



Osteocalcin and Risks of Incident Diabetes and Diabetic Kidney Disease: A 4.6-Year Prospective Cohort Study

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OBJECTIVE

We aimed to examine the relationship between osteocalcin (OC) and the risk of incident diabetes and the risk of incident diabetic kidney disease (DKD).

RESEARCH DESIGN AND METHODS

We followed 5,396 participants without diabetes (nondiabetes subcohort) and 1,174 participants with diabetes and normal kidney function (diabetes subcohort) at baseline. Logistic regression and modified Poisson regression models were used to estimate the relative risk (RR) of baseline OC levels with incident diabetes and DKD.

RESULTS

During a mean 4.6-year follow-up period, 296 cases of incident diabetes and 184 cases of incident DKD were identified. In the nondiabetes subcohort, higher OC levels were linearly associated with a decreased risk of diabetes (RR for 1-unit increase of \log_e -transformed OC 0.51 [95% CI 0.35–0.76]; RR for highest vs. lowest quartile 0.65 [95% CI 0.44–0.95]; P for trend < 0.05). In the diabetes subcohort, OC levels were linearly inversely associated with incident DKD (RR for 1-unit increase of \log_e -transformed OC 0.49 [95% CI 0.33–0.74]; RR for highest vs. lowest quartile 0.56 [95% CI 0.38–0.83]; P for trend < 0.05), even independent of baseline estimated glomerular filtration rate and urinary albumin-to-creatinine ratio. No significant interactions between OC and various subgroups on incident diabetes or DKD were observed.

CONCLUSIONS

Lower OC levels were associated with an increased risk of incident diabetes and DKD.

Increasing knowledge about metabolism has led to the emergence of bone-derived factors and hormones as important regulators of energy metabolism. Osteocalcin (OC), the most abundant noncollagenous protein in the bone matrix and synthesized by osteoblasts, is not only a marker of bone formation but also, importantly, a metabolically active hormone involved in the regulation of glucose and lipid metabolism (1–4). Circulating OC has two major forms: undercarboxylated and carboxylated. Although circulating undercarboxylated OC has been observed to stimulate β -cell proliferation, increasing insulin secretion and sensitivity in animal experiments (5,6), human studies

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more often reported the effect of circulating total OC because it is easier to measure. Currently, only five prospective cohort studies have reported the association of baseline total OC levels and risk of diabetes (7–11). The three studies with large samples (between 1,000 and 2,000 patients) showed that lower OC levels were associated with an increased risk of diabetes (9–11), but the other two studies (one with a small sample size and one including only men) did not find this association (7,8).

Diabetic kidney disease (DKD) is present in up to 36% of people with diabetes and is the most common diabetic chronic complication (12). The presence of DKD not only severely increases the complexity of diabetes management (13) but also significantly increases the risks of all-cause and cardiovascular mortality (14). Detecting some possible molecular markers related to incident DKD is essential for screening populations at high risk of DKD and for research into DKD onset mechanisms. Some adverse conditions related to lower OC levels, such as obesity, insulin resistance, and hyperglycemia (15), may further deteriorate and contribute to the development of DKD, especially hyperglycemia, a central upstream driver of DKD (16). Hence, we wanted to find out whether the relationship of OC with DKD is similar to that of OC with diabetes. However, to our knowledge, no population-based prospective studies have examined OC as a predictor for incident DKD. In this study, therefore, we aimed to simultaneously investigate the associations of total OC levels with incident diabetes and incident DKD on the basis of a 4.6-year community-based prospective cohort.

RESEARCH DESIGN AND METHODS

Study Participants

This community-based longitudinal observational study was embedded in the Shanghai Nичeng Cohort Study, which was designed to prospectively investigate the prevalence, incidence, and related factors of cardiometabolic diseases. The cohort has been described in detail previously (17). From the general population of the Nичeng area in Shanghai, 17,212 individuals aged 45–70 years were recruited between 2013 and 2014, and among them, 10,075 aged 55–70

years were invited to participate in the follow-up survey in 2018. Finally, 7,230 individuals were followed up (a follow-up rate of 71.8%), of whom 7,069 participated in the onsite survey and 161 died.

