Association of serum proprotein convertase subtilisin/kexin type 9 with early atherosclerosis in newly diagnosed type 2 diabetes mellitus

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Abstract *Background and Aim:* Proprotein convertase subtilisin/kexin type 9 (PCSK9) is rapidly gaining attention as a potential risk of developing atherosclerosis due to its crucial role in the regulation of low-density lipoprotein cholesterol (LDL-C) metabolism. The present study investigated the relationship between serum PCSK9 levels and early atherosclerosis as assessed by carotid intimal-medial thickness (CIMT) and brachial-ankle pulse wave velocity (ba-PWV) in newly diagnosed type 2 diabetes mellitus (T2DM).

Methods and Results: A total of 100 newly diagnosed T2DM were enrolled and further divided into the thickened CIMT group (n = 41) and the non-thickened CIMT group (n = 59) according to the results of color Doppler ultrasonography. Serum PCSK9 levels, CIMT, ba-PWV, and metabolic parameters were measured.

Patients in the thickened CIMT group had higher serum PCSK9 levels than patients in the non-thickened CIMT group (all $P < 0.05$). CIMT and ba-PWV were both positively correlated to serum PCSK9 levels, while serum PCSK9 levels were positively correlated to white blood cell count, neutrophil, lymphocyte, and high-sensitivity C-reactive protein ($P < 0.05$). Multiple linear regression indicated that serum PCSK9 level was an independent predictor of CIMT ($\beta = 0.637$, $P < 0.001$) and ba-PWV ($\beta = 0.600$, $P < 0.001$). Binary logistic regression analysis showed that serum PCSK9 levels were independent risk factors of thickened CIMT [$OR = 1.120$, $95\%CI$ ($1.041-1.204$), $P = 0.002$].

Conclusion: Serum PCSK9 levels are significantly correlated with CIMT and ba-PWV, independent of CAD risk factors. Therefore, serum PCSK9 level may have the potential to serve as a prescriptive biomarker for early arteriosclerosis in newly diagnosed T2DM.

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Introduction

Coronary artery disease (CAD) arising from atherosclerosis is the most common macrovascular complication and cause of death in type 2 diabetes mellitus (T2DM). T2DM patients exhibit dyslipidemia characterized by elevated plasma triglycerides (TG) levels, low high-density lipoprotein cholesterol (HDL-C) levels and increased low-density lipoprotein cholesterol (LDL-C) levels, which all associated with increased risk of CAD [1]. Particularly, LDL-C has been regarded as an independent risk factor and predictor of CAD [2]. Circulating LDL-C is removed from the plasma mainly by hepatic LDL receptor (LDLR) uptake. This pathway accounts for regulating the clearance of 56−80% serum LDL-C [3].

Proprotein convertase subtilisin/kexin type 9 (PCSK9) is a serine protease synthesized primarily in the liver. PCSK9 plays a key role in the regulation of LDL-C metabolism by increasing the degradation of LDLR on the surface of hepatocytes, which results in the reduction of the uptake of LDL-C via LDLR, thus higher serum PCSK9 expression leading to increase of serum LDL-C levels [4]. In light of this theory, serum PCSK9 is rapidly gaining attention as a potential risk of developing atherosclerosis. Gain-of-function mutations in PCSK9 can cause hypercholesterolemia and ultimately lead to premature CAD, whereas loss-of-function mutations in PCSK9 promotes LDL-C uptake and reduce the risk of CAD [5]. When fed with a 12-month Western diet, PCSK9 knockout mice accumulated 4-fold less aortic cholesteryl esters (CE) than WT mice, whereas mice overexpressed PCSK9 presented with higher CE contents in the aorta and more severe aortic lesions [6]. Recently, it has been shown that PCSK9 is also detectable in human atherosclerotic plaques [7]. Clinical studies have found a significant positive correlation between circulating PCSK9 levels and the severity of CAD or cardiovascular risk [8,9].

