.6–6.6)		6.1 (4.0–8.8)†	
HOMA-%B	48.5		73.0		26.7		45.5		77.4		24.7		61.9		80.6		33.1 (20.0–64.7)†	
	(25.8–77.9)		(55.5–98.3)		(17.4–40.7)†		(24.5–79.3)		(56.4–95.9)		(16.0–43.6)†		(31.4–92.1)*		(60.9–123.6)			

Data represent means±s.d. or median (interquartile range). BMI, body mass index; W, waist circumference; WHR, waist-to-hip ratio; SBP, systolic blood pressure; DBP, diastolic blood pressure; FINS, fasting serum insulin (normal range 3.42–16.44 mU/l); FPG, fasting plasma glucose (normal range 3.9–5.8 mmol/l); 2hPG, 2-h post-OGTT plasma glucose (normal range 3.9–7.8 mmol/l); HDL-c, high-density lipoprotein cholesterol (normal range 0.90–1.68 mmol/l); LDL-c, low-density lipoprotein cholesterol (normal range 2.84–4.10 mmol/l); TC, total cholesterol (normal range 2.80–5.90 mmol/l); TG, triglyceride (normal range 0.45–1.81 mmol/l). *P≤0.01 versus men; †P<0.01 versus premenopausal women; ‡P<0.01 versus NGT; §P<0.05 versus premenopausal women; ¶P<0.05 versus men.

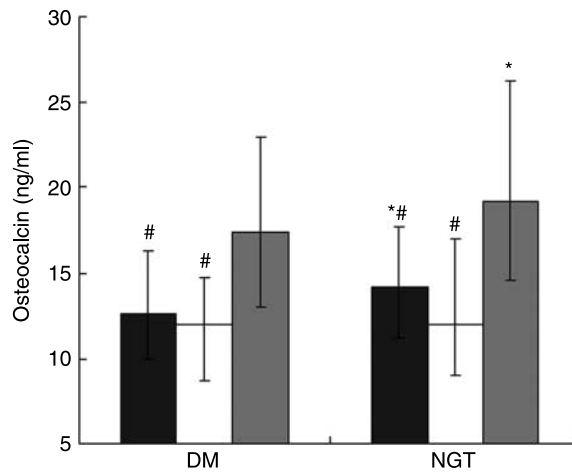


Figure 1 Serum osteocalcin concentrations in subjects with T2DM and NGT. Median (interquartile range) levels of serum osteocalcin in men, premenopausal women, and postmenopausal women are displayed. Black, men; white, premenopausal women; grey, postmenopausal women. * $P < 0.05$ versus DM after age and BMI adjustment; # $P < 0.001$ versus postmenopausal women after age and BMI adjustment.

Results

Characteristics of subjects

The principal characteristics of the study population are displayed in Table 1. Age, height, weight, waist circumference, WHR, % fat, BP, and lipid profile differed significantly among the three groups ($P < 0.01$). Men had significantly higher height, weight, waist circumference, WHR, diastolic BP and lower %fat, and HDL-c level than both premenopausal and postmenopausal women. There were no differences in the glucose tolerance status (FPG, 2hPG, HbA1c) among the three groups.

Serum osteocalcin concentrations in NGT and T2DM subjects

Serum concentrations of osteocalcin ranged from 4.6 to 39.5 ng/ml. We found a decreasing trend in osteocalcin concentrations with age in all three groups after we stratified subjects by every 10 years (a median level of 19.6–11.4 ng/ml in men, 13.5–10.7 ng/ml in premenopausal women, and 20.2–16.5 ng/ml in postmenopausal women, all $P < 0.05$). After adjustment for age, gender, and BMI, serum osteocalcin concentrations in the subjects with T2DM (median 15.1 ng/ml (interquartile range 10.8–18.3)) were significantly lower in comparison with the subjects with NGT (16.8 ng/ml (11.8–20.6); $P = 0.003$). Postmenopausal women had significantly higher serum osteocalcin concentrations than premenopausal women and men after adjustment for age and BMI in both NGT subjects (19.2 ng/ml (14.6–26.3) versus 12.0 ng/ml (9.0–17.0) and 14.2 ng/ml (11.2–17.7), $P < 0.001$) and diabetic patients (17.4 ng/ml (13.0–23.0) versus 12.0 ng/ml (8.7–14.7) and 12.6 ng/ml (10.0–16.3), $P < 0.001$; Fig. 1).