Participants were divided into two subcohorts according to the presence of diabetes at baseline. The nondiabetes subcohort (participants without prevalent diabetes at baseline) was included for analysis of the association between OC and incident diabetes, and the diabetes subcohort (participants with prevalent diabetes at baseline) was included for analysis of the association between OC and incident DKD. For the nondiabetes subcohort, we further excluded participants with missing data on OC, diseases, or drugs that may affect bone metabolism at baseline, and those with missing data on diabetes at follow-up were excluded. For the diabetes subcohort, participants with prevalent DKD, missing data on OC, diseases, or drugs that may affect bone metabolism at baseline; missing data on estimated glomerular filtration rate (eGFR) and/or urinary albumin-to-creatinine ratio (UACR) at baseline and follow-up, and a history of infection in the past 2 weeks at follow-up were excluded. Here, diseases that may affect bone metabolism included the presence of cirrhosis, thyroid diseases, parathyroid diseases, malignancy, and osteoporosis, and drugs that may affect bone metabolism included glucocorticoids, thyroid hormones, vitamin D, bisphosphonates, and vitamin K antagonists. Finally, a total of 5,396 participants were included in the nondiabetes subcohort, and 1,174 participants were included in the diabetes subcohort (Supplementary Fig. 1). The ethics committee of the Shanghai Sixth People's Hospital approved this study. Written informed consent was obtained from all participants.

Clinical Measurements

A standardized questionnaire was used to collect data on demographics, smoking status, drinking status, physical activity, medical history, and other information at baseline and follow-up. Current smokers were defined as those who smoked at least one cigarette per day in the past year. Current drinkers were defined as those who drank at least 1 g of alcohol

weekly in the past year. Height, weight, systolic blood pressure (SBP), and diastolic blood pressure (DBP) were measured according to an established standard method (18). BMI was calculated as weight in kilograms divided by the square of the height in meters.

Before and during this survey, all participants were required to have enough rest. Blood samples were collected during a morning visit after an overnight fast of at least 10 h. Electrochemiluminescence immunoassay (cobas e601; Roche Diagnostics GmbH, Mannheim, Germany) was used to detect the serum total OC, including both the intact molecule and the N-terminal/mid-region fragments, at baseline. The interassay coefficient of variation range was 1.1–1.6%. Hemoglobin A_{1c} (HbA_{1c}) was assessed by high-performance liquid chromatography. Fasting plasma glucose (FPG) was assessed by glucose oxidase method. Triglyceride (TG) and total cholesterol (TC) were assessed by enzymatic colorimetric method. HDL cholesterol (HDL-C) and LDL cholesterol (LDL-C) were assessed by direct method. Serum creatinine was assessed by sarcosine oxidase-PAP (phenol-aminophenazone peroxidase) method. Fasting insulin (FINS) was assessed by electrochemiluminescence immunoassay. Adiponectin was quantified using a latex particle-enhanced immunoturbidimetric assay (Antibody and Immunoassay Services, University of Hong Kong) (19). Urine creatinine and albumin were assessed by rate nephelometry assay. Insulin resistance was estimated by the HOMA of insulin resistance (HOMA-IR), calculated as FINS (mU/L) × FPG (mmol/L) / 22.5 (20).

Outcome Definition

Diabetes was diagnosed according to a self-reported history of diabetes, FPG ≥ 7.0 mmol/L, and/or HbA_{1c} $\geq 6.5\%$ on the basis of American Diabetes Association guidelines (21). eGFR was estimated using the Chronic Kidney Disease Epidemiology Collaboration equation (22). A reduced eGFR was defined as < 60 mL/min/1.73 m². Albuminuria was defined as UACR ≥ 30 mg/g. DKD was a clinical diagnosis based on the presence of albuminuria and/or reduced eGFR in the absence of signs or symptoms of other primary causes of kidney damage (23).

Statistical Analysis

Continuous variables are presented as median (interquartile range) and categorical variables as numbers (proportions). For continuous variables, distributions between two groups were compared using the Wilcoxon rank sum test, and for categorical variables, proportions between two groups were compared using Pearson χ^2 test. Spearman partial correlation coefficients controlling for age and sex were used to assess correlations between OC and other variables. Logistic regression models were used to estimate the risk ratio (RR) and 95% CI for incident diabetes. In addition, since incident DKD was not a rare event in our study (incidence rate >10%), RR estimated with logistic regression might be overestimated; thus, a modified Poisson regression model with robust error variance was used to estimate RR and 95% CI for incident DKD (24,25). The adjustment variables included in the regression analyses were selected according to the following criteria: The confounders were 1) reported in the relevant literature, 2) related to the outcome, and 3) not mediators in the causal pathway. Potential mediators were examined by using the counterfactual mediation approach, which was implemented using the PROC CAUSALMED procedure in SAS (SAS Institute). To test the linear trend, median values of each quartile group as a continuous variable were assigned to the regression models. Potential interactions between OC and the other adjustment variables on incident outcome were tested using the Wald test by adding a product variable to the regression models.

We conducted all statistical analyses using Stata/MP 16 (Stata Corp) and SAS 9.4 software. A two-sided $P < 0.05$ was considered statistically significant.