In fact, many cases of coronary artery disease (CAD) could be prevented by timely and appropriate primary or secondary prevention. Therefore, it is essential to establish a simple method for the detection of early atherosclerosis and intervene it earlier in order to prevent atherosclerosis progression. Carotid intima-media thickness (CIMT) and brachial−ankle pulse wave velocity (ba-PWV) are well-established markers of early atherosclerosis [10]. Although the association between PCSK9 and CAD is well known, there are limited data on the association between serum PCSK9 levels and early atherosclerosis. Therefore, this study aimed to explore the relationship between serum PCSK9 levels and early atherosclerosis as assessed by CIMT and ba-PWV in newly diagnosed type 2 diabetes mellitus.

Methods

Study population and data collection

This study included 100 newly diagnosed T2DM patients (66 males and 34 females; age 58.31 ± 13.04 years) who were recruited from inpatient of the Department of Endocrinology of Nanjing Central Hospital between January 2018 and June 2018 and diagnosed according to the World Health Organization criteria. They were further divided into the thickened CIMT group (n = 41) and the non-thickened CIMT group (n = 59) according to the results of color Doppler ultrasonography. Written consents were obtained from all participants. Subjects with the previous history of cardiovascular events, diabetic ketosis, systemic acute or chronic inflammatory diseases, thyroid diseases, history of usage of statin lipid-lowering drugs, and liver, kidney or other diseases associated with lipid metabolism disorders were excluded. This study design was approved by the Ethical Committee of the Nanjing Central Hospital (2018002).

Blood sampling and biochemical tests

Venous blood samples were collected after overnight fasting. Routine biochemical analyses including serum lipids, glucose, and uric acid were measured by enzymatic methods (Chemistry Analyzer Au2700, Olympus Medical Engineering Company, Japan). The white blood cell count (WBCC), neutrophil, lymphocyte, and monocyte differentials were determined using an automated blood cell counter (Beckman Coulter Ireland Inc Mervue, Galway, Ireland). The concentration of high-sensitivity C-reactive protein (hs-CRP) was determined using immunoturbidimetry. Serum insulin level was assessed by a specific time-resolved fluoro-immunoassay on an Auto DELFIA. Body weight, height, waist circumference (WC) and blood pressure were measured in accordance with international standards.

PCSK9 ELISA

To measure serum concentration of PCSK9, fasting blood sample was centrifuged at 4 °C (3,000 rpm, 15 min), and serum samples were separated and stored at −80 °C until analysis. Serum PCSK9 levels were measured by a sandwich ELISA assay as previously described [11].

Measurement of CIMT

The CIMT was measured by echo color Doppler ultrasonography. All measurements were performed and interpreted by experienced ultrasonographers. The CIMT was evaluated as the distance between the lumen-intima interface and the media-adventitia interface. Intima and media thicknesses were measured as the distance from the main edge of the first to the main edge of the second echogenic line. The mean value of CIMT in the right and left carotid arteries was used for the statistical analyses. Thickened IMT was characterized by the largest IMT (≥ 1.0 mm) in the common carotid artery on the left or right sides [12].

Measurement of ba-PWV

The ba-PWV value was measured by VP-1000 automated PWV/ABI analyzer after resting for 15−30 min. Ba-PWV was measured from the ascending point of right brachial

pulse volume recorder to the ascending point of each ankle pulse volume recorder. We adopted the average value of left and right ba-PWV for the statistical analyses.

Statistical analysis and calculations

Data are expressed as mean $\pm$ SD. Differences between two groups were tested by student t-tests. Differences among three groups were tested by one-way ANOVA with Bonferroni correction for pair wise comparisons. The correlations between parameters were analyzed by Pearson's correlation (parametric data) and Spearman rank correlation (non-parametric data). Multiple linear regression analyses were performed to determine the factors that were independent of the CIMT or ba-PWV, and factors that were not significant in the univariate analysis were excluded from this analysis. The binary logistic regression analysis was performed to explore the relationship between serum PCSK9 levels and early atherosclerosis status (0 = the non-thickened CIMT group, 1 = the thickened CIMT group). Data were analyzed using SPSS18.0 statistical software, with significance defined as p < 0.05 (two-sided). HOMA-IR = fasting blood glucose (mmol/L) × fasting insulin (uIU/ml)/22.5; BMI = weight (Kg)/height (m)^2.

