Association of serum pentraxin 3 concentrations with diabetic nephropathy

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ABSTRACT

Pentraxin 3 (PTX3), a member of a superfamily of conserved proteins, attenuates renal damage in diabetic mice. This study aims to determine whether serum PTX3 concentrations are correlated with the presence of diabetic nephropathy (DN). A total of 160 patients with type 2 diabetes mellitus (T2DM) and 54 healthy subjects were enrolled in this study. Patients with T2DM were divided into three groups in accordance with the levels of urinary albumin excretion (UAE). Serum PTX3 concentrations were determined using an ELISA kit. Serum PTX3 concentrations were significantly higher in patients with T2DM compared with the controls. Patients with T2DM with macroalbuminuria showed higher serum PTX3 concentrations compared with the other three groups. However, there were no significant differences of serum PTX3 concentrations between patients with T2DM with normoalbuminuria and microalbuminuria. Furthermore, a simple regression analysis has shown that serum PTX3 concentrations in patients with T2DM were negatively correlated with body mass index, and positively correlated with blood urea nitrogen, serum creatinine, and UAE. Serum PTX3 concentrations are correlated with DN.

Significance of this study

What is already known about this subject?

- Inflammation is a clear mechanism of diabetic nephropathy (DN).
- Pentraxin 3 (PTX3) plays an important role in inflammation.
- PTX3 attenuated renal damage in mice with DN by promoting M2 macrophage differentiation.

What are the new findings?

- Serum PTX3 concentrations were significantly higher in patients with T2DM compared with the controls.
- Patients with T2DM with macroalbuminuria showed significantly increased serum PTX3 concentrations compared with the controls and patients with T2DM with normoalbuminuria and microalbuminuria.
- Serum PTX3 concentrations in patients with T2DM were positively correlated with renal function parameters such as blood urea nitrogen, serum creatinine and urinary albumin excretion.

How might these results change the focus of research or clinical practice?

- PTX3 may be involved in the mechanism of the development and progression of DN.

INTRODUCTION

Type 2 diabetes mellitus (T2DM) represents >90% of the diabetic population in the world and contributes to 9% of global mortality corresponding to 4 million deaths per year.[1] Diabetic nephropathy (DN), one important microvascular complication of T2DM, is currently the most common cause of end-stage renal disease in developed countries.[2] Traditionally, metabolic and hemodynamic alterations caused by hyperglycemia and hypertension are believed to contribute to renal injury in people with diabetes.[3] Recently, more and more evidence suggest that inflammation plays a role in the pathogenesis of DN.[4]

Pentraxins, a member of acute-phase protein family, had a cyclic multimeric structure.[5] C reactive protein is a typical short pentraxin with response to inflammation.[5] Pentraxin 3 (PTX3), belonging to the long pentraxin group, is an acute-phase glycoprotein.[6] PTX3 is produced by monocytes, macrophages, vascular endothelial cells, fibroblasts and smooth muscle cells on inflammatory stimulants.[7] PTX3 plays a role in the initiating inflammation, inhibiting angiogenesis, promoting restenosis, and the formation of advanced atherosclerotic lesions.[8 9] Recent studies have shown the protective role of PTX3 in nephropathy. PTX3 attenuated renal damage in mice with DN by promoting M2 macrophage differentiation.[10] Therefore, it is hypothesized that PTX3 may be involved in the pathogenesis of DN development.

The aim of this study was to investigate the correlation of serum PTX3 concentrations with DN.

MATERIALS AND METHODS

Patients

This study randomly recruited a consecutive population of 160 patients with T2DM from the inpatient endocrinology department of our hospital. They were all diagnosed with T2DM according to the American Diabetic Association criteria. These patients were then divided into

three groups according to the levels of urinary albumin excretion (UAE): those with normoalbuminuria (UAE<30 mg/24 hour; n=74), those with microalbuminuria (30≤UAE≤300 mg/24 hour; n=52), and those with macroalbuminuria (UAE>300 mg/24 hour; n=34).[11] Patients with concomitant valvular heart disease, cardiomyopathy, acute renal failure, acute and chronic viral or bacterial infections, asthma, tumors, connective tissue diseases, and type 1 diabetes mellitus were excluded from this study.[11] The control group randomly enrolled a consecutive population of 54 healthy subjects who had routine medical check-up in our hospital. Those subjects had no history or clinical symptom of diabetes.

The study was approved by the Hospital Ethics Board and all patients provided written informed consent.

Measurements

Serum PTX3 concentrations were measured using ELISA (R and D Systems, Inc, Minneapolis, Minnesota, USA).

Statistical analysis

Data are expressed as means±SD or median (IQR). The differences of characteristics between three groups of patients with T2DM and control subjects were compared using χ^2 tests, one-way analysis of variance (ANOVA), or Kruskal-Wallis test. The correlation between serum PTX3 and other parameters were analyzed using simple and multiple stepwise linear regression analysis. The p Values <0.05 were considered to be statistically significant.

RESULTS

Baseline clinical characteristics of patients with T2DM and controls

As presented in table 1, higher body mass index (BMI), systolic blood pressure, diastolic blood pressure, fasting plasma glucose, homeostasis model assessment of insulin resistance, triglycerides and UAE, as well as decreased levels of high-density lipoprotein cholesterol (HDL-C) were found in patients with T2DM than those in controls. Furthermore, patients with T2DM and macroalbuminuria had higher blood urea nitrogen (BUN), creatinine (Cr) and UAE than the other three groups.