RESULTS

Baseline clinical and demographic characteristics of the nondiabetes and diabetes subcohorts are presented in Table 1. Although the participants in whom DKD developed were significantly older and included a higher proportion of women, there were no differences in median ages and sex proportions between the participants with and without diabetes. Participants in whom either diabetes or DKD developed all included a higher proportion of noncurrent drinkers, had a lower OC level, and had unfavorable

metabolic profiles characterized by higher levels of BMI, SBP, HOMA-IR, FPG, HbA_{1c}, and TG but lower levels of HDL-C (Table 1). Significant negative correlations were found between OC and BMI, HOMA-IR, FPG, and HbA_{1c} after adjustment for age and sex (all $P < 0.001$) (Supplementary Table 1).

There was a linearly inverse association between OC and the risk of incident diabetes (P for trend < 0.05); RR of incident diabetes was 0.51 (95% CI 0.35–0.76) per 1-unit increase of log_e-transformed OC (Table 2). In the multivariable model (model 3), the participants with the highest quartile of OC had a 35% lower risk of incident diabetes (RR 0.65 [95% CI 0.44–0.95]) compared with those with the lowest quartile of OC.

The multivariable regression models showed a linear association between baseline OC concentrations and risk of incident DKD (RR for incident DKD per 1-unit increase of log_e-transformed OC 0.49 [95% CI 0.33–0.74]). Higher baseline OC concentrations were significantly associated with a decreased risk of incident DKD (P for linear trend < 0.05); compared with those with the lowest quartile, RR of incident DKD for the participants with the highest quartile of OC was 0.56 (95% CI 0.38–0.83) (Table 3).

The associations between baseline OC and incident diabetes or DKD among the various subgroups are shown in Fig. 1. Statistically significant inverse associations between OC and the risk of incident diabetes were found among the various age and following subgroups: women, participants without a family history of diabetes, overweight or obese participants (BMI ≥ 24 kg/m²), and participants with normal SBP and DBP ($< 140/90$ mmHg) (Fig. 1A). The associations between OC and the risk of incident DKD reached statistical significance in the various sex, age, and blood pressure subgroups and only in participants with diabetes duration < 5 years or who were overweight or obese (BMI ≥ 24 kg/m²) (Fig. 1B). However, there were no significant interaction effects between OC and any one of these subgroups on incident diabetes or DKD (all P for interaction > 0.05).

CONCLUSIONS

Our study is currently the largest community-based prospective cohort study

investigating the association between OC and incident diabetes and the first to report on the association between OC and incident DKD using a prospective cohort. This study demonstrates that higher serum total OC concentrations are associated with a reduced risk of incident diabetes and DKD. Our data provide further insight into the impact of OC on metabolic diseases in addition to its role as a marker of bone formation.

OC and Risks of Incident Diabetes and DKD

The reported association between total OC and risk of diabetes have been inconsistent among several longitudinal cohort studies (7–11). In agreement with our results, two longitudinal studies, one including 1,870 hospitalized patients from China (9) and the other including only 1,691 postmenopausal women from Japan (10), reported an inverse association between OC and incident diabetes. In a study of 1,455 older women in the U.S., the authors argued that lower OC levels are associated with increased risks of incident diabetes, approaching statistical significance (hazard ratio 0.85 per SD increase [95% CI 0.71–1.02]; $P = 0.075$) (11). Inconsistent with our results, two studies found no significant association between OC and risk of incident diabetes (7,8), but one was a small sample of 307 individuals (8), the other included only men (7), and the diagnosis of diabetes in both studies did not consider HbA_{1c} levels. Therefore, the association between OC and risk of incident diabetes is not yet conclusive. Our study in 5,396 participants without diabetes at baseline demonstrated that serum OC is inversely associated with incident diabetes. Notably, we did not find any significant interaction effects in the various subgroups, indicating that our results are relatively credible. In addition, the association between OC and diabetes was attenuated by adding BMI to the regression model but did not materially influence the results (data not shown).

So far, there have been no prospective studies on OC and the risk of incident DKD. Some conclusions regarding the association of OC and DKD, drawn from some small-sample cross-sectional studies, are still controversial (26–28). Zhao et al. (26) found a significant positive correlation between UACR and OC in a cross-sectional study of 297 patients with

Table 1—Baseline characteristics of the study population with or without incident diabetes or DKD