Results

Characteristics and serum PCSK9 levels in newly diagnosed T2DM participants

Clinical and biochemical characteristics of the participants are listed in Table 1. Compared with patients in the non-thickened CIMT group, patients in the thickened CIMT group were older and had higher systolic blood pressure (SBP), LDL-C, serum PCSK9 levels, CIMT, ba-PWV, fasting blood glucose (FBG), fasting insulin (FINS), HOMA-IR, WBCC and hs-CRP (all P < 0.05).

The relationships between early atherosclerosis makers, serum PCSK9 levels and other metabolic factors

Table 2 showed that CIMT was positively correlated to age, SBP, total cholesterol (TC), LDL-C, FBG, FINS, HOMA-IR, serum PCSK9 levels, WBCC and hs-CRP (P < 0.05) in newly diagnosed T2DM patients. Nevertheless, we did not observe the statistically significant relationship between CIMT and BMI, WC, diastolic blood pressure (DBP), TG, HDL-C, serum uric acid, neutrophil count, lymphocyte count and monocyte count. The relationships between ba-PWV and metabolic factors, including serum PCSK9 levels were parallel to the associations between CIMT and metabolic factors.

The relationships between serum PCSK9 levels, hs-CRP, WBCC and subpopulations

To explore the relationships between serum PCSK9 levels, hs-CRP, WBCC and subpopulations, spearman correlation analyses were performed. Serum PCSK9 levels were

Table 1 General characteristics and laboratory data of newly diagnosed type 2 diabetes.

	non-thickened CIMT group (n = 59)	thickened CIMT group (n = 41)	P value
Age (years)	53.71 ± 11.97	64.93 ± 11.69	<0.01
BMI(kg/m^2)	24.74 ± 3.36	24.73 ± 2.65	0.98
WC(cm)	89.44 ± 8.09	90.61 ± 8.02	0.47
SBP(mmHg)	127.51 ± 11.44	133.71 ± 15.28	0.22
DBP(mmHg)	79.71 ± 7.73	79.98 ± 11.22	0.89
TC (mmol/L)	4.70 ± 0.80	5.03 ± 0.87	0.06
TG (mmol/L)	1.73 ± 0.99	1.80 ± 1.23	0.75
LDL-C (mmol/L)	2.54 ± 0.74	2.99 ± 0.84	0.20
HDL-C (mmol/L)	1.16 ± 0.34	1.08 ± 0.24	<0.01
UA (umol/L)	328.95 ± 83.88	324.33 ± 98.31	0.80
PCSK9 (ng/ml)	85.40 ± 27.23	133.09 ± 33.19	<0.01
CIMT(mm)	0.81 ± 0.09	1.11 ± 0.10	<0.01
ba-PWV(cm/sec)	1464.76 ± 241.19	1902.10 ± 326.59	<0.01
FBG (mmol/L)	7.83 ± 1.79	8.87 ± 2.09	<0.01
FINS(uIU/ml)	7.97 ± 3.27	12.49 ± 4.97	<0.01
HOM-IR	2.75 ± 1.05	4.92 ± 1.09	<0.01
WBCC (10^9/L)	5.89 ± 1.22	7.09 ± 1.31	<0.01
Neutrophil count (10^9/L)	3.34 ± 0.78	3.70 ± 1.15	0.06
Lymphocyte count (10^9/L)	1.98 ± 0.61	2.11 ± 0.64	0.32
Monocyte count (10^9/L)	0.41 ± 0.12	0.46 ± 0.16	0.07
hs-CRP (mg/L)	0.84 ± 0.58	1.86 ± 0.97	<0.01

Values are presented as mean ± standard deviation.
BMI, body mass index; WC, waist circumference; SBP, systolic blood pressure; DBP, diastolic blood pressure; TC, total cholesterol; TG, triacylglyceride; LDL-C, high-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; UA, serum uric acid; PCSK9, Proprotein convertase subtilisin/kexin type 9; CIMT, carotid intima-medial thickness; ba-PWV,brachial-ankle pulse wave velocity; FBG, fasting glucose; FINS, fasting insulin; WBCC: while blood cell count; hs-CRP: high sensitivity C reactive protein.

positively correlated to WBCC($r = 0.542, P < 0.001$), neutrophil count ($r = 0.231, P = 0.021$), lymphocyte count ($r = 0.237, P = 0.017$) and hs-CRP ($r = 0.662, P < 0.001$) in newly diagnosed T2DM patients. While no significant correlation of serum PCSK9 levels with the monocyte count ($r = 0.157, P = 0.120$) was found.