Correlations of serum osteocalcin with anthropometric parameters, glucose, lipid profile, insulin secretion, and insulin sensitivity

In whole participants, the correlation analysis showed that serum osteocalcin was inversely correlated with FPG ($r = -0.142$, $P = 0.001$), 2hPG ($r = -0.089$, $P = 0.048$), and HbA1c ($r = -0.167$, $P < 0.001$), and positively correlated with TG ($r = 0.095$, $P = 0.035$), FINS ($r = 0.144$, $P = 0.001$), and HOMA-%B ($r = 0.216$, $P < 0.001$). However, no significant correlation was found between serum osteocalcin and HOMA-IR ($r = 0.031$, $P = 0.492$).

To further determine which variables were independently associated with serum osteocalcin, multiple stepwise regression analysis was performed in men and women separately (Table 2). As a result, age, %fat,

Table 2 Multiple stepwise regression analysis showing variables independently associated with serum osteocalcin in men and women respectively.

Independent variables enter the model		β	S.E.M.	Standardized β	t	P
Men	Age	-0.003	0.001	-0.248	-4.038	<0.001
	%fat	-0.004	0.002	-0.175	-2.638	0.009
	HDL-c	-0.250	0.103	-0.153	-2.421	0.016
	FPG	-0.010	0.005	-0.130	-2.086	0.038
	FINS	0.107	0.047	0.144	2.283	0.023
Premenopausal women	Age	-0.008	0.003	-0.271	-2.300	0.025
	TG	0.167	0.081	0.251	2.054	0.044
	HbA1c	-0.052	0.019	-0.330	-2.703	0.009
Postmenopausal women	Age	-0.003	0.002	-0.152	-2.094	0.038
	HbA1c	-0.041	0.010	-0.296	-4.074	<0.001

Age, BMI, waist circumference, WHR, %fat, TC, TG, LDL-c, HDL-c, FPG, 2hPG, HbA1c, and FINS were selected as independent variables. FINS, fasting serum insulin; FPG, fasting plasma glucose; HbA1c; HDL-c, high-density lipoprotein cholesterol; TG, triglyceride.

Table 3 Multiple stepwise regression analysis to assess the relationship between osteocalcin and insulin secretion.

Independent variables enter the model		β	S.E.M.	Standardized β	t	P
HOMA-%B	Age	0.004	0.001	0.145	3.753	<0.001
	BMI	0.015	0.004	0.152	3.880	<0.001
	HbA1c	-0.149	0.012	-0.501	-12.577	<0.001
	Osteocalcin	0.289	0.081	0.139	3.551	<0.001

Age, gender, BMI, waist circumference, WHR, %fat, lipid profile, HbA1c, and osteocalcin were selected as independent variables. BMI, body mass index.

HDL-c, FPG, and FINS were independently associated with osteocalcin in men. Besides age and HbA1c, which were also independently correlated with osteocalcin in postmenopausal women, TG was shown to be an independent factor influencing osteocalcin in premenopausal women. Moreover, in order to eliminate the effects of lipid-lowering agents such as statins or fibrates, we exclude subjects who had ever taken these drugs (six men, two premenopausal women, and 12 postmenopausal women). Exclusion of this group made little difference to the regression coefficients of lipid parameters in the analyses above ($\beta = -0.270$, $P = 0.012$ for HDL-C in men, and $\beta = 0.170$, $P = 0.043$ for TG in premenopausal women; data not shown).

In addition, as shown in Table 3, to assess the influence of osteocalcin on insulin secretion, multiple regression model with HOMA-%B as a dependent variable further revealed that serum osteocalcin was an independent factor associated with HOMA-%B. However, when HOMA-IR was the dependent variable, osteocalcin was not a significant independent determinant, which was excluded in the final model.

Multiple logistic regression analysis

In all subjects, multiple logistic regression analysis was performed using the presence of T2DM as a dependent variable (Table 4). The analysis involved age, gender, BMI, %fat, BP, serum osteocalcin, and lipid profile (TC, TG, LDL-c, HDL-c). As a result, osteocalcin (odds ratio, OR = 0.136), systolic BP (OR = 1.016), and TG (OR = 5.203) were found to be independent factors for diabetes. However, other variables were all excluded in the final model.

Discussion

Osteocalcin is a 49-amino acid bone matrix noncollagen protein, which is one of the most mature secretory products of osteoblasts (10). Serum osteocalcin concentration has been found to represent the *de novo* synthesis of osteocalcin by osteoblasts (11), and is a specific biochemical marker of bone turnover and bone formation (12, 13). Besides its functions in bone mineralization and calcium homeostasis, recent animal studies have reported its novel role in the regulation of glucose metabolism and insulin secretion and sensitivity. However, relevant evidences are still limited in humans.