Characteristic	Nondiabetes subcohort			Diabetes subcohort			
	Total (n = 5,396)	No diabetes (n = 5,100)	Incident diabetes (n = 296)	Total (n = 1,174)	No DKD (n = 990)	Incident DKD (n = 184)	P
Age, years	61.6 (58.7–65.1)	61.6 (58.7–65.1)	62.3 (58.8–65.5)	61.9 (58.9–65.6)	61.7 (58.8–65.3)	63.9 (59.7–67.1)	<0.001
Women	3,005 (55.7)	2,836 (55.6)	169 (57.1)	703 (59.9)	575 (58.1)	128 (69.6)	0.004
Current smoker	1,180 (21.9)	1,115 (21.9)	65 (22.0)	209 (17.8)	184 (18.6)	25 (13.6)	0.10
Current drinker	829 (15.4)	796 (15.6)	33 (11.1)	154 (13.1)	144 (14.5)	10 (5.4)	<0.001
Physical activity*	189 (3.5)	179 (3.5)	10 (3.4)	48 (4.1)	40 (4.0)	8 (4.3)	0.85
Osteocalcin, ng/mL	21.8 (17.2–27.8)	21.9 (17.3–27.9)	20.4 (16.5–26.2)	18.1 (14.2–22.9)	18.4 (14.3–23.3)	17.0 (13.2–21.5)	0.004
BMI, kg/m ²	24.6 (22.6–26.7)	24.5 (22.5–26.6)	26.0 (24.0–28.2)	26.0 (23.8–27.9)	25.9 (23.7–27.9)	26.7 (24.6–28.0)	0.013
SBP, mmHg	131.0 (123.0–142.0)	131.0 (123.0–141.5)	138.0 (129.0–148.5)	137.0 (127.0–149.0)	135.3 (127.0–147.0)	140.0 (130.0–155.0)	<0.001
DBP, mmHg	82.0 (79.0–89.0)	82.0 (79.0–88.5)	84.0 (80.0–90.0)	83.0 (80.0–90.0)	83.0 (80.0–89.0)	83.0 (80.0–90.0)	0.91
TC, mmol/L	5.1 (4.5–5.8)	5.1 (4.5–5.7)	5.2 (4.6–6.0)	5.3 (4.6–6.0)	5.3 (4.6–6.0)	5.3 (4.5–5.9)	0.95
TG, mmol/L	1.3 (0.9–1.9)	1.3 (0.9–1.8)	1.6 (1.1–2.3)	1.5 (1.1–2.3)	1.5 (1.1–2.2)	1.8 (1.3–2.7)	<0.001
HDL-C, mmol/L	1.3 (1.1–1.5)	1.3 (1.1–1.6)	1.2 (1.0–1.4)	1.3 (1.1–1.5)	1.3 (1.1–1.5)	1.2 (1.0–1.4)	0.006
LDL-C, mmol/L	3.0 (2.6–3.6)	3.0 (2.6–3.6)	3.2 (2.7–3.9)	3.2 (2.6–3.7)	3.2 (2.6–3.7)	3.2 (2.6–3.7)	0.58
FINS, μU/mL	6.5 (4.6–9.2)	6.4 (4.6–9.0)	8.3 (5.8–11.7)	8.7 (5.8–13.1)	8.3 (5.8–12.7)	10.4 (6.9–16.1)	<0.001
HOMA-IR	1.7 (1.2–2.4)	1.6 (1.1–2.3)	2.3 (1.6–3.3)	3.1 (2.0–4.7)	2.9 (2.0–4.6)	3.7 (2.5–5.8)	<0.001
FFPG, mmol/L	5.8 (5.4–6.1)	5.7 (5.4–6.1)	6.3 (6.0–6.6)	7.7 (7.0–8.9)	7.6 (7.0–8.8)	8.1 (7.0–9.8)	0.011
HbA _{1c} %	5.6 (5.4–5.8)	5.6 (5.3–5.8)	6.0 (5.8–6.2)	6.7 (6.2–7.5)	6.7 (6.2–7.5)	7.0 (6.5–7.9)	<0.001
mmol/mol	37.7 (35.5–39.9)	37.7 (34.4–39.9)	42.1 (39.9–44.3)	49.7 (44.3–58.5)	49.7 (44.3–58.5)	52.4 (47.5–62.8)	<0.001
eGFR, mL/min/1.73 m ²	95.4 (89.5–99.6)	95.4 (89.4–99.6)	95.4 (90.5–100.0)	96.4 (91.4–101.3)	96.7 (91.8–101.6)	94.5 (80.4–100.0)	<0.001
UA CR, mg/mg	6.4 (4.4–10.7)	6.4 (4.4–10.6)	7.3 (5.2–13.3)	7.4 (5.0–11.9)	6.9 (4.9–10.6)	12.0 (6.5–18.0)	<0.001
Adiponectin, μg/mL	4.2 (3.1–5.5)	4.2 (3.1–5.6)	3.5 (2.6–4.5)	3.5 (2.6–4.7)	3.6 (2.7–4.7)	3.3 (2.5–4.8)	0.14

Data are n (%) for categorical measures or median (interquartile range) for continuous measures. *Physical activity was defined as leisure-time physical activity ≥30 min/day.