Multiple linear regression analyses of the relationships between early atherosclerosis makers and serum PCSK9 levels

The association between CIMT with serum PCSK9 levels remained significant ($\beta = 0.637, P < 0.001$) when age, SBP, TC, LDL-C, FBG, FINS, HOMA-IR, WBCC, and hs-CRP were further adjusted by multiple linear regression. After adjustment for other parameters such as serum lipids, WBC, and hs-CRP, serum PCSK9 levels were still positively correlated to ba-PWV ($\beta = 0.600, P < 0.001$) (Table 3).

CIMT and ba-PWV compared across the tertiles of serum PCSK9 levels

To further demonstrate the relationship between serum PCSK9 levels and CIMT, participants were stratified into

Table 2 Pearson correlation analysis of markers of early athero-sclerosis with serum PCSK9 levels and other parameters.

	CIMT		ba-PWV	
	r	P value	r	P value
Age (years)	0.357	<0.001	0.518	<0.001
BMI(kg/m^2)	−0.038	0.706	−0.121	0.231
WC(cm)	0.032	0.775	−0.058	0.563
SBP(mmHg)	0.259	0.009	0.347	<0.001
DBP(mmHg)	0.073	0.471	−0.076	0.455
TC (mmol/L)	0.449	<0.001	0.428	<0.001
TG (mmol/L)	0.162	0.107	0.182	0.070
LDL-C (mmol/L)	0.494	<0.001	0.344	<0.001
HDL-C (mmol/L)	0.003	0.980	−0.011	0.913
UA (umol/L)	−0.109	0.284	−0.183	0.071
FBG (mmol/L)	0.359	<0.001	0.457	<0.001
FINS(uIU/ml)	0.344	<0.001	0.226	0.024
HOMA-IR	0.387	<0.001	0.393	<0.001
PCSK9 (ng/ml)	0.716	<0.001	0.714	<0.001
WBCC (10^9/L)	0.457	<0.001	0.324	0.001
Neutrophil count (10^9/L)	0.194	0.053	−0.007	0.941
Lymphocyte count (10^9/L)	0.084	0.406	0.104	0.304
Monocyte count (10^9/L)	−0.020	0.840	0.063	0.530
hs-CRP (mg/L)	0.652	<0.001	0.531	<0.001

BMI, body mass index; WC, waist circumference; SBP, systolic blood pressure; DBP, diastolic blood pressure; TC, total cholesterol; TG, triacylglyceride; LDL-C, high-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; UA, serum uric acid; PCSK9, Proprotein convertase subtilisin/kexin type 9; CIMT, carotid intimal-medial thickness; ba-PWV,brachial-ankle pulse wave velocity; FBG, fasting glucose; FINS, fasting insulin; WBCC: while blood cell count; hs-CRP: high sensitivity C reactive protein.

three groups by the tertiles of serum PCSK9 levels in 100 newly diagnosed T2DM (tertile 1 < 83.32 ng/ml, n = 33; tertile 2 83.32–119.17 ng/ml,n = 34 and tertile 3 > 119.17 ng/ml, n = 33), and analyzed whether CIMT varied in different serum PCSK9 levels tertiles. A continuous rise in CIMT was observed along the tertiles of serum PCSK9 levels [(0.80 ± 0.13 vs 0.91 ± 0.09 vs 1.09 ± 0.17) mm, P < 0.05] (Fig. 1A). Ba-PWV was higher in patients

Table 3 Associations of markers of early atherosclerosis with serum PCSK9 levels and other parameters using multiple linear regression analysis.