In animal studies, as a model of a gain of osteocalcin bioactivity, *Esp*^{-/-} mice were protected from obesity; in contrast, *Ocn*^{-/-} mice showed an increase in fat mass and adipocyte number (3). Consistent with these studies, we found %fat was negatively correlated with serum osteocalcin in men. In addition, similarly to previous clinical studies (14–17), we observed significantly lower osteocalcin concentrations in patients with T2DM compared with NGT subjects. Moreover, we found FPG was independently correlated with osteocalcin in men, and HbA1c was independently correlated with osteocalcin in both premenopausal and postmenopausal women. Studies *in vivo* or *in vitro* have demonstrated that in hyperglycemic states, the osteoblast mass and possibly function are decreased, which suppresses osteocalcin synthesis and secretion (18, 19), resulting in impaired bone turnover and reduced bone formation (20). Conversely, osteocalcin also has an impact on glucose regulation. In line with observations from animal studies, which showed that recombinant osteocalcin could increase insulin secretion (3, 4), we found osteocalcin was positively associated with basal insulin secretion.

Table 4 Multivariate logistic regression analysis with type 2 diabetes mellitus as a dependent variable.

	B	S.E.M.	Wald	P	Exp(B)	95% CI	
Osteocalcin	-1.996	0.579	11.883	0.001	0.136	0.044	0.423
Systolic BP	0.016	0.008	3.922	0.048	1.016	1.000	1.033
TG	1.649	0.686	5.780	0.016	5.203	1.356	19.964

The analysis also included age, gender, BMI, %fat, diastolic blood pressure, TC, LDL-c, and HDL-c, which were all excluded in the final model. BP, blood pressure; TG, triglyceride.

A novel observation of this study was the relationship of serum osteocalcin with lipid profile in both genders: negatively for HDL-c in men and positively for TG in premenopausal women. Although recent animal-based studies suggested osteocalcin might have a beneficial effect on blood TG levels (3, 4), the clinical relevance of osteocalcin with lipid profile remained poorly characterized. Fernandez-Real *et al.* studied 19 obese non-diabetic women in Spain, aged 40–60 years. Their baseline serum osteocalcin was not associated with TG. After diet or diet plus resistance training intervention, the postintervention serum osteocalcin was negatively associated with TG ($r = -0.54$, $P = 0.01$). However, they did not observe the relationship between the change in osteocalcin and the change in TG (5). Another study reported that no correlations between osteocalcin and lipid profile were found in 339 postmenopausal women from Korea (21), which was in line with our findings. The discrepancy from the above studies raised the possibility that the effect of osteocalcin on lipid metabolism in humans might be different from that in animals. The pathophysiologic mechanisms underlying this association are unknown. Although relevant data about the relationship between osteocalcin and lipid profile are scanty, there is increasing evidence showing a connection between lipids and bone metabolism in recent years. A number of studies focusing on the association between lipid profile and bone mass density (BMD) suggested that TG was positively associated with BMD and inversely associated with the presence of vertebral fractures (22–24). Adami *et al.* also found reduced bone mass to be associated with a favorable lipid profile in healthy men (25). However, the literature concerning relationships between HDL-c levels and BMD was inconsistent. Studies in both men and postmenopausal women found an inverse relationship between fasting HDL-c level and BMD (26, 27), while a study in older Japanese women reported a positive relationship (24). These observations did imply that there might be some common mechanism underlying both lipid and bone metabolism. Kha *et al.* demonstrated that certain oxysterols displayed osteogenic activities by regulating the differentiation of mesenchymal cells into osteogenic cells (28). We speculate that as a marker of bone turnover and bone formation, osteocalcin might be the possible link between dyslipidemia and BMD. However, we did not measure BMD, which might have provided more insight into the association between osteocalcin and lipid metabolism. Clearly, more studies are warranted to clarify the pathophysiologic relationship between osteocalcin and lipid profile.

In this study, we did not find the correlations between serum osteocalcin and insulin resistance as several studies reported (5, 21, 29). Previous multiethnic cohort studies have reported the ethnic differences in insulin sensitivity and β -cell function (30). In the present study, the BMI of the subjects was relatively

low in comparison with those in above studies, which may contribute to insignificant correlation between osteocalcin and HOMA-IR. Besides that, more precise evaluations of insulin resistance like glucose clamp are still warranted to investigate the association between osteocalcin and insulin resistance.