Table 2—Effects of OC on incident diabetes

	Per 1-unit increase	Quartile 1	Quartile 2	Quartile 3	Quartile 4	<i>P</i> for trend
Participants, <i>n</i>		1,350	1,349	1,349	1,348	
Events, <i>n</i> (%)		80 (5.9)	97 (7.2)	57 (4.2)	62 (4.6)	
Model 1	0.51 (0.35–0.75)	Reference	1.16 (0.84–1.58)	0.62 (0.43–0.90)	0.65 (0.44–0.95)	0.002
Model 2	0.50 (0.34–0.74)	Reference	1.15 (0.84–1.58)	0.61 (0.42–0.88)	0.64 (0.44–0.94)	0.002
Model 3	0.51 (0.35–0.76)	Reference	1.15 (0.84–1.59)	0.62 (0.43–0.91)	0.65 (0.44–0.95)	0.003

Data are RR (95% CI) unless otherwise indicated. RR for per 1-unit increase was estimated from 1-unit increase of log_e-transformed OC. Model 1 was adjusted for age and sex. Model 2 was adjusted for age, sex, family history of diabetes (yes/no), physical activity ≥30 min/day (yes/no), current smoker (yes/no), and current drinker (yes/no). Model 3 was adjusted for age, sex, family history of diabetes (yes/no), physical activity ≥30 min/day (yes/no), current smoker (yes/no), current drinker (yes/no), SBP, TC, LDL-C, and UACR.

type 2 diabetes; however, we should be alert to the possibility of reverse causation. In cross-sectional studies, reduced renal function may result in decreased OC excretion and, consequently, in an increasing circulating OC level (i.e., the exposure-disease relationship may be distorted). Additionally, a case-control study of 46 adolescents and young adults with type 1 diabetes reported that OC levels were significantly lower in those with persistent microalbuminuria than in those with normoalbuminuria (29). Our prospective cohort study demonstrates that serum OC has an independent and robust inverse association with incident DKD, even after adjustment for eGFR and UACR levels at baseline.

The Potential Mechanism Between OC and Diabetes and DKD

It has been demonstrated that OC-null mice can accumulate abnormal amounts of visceral fat and display decreased β-cell proliferation, hyperglycemia, decreased insulin secretion, and insulin resistance (6). Furthermore, the administration of OC could significantly weaken the deleterious effect on glucose metabolism and fat

mass in wild-type mice (30). In our study, we found that OC concentrations were inversely associated with BMI, FINS, HOMA-IR, and blood glucose (FPG and HbA_{1c}) at baseline, which is consistent with these animal experiments to some extent. Meanwhile, we also found that most of the association between OC and incident diabetes was mediated through BMI, insulin resistance, blood glucose, and adiponectin (Supplementary Table 2). Of note, adiponectin is an adipokine and enhances insulin sensitivity (31). Animal studies have shown that OC can promote insulin sensitivity by increasing the expression of adiponectin in adipocytes (6,30). Our results provided further prospective evidence in humans that the effect of OC on diabetes might be partially mediated by adiponectin.

Our results suggest that the effects of OC on DKD are partially mediated through TG, HDL-C, and HbA_{1c}, but they all account for only a small percentage of mediation (Supplementary Table 3). Therefore, some other pathogenic pathways need to be considered. For example, a previous study found that OC could increase endothelial nitric oxide

synthase levels by activating the Akt/endothelial nitric oxide synthase pathway and played an important role in regulating endothelial function (32). Since endothelial function was a key contributor to DKD progression (16,33), it is also logical to propose that OC is associated with incident DKD. Overall, the underlying mechanistic link of OC with incident DKD remains unclear, and further research is warranted to explore more potential mechanisms between OC and DKD.

Additional Predictive Value of OC Compared With the Traditional Risk Model

Identifying patients at high risk of incident diabetes and DKD could help us to provide better treatment strategies and improve clinical outcomes for patients. OC was significantly associated with incident diabetes and DKD. However, whether OC adds information to traditional prediction risk models is still not clear. Thus, we assessed the incremental predictive performance of OC based on previously established diabetes and DKD risk prediction models (34,35). We found

Table 3—Effects of OC on incident DKD

	Per 1-unit increase	Quartile 1	Quartile 2	Quartile 3	Quartile 4	<i>P</i> for trend
Participants, <i>n</i>		294	294	293	293	
Events, <i>n</i> (%)		57 (19.4)	49 (16.7)	43 (14.7)	35 (11.9)	
Model 1	0.39 (0.26–0.59)	Reference	0.76 (0.53–1.07)	0.62 (0.43–0.90)	0.45 (0.30–0.67)	<0.001
Model 2	0.44 (0.29–0.68)	Reference	0.75 (0.53–1.07)	0.66 (0.45–0.97)	0.50 (0.33–0.76)	0.003
Model 3	0.49 (0.33–0.74)	Reference	0.87 (0.62–1.21)	0.66 (0.45–0.97)	0.56 (0.38–0.83)	0.004

Data are RR (95% CI) unless otherwise indicated. RR for per 1-unit increase was estimated from 1-unit increase of log_e-transformed OC. Model 1 was adjusted for age and sex. Model 2 was adjusted for age, sex, diabetes duration, current smoker (yes/no), current drinker (yes/no), glucose-lowering treatment (yes/no), antihypertensive treatment (yes/no), and prevalent cardiovascular disease (yes/no). Model 3 was adjusted for age, sex, diabetes duration, current smoker (yes/no), current drinker (yes/no), glucose-lowering treatment (yes/no), antihypertensive treatment (yes/no), prevalent cardiovascular disease (yes/no), BMI, SBP, TC, LDL-C, FPG, eGFR, and UACR.