Variables	CIMT		ba-PWV	
	Standardized β	P value	Standardized β	P value
Age (years)	0.253	<0.001	0.428	<0.001
SBP(mmHg)	0.020	0.741	0.142	0.019
TC (mmol/l)	−0.215	0.537	0.208	0.065
LDL-C (mmol/l)	0.280	0.010	0.188	0.082
FBG (mmol/l)	0.123	0.304	0.073	0.542
FINS(uIU/ml)	0.115	0.667	0.119	0.338
HOMA-IR	0.071	0.811	0.199	0.507
PCSK9	0.637	<0.001	0.600	<0.001
WBCC (10^9/L)	0.023	0.723	0.065	0.314
hs-CRP (mg/L)	0.100	0.208	0.009	0.907

TC, total cholesterol; LDL-C, high-density lipoprotein cholesterol; PCSK9, Proprotein convertase subtilisin/kexin type 9; CIMT, carotid intimal-medial thickness; ba-PWV,brachial-ankle pulse wave velocity; FBG, fasting glucose; FINS, fasting insulin; WBCC: while blood cell count; hs-CRP: high sensitivity C reactive protein.

belonging to the highest tertiles of serum PCSK9 levels than in those belonging to the middle and the lowest tertiles [(1944.13 ± 277.81 vs 1575.60 ± 331.60 vs 1414.54 ± 201.53) cm/sec, P < 0.05] (Fig. 1B).

The relationship between serum PCSK9 levels and early atherosclerosis by binary logistic regression analysis

According to binary logistic regression analysis, serum PCSK9 levels were positively correlated with thickened carotid IMT [OR = 1.057, 95% CI (1.034−1.081), P < 0.001] without adjustment. When other risk factors such as age, lipid levels, WBCC, and subpopulations and hs-CRP were forced in the model, serum PCSK9 levels were still significantly related to thickened CIMT [OR = 1.120,95% CI (1.041−1.204), P = 0.002] (Table 4).

Discussion

Here for the first time, we demonstrated a relation between serum PCSK9 levels and markers of early atherosclerosis in newly diagnosed T2DM. A major finding of our study is that serum PCSK9levels were positively and significantly associated with carotid IMT and ba-PWV even after correction for other confounding factors.
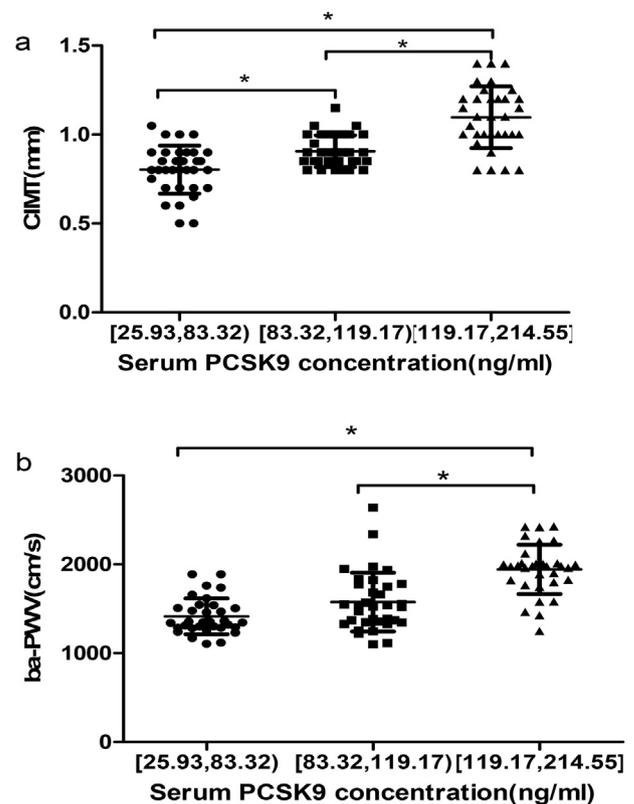

Figure 1 Markers of early atherosclerosis across the tertiles of serum Proprotein convertase subtilisin/kexin type 9(PCSK9) levels in 100 newly diagnosed type 2 diabetes (T2DM): (a) carotid intima-medial thickness (CIMT) across the tertiles of serum PCSK9 levels; (b) brachial-ankle pulse wave velocity (ba-PWV) across the tertiles of serum PCSK9 levels. *P < 0.05.