There are several limitations in this study. First, the sample size was not large enough, especially premenopausal women. Secondly, due to a design to compare serum osteocalcin concentrations between subjects with NGT and T2DM, we did not include patients with impaired glucose regulation, i.e. impaired fasting glucose (IFG) and/or impaired glucose tolerance (IGT). However, as IFG and IGT are two important categories of prediabetes, which represent an intermediate stage between NGT and DM, further studies are needed in this population. Furthermore, because of its cross-sectional nature, we are not able to clarify the cause-effect between osteocalcin and lipid metabolism, and relevant prospective researches are warranted in future.

In conclusion, we investigated the association between osteocalcin and factors related to adiposity, glucose, and lipid metabolism in Chinese individuals. Serum osteocalcin concentrations were positively associated with insulin secretion and inversely correlated with glucose levels and adiposity. Moreover, for the first time, we found an association between serum osteocalcin and lipid metabolism in the Chinese population. These conclusions supported the recent concept that as an endocrine organ bone has close crosstalks with energy metabolism and might contribute to the onset and severity of metabolic disorders.

Declaration of interest

We declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

Funding

This work was funded by Chinese National 973 Project (2007CB914702).

References

- 1 Ducey P, Amling M, Takeda S, Priemel M, Schilling AF, Beil FT, Shen J, Vinson C, Rueger JM & Karsenty G. Leptin inhibits bone formation through a hypothalamic relay: a central control of bone mass. *Cell* 2000 **100** 197–207.
- 2 Eleftheriou F, Ahn JD, Takeda S, Starbuck M, Yang X, Liu X, Kondo H, Richards WG, Bannion TW, Noda M, Clement K, Vaisse C & Karsenty G. Leptin regulation of bone resorption by the sympathetic nervous system and CART. *Nature* 2005 **434** 514–520.
- 3 Lee NK, Sowa H, Hinoi E, Ferron M, Ahn JD, Confavreux C, Dacquin R, Mee PJ, McKee MD, Jung DY, Zhang Z, Kim JK, Mauvais-Jarvis F, Ducey P & Karsenty G. Endocrine regulation of energy metabolism by the skeleton. *Cell* 2007 **130** 456–469.
- 4 Ferron M, Hinoi E, Karsenty G & Ducey P. Osteocalcin differentially regulates beta cell and adipocyte gene expression and affects the development of metabolic diseases in wild-type mice. *PNAS* 2008 **105** 5266–5270.