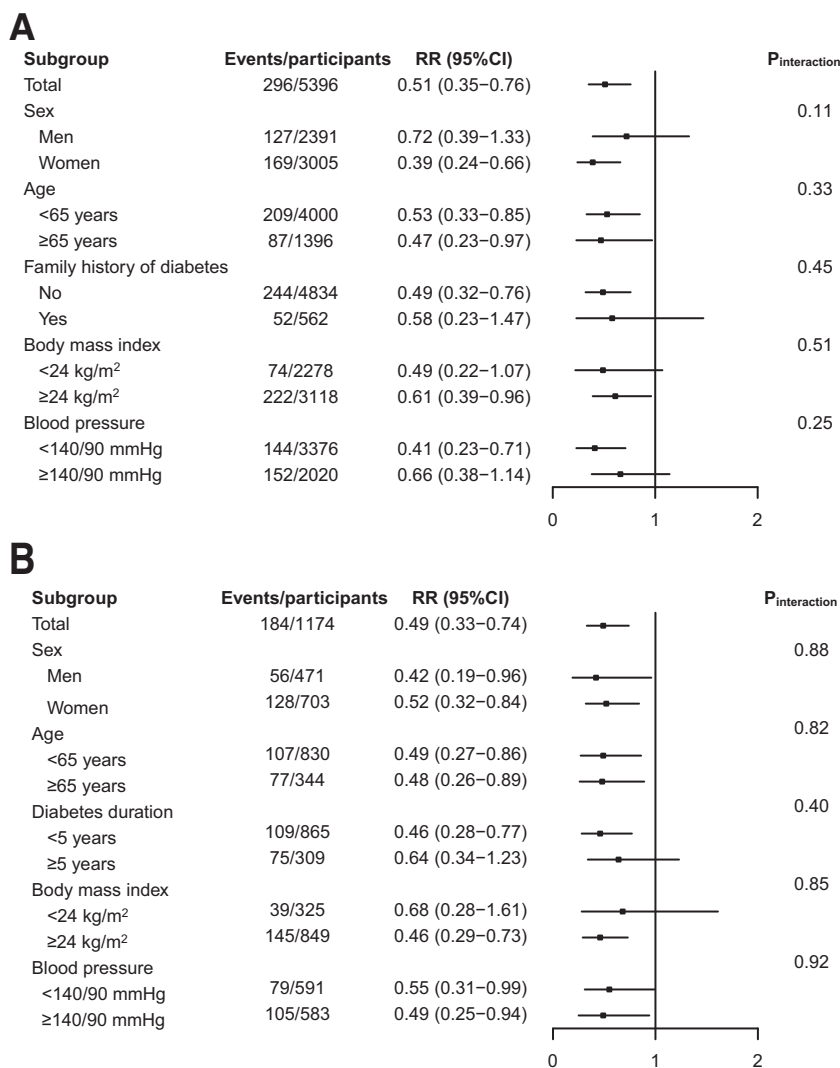


Figure 1—Effects of OC on incident diabetes or DKD in various subgroups. RR was estimated per 1-unit increase of \log_e -transformed OC. **A:** OC and the risk of incident diabetes. The model was adjusted for age, sex, family history of diabetes (yes/no), physical activity ≥ 30 min/day (yes/no), current smoker (yes/no), current drinker (yes/no), SBP, TC, LDL-C, and UACR. **B:** OC and the risk of incident DKD. The model was adjusted for age, sex, diabetes duration, current smoker (yes/no), current drinker (yes/no), glucose-lowering treatment (yes/no), antihypertensive treatment (yes/no), prevalent cardiovascular disease (yes/no), BMI, SBP, TC, LDL-C, FPG, eGFR, and UACR.

that adding OC could not improve the performance of the traditional diabetes risk prediction model. The reason might be related to the presence of BMI and HbA_{1c} in the traditional diabetes model, both of which are associated with OC. On the other hand, we found that an additional predictive value was detected when adding OC to the DKD risk model, as shown by the significantly improved integrated discriminant index values (Supplementary Table 4). Although statistical improvements were slight, any small improvement in risk prediction might be vital for screening patients with diabetes at a higher risk of DKD.

We also believe that the value of OC lies not only in adding additional predictive value to the traditional model but also in providing some new insights (e.g., endocrine role of bone in metabolic disease) into the pathogenetic pathway of diabetes and DKD.