Table 4 The relationship between serum PCSK9 levels and subclinical atherosclerosis by binary logistic regression analysis.

Model	β	SE	Wald x2	OR	OR (95%CI)	P
1	0.056	0.011	14.136	1.057	1.034−1.081	<0.001
2	0.104	0.025	17.041	1.110	1.056−1.166	<0.001
3	0.113	0.037	9.317	1.120	1.041−1.204	0.002

Binary logistic regression model 1 without adjustment.
Binary logistic regression model 2 included adjustment for age, BMI, WC, SBP, DBP, TC, TG, HDL-C, LDL-C, UA, and HOMA-IR.
Binary logistic regression model 2 included adjustment for age, BMI, WC, SBP, DBP, TC, TG, HDL-C, LDL-C, UA, HOMA-IR, WBCC and subpopulations, and hs-CRP.
BMI, body mass index; WC, waist circumference; SBP, systolic blood pressure; DBP, diastolic blood pressure; TC, total cholesterol; TG, triacylglyceride; LDL-C, high-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; UA, serum uric acid; PCSK9, Proprotein convertase subtilisin/kexin type 9; WBCC: while blood cell count; hs-CRP: high sensitivity C reactive protein.

It is well known that T2DM is associated with an excessively high rate of mortality and morbidity from cardiovascular diseases, and T2DM is considered as a coronary artery disease equivalent [13]. Therefore, early detection of risk factors related to atherosclerosis is necessary to decrease mortality and morbidity risk in patients with T2DM. A number of methods have been applied for the assessment of cardiovascular risks and early atherosclerosis. In particular, CIMT has been recognized as a reliable surrogate marker of early atherosclerosis and is widely used in clinical settings, due to its advantages such as non-invasive, economical, easily applicable and reliable. In recent years, PCSK9, as a biomarker in predicting cardiovascular diseases, has gained tremendous attention. The relationship between serum PCSK9 levels and CIMT has been studied in some disorders. For example, Lee et al. showed that serum PCSK9 levels were independently associated with CIMT after adjusting for age, the lipid profile and other traditional CVD risk factors in hypertensive Korean patients [14]. A similar finding was observed by Chan et al. in asymptomatic Australian subjects [15]. Another three studies examined the association of PCSK9 concentrations with CIMT in different clinical groups. In two studies of familial hypercholesterolemia, a positive correlation between blood levels of PCSK9 and CIMT was revealed [16,17]. In a cohort of 120 healthy patients, free of manifest cardiovascular diseases), multivariable regression analysis after adjusting for gender, age, BMI, and LDL-C, plasma PCSK9 levels significantly correlated with carotid IMT in the group of obese patients [18]. Nevertheless, to be inconsistent with the results of Yang et al. [19] and Zhu et al. [20]. In the study conducted by Yang et al. a positive correlation between PCSK9 concentration and CIMT in hypertensives was found in univariate analyses, whereas such positive associations were absent after adjusting for age and there was no relationship between PCSK9 and carotid IMT in normotensives. Zhu et al also reported PCSK9 concentration was positively related to CIMT in univariate analyses, however, in backward multiple regression models, PCSK9 concentration was not an independent predictor of CIMT in 1527 middle-aged men free of vascular diseases. However, limited studies were focused on the association between serum PCSK9 levels and CIMT in patients with newly diagnosed T2DM. Our study found that patients with newly diagnosed T2DM in the thickened CIMT group had higher serum PCSK9 levels than patients in the non-thickened CIMT group. Serum PCSK9 levels were positively correlated with CIMT, which is independent of risk factors such as age and lipid profiles. When the participants with newly diagnosed T2DM were stratified into three groups by the tertiles of serum PCSK9 levels, CIMT gradually and significantly increased from the lowest to the highest tertiles of serum PCSK9 levels. Binary logistic regression analysis showed that serum PCSK9 level was an independent risk factor of early atherosclerosis. These findings suggested that serum PCSK9 levels were significantly correlated with CIMT independent of any risk factor in newly diagnosed T2DM.