- 5 Fernández-Real JM, Izquierdo M, Ortega F, Gorostiaga E, Gómez-Ambrosi J, Moreno-Navarrete JM, Frühbeck G, Martínez C, Idoate F, Salvador J, Forga L, Ricart W & Ibañez J. The relationship of serum osteocalcin concentration to insulin secretion, sensitivity, and disposal with hypocaloric diet and resistance training. *Journal of Clinical Endocrinology and Metabolism* 2009 **94** 237–245.
- 6 Kanazawa I, Yamaguchi T, Yamamoto M, Yamauchi M, Kurioka S, Yano S & Sugimoto T. Serum osteocalcin level is associated with glucose metabolism and atherosclerosis parameters in type 2 diabetes mellitus. *Journal of Clinical Endocrinology and Metabolism* 2009 **94** 45–49.
- 7 World Health Organization. Definition, diagnosis and classification of diabetes mellitus and its complications: report of a WHO consultation. Part 1: Diagnosis and Classification of Diabetes Mellitus. Geneva: World Health Organization, 1999.
- 8 Expert Committee on the Diagnosis and Classification of Diabetes Mellitus. Diagnosis and classification of diabetes mellitus. *Diabetes Care* 2004 **27** S5–S10.
- 9 Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF & Turner RC. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia* 1985 **28** 412–419.
- 10 Hauschka PV, Lian JB, Cole DE & Gundberg CM. Osteocalcin and matrix Gla protein: vitamin K-dependent proteins in bone. *Physiological Reviews* 1989 **69** 990–1047.
- 11 Price PA, Williamson MK & Lothringer JW. Origin of the vitamin K-dependent bone protein found in plasma and its clearance by kidney and bone. *Journal of Biological Chemistry* 1981 **256** 12760–12766.
- 12 Verhaeghe J, Van Herck E, Visser WJ, Suiker AM, Thomasset M, Einhorn TA, Faierman E & Bouillon R. Bone and mineral metabolism in BB rats with long-term diabetes. Decreased bone turnover and osteoporosis. *Diabetes* 1990 **39** 477–482.
- 13 Epstein S. Serum and urinary markers of bone remodeling: assessment of bone turnover. *Endocrine Reviews* 1988 **9** 437–449.
- 14 Pietschmann P, Schernthaner G & Woloszczuk W. Serum osteocalcin concentrations in diabetes mellitus: analysis of the type of diabetes and microvascular complication. *Diabetologia* 1988 **31** 892–895.
- 15 Rosato MT, Schneider SH & Shapses SA. Bone turnover and insulin-like growth factor I levels increase after improved glycemic control in noninsulin-dependent diabetes mellitus. *Calcified Tissue International* 1998 **63** 107–111.
- 16 Verhaeghe J, Suiker AM, Visser WJ, Van Herck E, Van Bree R & Bouillon R. The effects of systemic insulin, insulin-like growth factor-1 and growth hormone on bone growth and turnover in spontaneously diabetic BB rats. *Journal of Endocrinology* 1992 **134** 485–492.
- 17 Krakauer JC, McKenna MJ, Buderer NF, Rao DS, Whitehouse FW & Parfitt AM. Bone loss and bone turnover in diabetes. *Diabetes* 1995 **44** 775–782.
- 18 Verhaeghe J, Suiker AM, Nyomba BL, Visser WJ, Einhorn TA, Dequeker J & Bouillon R. Bone mineral homeostasis in spontaneously diabetic BB rats. II. Impaired bone turnover and decreased osteocalcin synthesis. *Endocrinology* 1989 **124** 573–582.
- 19 Gerdhem P, Isaksson A, Akesson K & Obrant KJ. Increased bone density and decreased bone turnover, but no evident alteration of fracture susceptibility in elderly women with diabetes mellitus. *Osteoporosis International* 2005 **16** 1506–1512.
- 20 Rico H, Hernandez ER, Cabranes JA & Gomez-Castresana F. Suggestion of a deficient osteoblastic function in diabetes mellitus: the possible cause of osteopenia in diabetics. *Calcified Tissue International* 1989 **45** 71–73.
- 21 Im JA, Yu BP, Jeon JY & Kim SH. Relationship between osteocalcin and glucose metabolism in postmenopausal women. *Clinical Chimica Acta* 2008 **396** 66–69.
- 22 Tang YJ, Sheu WH, Liu PH, Lee WJ & Chen YT. Positive associations of bone mineral density with body mass index, physical activity, and blood triglyceride level in men over 70 years old: a TCVDHAGE study. *Journal of Bone and Mineral Metabolism* 2007 **25** 54–59.
- 23 Bagger YZ, Rasmussen HB, Alexandersen P, Werge T, Christiansen C & Tankó LB; PERF Study Group. Links between cardiovascular disease and osteoporosis in postmenopausal women: serum lipids or atherosclerosis *per se*? *Osteoporosis International* 2007 **18** 505–512.
- 24 Yamaguchi T, Sugimoto T, Yano S, Yamauchi M, Sowa H, Chen Q & Chihara K. Plasma lipids and osteoporosis in postmenopausal women. *Endocrine Journal* 2002 **49** 211–217.
- 25 Adami S, Braga V & Gatti D. Association between bone mineral density and serum lipids in men. *Journal of the American Medical Association* 2001 **286** 791–792.
- 26 Dennison EM, Syddall HE, Aihie Sayer A, Martin HJ & Cooper C; Hertfordshire Cohort Study Group. Lipid profile, obesity and bone mineral density: the Hertfordshire cohort study. *Quarterly Journal of Medicine* 2007 **100** 297–303.
- 27 Adami S, Braga V, Zamboni M, Gatti D, Rossini M, Bakri J & Battaglia E. Relationship between lipids and bone mass in 2 cohorts of healthy women and men. *Calcified Tissue International* 2004 **74** 136–142.
- 28 Kha HT, Basseri B, Shouhed D, Richardson J, Tetradis S, Hahn TJ & Parhami F. Oxysterols regulate differentiation of mesenchymal stem cells: pro-bone and anti-fat. *Journal of Bone and Mineral Research* 2004 **19** 830–840.
- 29 Pittas AG, Harris SS, Eliades M, Stark P & Dawson-Hughes B. Association between serum osteocalcin and markers of metabolic phenotype. *Journal of Clinical Endocrinology and Metabolism* 2009 **94** 827–832.
- 30 Torróns JI, Skurnick J, Davidow AL, Korenman SG, Santoro N, Soto-Greene M, Lasser N & Weiss G; Study of Women's Health Across the Nation (SWAN). Ethnic differences in insulin sensitivity and beta-cell function in premenopausal or early perimenopausal women without diabetes: the Study of Women's Health Across the Nation (SWAN). *Diabetes Care* 2004 **27** 354–361.

Received 4 August 2009

Accepted 9 August 2009