Strengths and Limitations

This study has several strengths. First, it is the largest sample size used to study the association between baseline OC and the risk of incident diabetes and, at the same time, the first to report on the association between baseline OC and the risk of incident DKD in a community-

based cohort. In addition, complete and multifaceted data collection allowed us to assess common confounders and infer causal mechanism exploration.

Several limitations must also be considered. First, the lack of data on bone turnover markers other than OC limited our ability to make a broader exploration of the association of bone with diabetes and DKD. Second, a single morning urine sample was collected, and results based on a single measurement may be affected by day-to-day variability of UACR within individuals. However, it has been shown that the single testing of UACR could be a feasible alternative in a large community cohort study (36–38). Meanwhile, when DKD was defined as the presence of macroalbuminuria (UACR ≥ 300 mg/g) and/or reduced eGFR, the inverse association of OC with incident DKD still existed, supporting the robustness of our findings (data not shown). Third, our cohort only included the middle-aged and older population, making our results inapplicable to younger age-groups. Fourth, we did not perform screening programs for diabetes classification, such as islet autoantibody testing, so we could not have a clear classification of diabetes type. Finally, although we excluded osteoporosis by use of osteoporosis medications or an underlying diagnosis, it remains possible that a small portion of osteoporosis was missed, especially in postmenopausal women. Therefore, to better exclude the potential effect of residual confounding, further studies are needed with better designs to take confounders fully into account.

In conclusion, circulating OC has the potential to serve as a biomarker for the detection of incident diabetes and DKD and may even be targeted as a future therapeutic for diabetes and its related microvascular diseases. Our results are preliminary and hypothesis generating; thus, further prospective observational and interventional studies are needed to clarify the underlying mechanisms and establish causality.