Ba-PWV is a promising yet relatively simple test that measures the stiffness of both aortic and peripheral arteries, and a number of research groups propose using this technique to detect early atherosclerotic changes. Our study found that some CVD risk factors such as age, SBP, TC, LDL-C, HOMA-IR, and hs-CRP were all significantly associated with ba-PWV, which partially correlated with previous studies [21]. Han et al. found that plasma levels of PCSK9 were not correlated with cf-PWV in a community-based population [22]. However, studies about the correlation between serum PCSK9 levels and ba-PWV are limited especially in patients with newly diagnosed T2DM. Our study showed that serum PCSK9 levels were positively correlated with ba-PWV, which is independent of any risk factor such as age and lipid profile, consistent with another study in low-risk obese patients [18].

It is now well-recognized that PCSK9 plays a key role in atherogenesis via LDLR dependent pathways. Circulating PCSK9 increases the degradation of LDLR on the surface of hepatocytes, thus decreases the uptake of LDL-C via LDLR, leading to the increase of serum LDL-C levels, which is an independent risk factor of atherosclerosis. In our study, we found that serum PCSK9 levels were positively correlated with CIMT and ba-PWV, independent of LDL-C, TG and other traditional CV risk factors. Thus, we speculated that the harmful effects of PCSK9 on atherogenesis might be mediated by mechanisms independent of LDLR pathways. Several experimental and clinical studies suggested that PCSK9 plays a potential role in inflammation. For example, Tang et al. [23] showed that PCSK9 gene interference could suppress atherosclerosis directly through decreasing vascular inflammation and inhibiting the TLR4/NF-κB signaling pathway without affecting plasma cholesterol level in high-fat diet-fed apoE knockout mice. Li et al. [24] found that plasma PCSK9 levels were significantly and independently associated with the WBCC and its subsets, suggesting a potential interaction between PCSK9 and chronic inflammation in patients with CVD. Here we speculated that the potential impacts of PCSK9 on CIMT and ba-PWV may involve mechanisms relating to

inflammatory pathways. In our study, we found that serum PCSK9 levels were positively correlated to WBCC, neutrophil count, lymphocyte count and hs-CRP in newly diagnosed T2DM patients. Interestingly, after adjustment for these inflammation makers, serum PCSK9 levels were still correlated with early atherosclerosis. Measurements of other inflammatory markers such as IL-6, TNF-α, NF-kb may further help to elucidate inflammation pathways involved in the association between PCSK9 and early atherosclerosis makers.

This study has several limitations. Firstly, it is difficult to determine whether serum PCSK9 levels have a causative effect on early atherosclerosis because of the cross-sectional design. Studies of larger subject number with a long-term follow up are warranted to provide more definitive evidence. Secondly, we did not detect other inflammatory factors such as TNF-a and IL-6, so the potential role of PCSK9 in early atherosclerosis via inflammation needs to be explored in our further research. Thirdly, it is uncertain whether these results are generalizable in other ethnic groups.

In conclusion, we firstly reported that serum PCSK9 level significantly correlated with CIMT and ba-PWV, independent of metabolic factors including serum lipid profiles and inflammation markers in newly diagnosed T2DM patients. Serum PCSK9 levels could serve as a prescriptive biomarker for early arteriosclerosis, defined by CIMT and ba-PWV, which may be widely used in newly diagnosed T2DM patients or people subjected to high risks of developing T2DM.

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Authors' contribution

WG, YG, HZ and QZ participated in the study design. WG, YG, JL (Jie Li) QP, JL (Jing Lu), XL, WZ and NX were involved in the conduct of the study and data collection. WG and YG made contributions to data analysis and results interpretation. WG, YG, HZ and QZ wrote and modified the manuscript and prepared tables and figures. All authors read and approved the final manuscript.

Disclosure of interest

The authors declare that they have no competing interest.

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