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References

- Dirckx N, Moorer MC, Clemens TL, Riddle RC. The role of osteoblasts in energy homeostasis. *Nat Rev Endocrinol* 2019;15:651–665
- Magni P, Macchi C, Sirtori CR, Corsi Romanelli MM. Osteocalcin as a potential risk biomarker for cardiovascular and metabolic diseases. *Clin Chem Lab Med* 2016;54:1579–1587
- Cipriani C, Colangelo L, Santori R, et al. The Interplay between bone and glucose metabolism. *Front Endocrinol (Lausanne)* 2020;11:122
- Fukumoto S, Martin TJ. Bone as an endocrine organ. *Trends Endocrinol Metab* 2009;20:230–236
- Ferron M, McKee MD, Levine RL, Ducy P, Karsenty G. Intermittent injections of osteocalcin improve glucose metabolism and prevent type 2 diabetes in mice. *Bone* 2012;50:568–575
- Lee NK, Sowa H, Hinoi E, et al. Endocrine regulation of energy metabolism by the skeleton. *Cell* 2007;130:456–469
- Hwang YC, Jee JH, Jeong IK, Ahn KJ, Chung HY, Lee MK. Circulating osteocalcin level is not associated with incident type 2 diabetes in middle-aged male subjects: mean 8.4-year retrospective follow-up study. *Diabetes Care* 2012;35:1919–1924
- Liatis S, Sfikakis PP, Tsiakou A, et al. Baseline osteocalcin levels and incident diabetes in a 3-year prospective study of high-risk individuals. *Diabetes Metab* 2014;40:198–203
- Shu H, Pei Y, Chen K, Lu J. Significant inverse association between serum osteocalcin and incident type 2 diabetes in a middle-aged cohort. *Diabetes Metab Res Rev* 2016;32:867–874
- Urano T, Shiraki M, Kuroda T, et al. Low serum osteocalcin concentration is associated with incident type 2 diabetes mellitus in Japanese women. *J Bone Miner Metab* 2018;36:470–477
- Massera D, Biggs ML, Walker MD, et al. Biochemical markers of bone turnover and risk of incident diabetes in older women: the Cardiovascular Health Study. *Diabetes Care* 2018;41:1901–1908
- International Diabetes Federation. IDF Diabetes Atlas. 9th ed. Accessed 19 April 2021. Available from https://www.diabetesatlas.org/upload/resources/material/20200302_133351_IDFATLAS9e-final-web.pdf
- Perkovic V, Agarwal R, Fioretto P, et al.; Conference Participants. Management of patients with diabetes and CKD: conclusions from a “Kidney Disease: Improving Global Outcomes” (KDIGO) Controversies Conference. *Kidney Int* 2016;90:1175–1183
- Afkarian M, Sachs MC, Kestenbaum B, et al. Kidney disease and increased mortality risk in type 2 diabetes. *J Am Soc Nephrol* 2013;24:302–308
- Kunutsor SK, Apekey TA, Laukkanen JA. Association of serum total osteocalcin with type 2 diabetes and intermediate metabolic phenotypes: systematic review and meta-analysis of observational evidence. *Eur J Epidemiol* 2015;30:599–614
- Anders HJ, Huber TB, Isermann B, Schiffer M. CKD in diabetes: diabetic kidney disease versus nondiabetic kidney disease. *Nat Rev Nephrol* 2018;14:361–377
- Chen P, Hou X, Hu G, et al. Abdominal subcutaneous adipose tissue: a favorable adipose depot for diabetes? *Cardiovasc Diabetol* 2018;17:93
- Luepker RV, Evans A, McKeigue P, Reddy KS. *Cardiovascular Survey Methods*. 3rd ed. Geneva, World Health Organization, 2004
- Liu D, Wu L, Gao Q, et al. FGF21/adiponectin ratio predicts deterioration in glycemia: a 4.6-year prospective study in China. *Cardiovasc Diabetol* 2021;20:157
- 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
- American Diabetes Association. Diagnosis and classification of diabetes mellitus. *Diabetes Care* 2010;33(Suppl. 1):S62–S69
- Levey AS, Stevens LA, Schmid CH, et al.; CKD-EPI (Chronic Kidney Disease Epidemiology Collaboration). A new equation to estimate glomerular filtration rate. *Ann Intern Med* 2009;150:604–612
- American Diabetes Association. 11. Microvascular Complications and Foot Care: *Standards of Medical Care in Diabetes-2021*. *Diabetes Care* 2021;44(Suppl. 1):S151–S167
- McNutt LA, Wu C, Xue X, Hafner JP. Estimating the relative risk in cohort studies and clinical trials of common outcomes. *Am J Epidemiol* 2003;157:940–943
- Zou G. A modified Poisson regression approach to prospective studies with binary data. *Am J Epidemiol* 2004;159:702–706
- Zhao X, Zhang XM, Yuan N, Yu XF, Ji LN. Associations of bone mineral density and bone metabolism indices with urine albumin to creatinine ratio in Chinese patients with type 2 diabetes. *Exp Clin Endocrinol Diabetes* 2019;127:50–55
- Ren H, Ma X, Shao Y, Han J, Yang M, Wang Q. Correlation Between Serum miR-154-5p and osteocalcin in males and postmenopausal females of type 2 diabetes with different urinary albumin creatinine ratios. *Front Endocrinol (Lausanne)* 2019;10:542
- Zhu X, Zhou Y, Hong S, Xue Y, Cui Y. Correlation between serum bone turnover markers and estimated glomerular filtration rate in Chinese patients with diabetes. *Dis Markers* 2021;2021:6731218
- Verrotti A, Basciani F, Carle F, Morgese G, Chiarelli F. Calcium metabolism in adolescents and young adults with type 1 diabetes mellitus without and with persistent microalbuminuria. *J Endocrinol Invest* 1999;22:198–202
- Ferron M, Hinoi E, Karsenty G, Ducy P. Osteocalcin differentially regulates beta cell and adipocyte gene expression and affects the development of metabolic diseases in wild-type mice. *Proc Natl Acad Sci U S A* 2008;105:5266–5270
- Yamauchi T, Kamon J, Waki H, et al. The fat-derived hormone adiponectin reverses insulin resistance associated with both lipodystrophy and obesity. *Nat Med* 2001;7:941–946
- Dou J, Li H, Ma X, et al. Osteocalcin attenuates high fat diet-induced impairment of endothelium-dependent relaxation through Akt/eNOS-dependent pathway. *Cardiovasc Diabetol* 2014;13:74
- Reidy K, Kang HM, Hostetter T, Susztak K. Molecular mechanisms of diabetic kidney disease. *J Clin Invest* 2014;124:2333–2340
- Heianza Y, Arase Y, Hsieh SD, et al. Development of a new scoring system for predicting the 5 year incidence of type 2 diabetes in Japan: the Toranomon Hospital Health Management Center Study 6 (TOPICS 6). *Diabetologia* 2012;55:3213–3223
- Jiang W, Wang J, Shen X, et al. Establishment and validation of a risk prediction model for early diabetic kidney disease based on a systematic review and meta-analysis of 20 cohorts. *Diabetes Care* 2020;43:925–933
- Hemmati R, Gharipour M, Khosravi A, Jozan M. A cost-benefit and accurate method for assessing microalbuminuria: single versus frequent urine analysis. *Int J Hypertens* 2013;2013:752903
- Blecker S, Matsushita K, Köttgen A, et al. High-normal albuminuria and risk of heart failure in the community. *Am J Kidney Dis* 2011;58:47–55
- Foster MC, Ghuman N, Hwang SJ, Murabito JM, Fox CS. Low ankle-brachial index and the development of rapid estimated GFR decline and CKD. *Am J Kidney Dis* 2013;61:204–210