Serum PTX3 concentrations

Decreased serum PTX3 concentrations were shown in the controls compared with the case groups (figure 1). Serum PTX3 concentrations were elevated in patients with T2DM with macroalbuminuria than those in the other three groups (figure 1). Moreover, serum PTX3 concentrations showed no significant differences between patients with T2DM with microalbuminuria and normoalbuminuria (figure 1).

The association of serum PTX3 concentrations with other clinical characteristics

Simple regression analysis showed that serum PTX3 concentrations in patients with T2DM were negatively correlated with BMI (r=−0.206, p=0.009), and positively correlated with BUN (r=0.240, p=0.002), Cr (r=0.236, p=0.003), and UAE (r=0.314, p<0.001) (table 2). However, only BMI (β=−0.236, p=0.002) and UAE (β=0.242, p=0.005) remained associated with serum PTX3 concentrations after multiple stepwise regression analysis.

DISCUSSION

This study provides the first report of the association of serum PTX3 concentrations with DN which is assessed using UAE.

DN is the leading cause of end-stage renal disease in developed countries.[2] This study has shown that serum PTX3 concentrations were correlated with the presence of DN which is evaluated using UAE. This indicates that serum PTX3 might be involved in the mechanism of the development and progression of DN. Suliman *et al*[12] reported that PTX3 was independently associated with UAE. Patients with T2DM showed significantly higher serum PTX3 concentrations than control subjects.[12] In addition, patients with T2DM with more proteinuria had higher PTX3 concentrations than those with less proteinuria.[12] Serum PTX3 and proteinuria were both significantly decreased after renin angiotensin system blockade treatment.[13] Furthermore, serum PTX3 levels were positively correlated with 24 hour proteinuria.[13] Katakami *et al*[14] also reported that plasma PTX3 concentrations were an independent determinant of UAE in patients with type 1 diabetes. However, other studies are inconsistent with respect to the relationship between serum PTX3 concentrations and DN. A study performed in a Malay population showed that plasma PTX3 levels in patients with DN were decreased compared with those without DN.[15] Katakami *et al*[14] reported that patients with type 1 diabetes with albuminuria showed higher serum PTX3 concentrations than those without albuminuria; however, the differences were not significant. The explanation for these conflicting data is unclear but may be attributable to differences in disease advancement, racial differences or assays applied. Our explanation is supported by another study showing the racial differences of PTX3. Higher PTX3 was associated with lower glomerular filtration rate in blacks, but not in Hispanics, Chinese or whites.[16]

This study indicated that serum PTX3 concentrations were positively correlated with renal function parameters, such as BUN and Cr. This finding was supported by other investigations. Serum PTX3 were found to be positively associated with plasma Cr in patients with acute coronary syndromes.[17] Furthermore, an investigation performed in patients with nephropathia epidemica demonstrated that patients with higher serum PTX3 levels had higher serum creatinine levels than patients with lower serum PTX3 levels.[18] PTX3 might be involved in renal failure development and progression.

The role of PTX3 in renal injury is controversial. Some studies reported the promoting role of PTX3 in renal injury or diseases. After 24 hours of reperfusion, PTX3 knockout mice showed elevated expression of endothelial adhesion molecules.[19] This indicates that endothelial PTX3 plays an important role in the pathogenesis of ischemic acute kidney injury by promoting inflammation.[19] However, most investigations focusing on the exact role of PTX3 in renal injury showed the protective function of PTX3 in renal disease. In the same study aforementioned, the author also demonstrated that decreased endothelial expression of cell adhesion molecules was found in PTX3

Table 1 Clinical characteristics of patients with T2DM and controls

		Patients with T2DM			
	Control	Normoalbuminuria	Microalbuminuria	Macroalbuminuria	p Value
N	54	74	52	34	
Age (years)	52.57±7.49	53.74±11.44	56.48±13.82	60.32±10.78*†	0.008
Gender (male/female)	29/25	43/31	27/25	20/14	0.874
BMI (kg/m^2)	24.09±1.34	26.07±3.38*	26.34±2.97*	26.49±2.89*	<0.001
SBP (mm Hg)	121.35±11.42	137.03±24.74*	144.71±30.80*	153.41±22.54*†	<0.001
DBP (mm Hg)	76.17±7.45	83.11±13.36*	86.35±17.83*	84.56±12.21*	0.001
FPG (mmol/L)	5.14±0.42	7.13±1.89*	7.89±2.28*†	7.62±2.60*	<0.001
HOMA-IR	1.79±0.66	2.66±0.72*	3.17±0.96*	3.20±0.98*	<0.001
TG (mmol/L)	1.12±0.55	1.84±0.54*	2.48±0.76*†	2.15±0.64*	<0.001
TC (mmol/L)	5.04±0.86	5.11±1.13	5.41±1.36	5.30±0.92	0.286
HDL-C(mmol/L)	1.44±0.30	1.11±0.22*	1.09±0.18*	1.15±0.26*	<0.001
LDL-C (mmol/L)	3.20±0.73	3.42±0.93	3.64±1.03*	3.50±0.73	0.083
BUN (nmol/L)	5.01±1.06	5.15±1.31	5.46±1.83	7.99±3.18*†‡	<0.001
Cr (μmol/L)	57.19±7.64	63.92±13.76	63.51±20.74	111.41±34.66*†‡	<0.001
UAE (mg/24 hour)	1.14±0.69	16.22±4.67*	74.71±48.49*†	>300*†‡	<0.001

*Significant versus control subjects.
†Significant versus patients with T2DM with normoalbuminuria.
‡Significant versus patients with T2DM with microalbuminuria.
BMI, body mass index; BUN, blood urea nitrogen; Cr, creatinine; DBP, diastolic blood pressure; FPG, fasting plasma glucose; HDL-C, high-density lipoprotein cholesterol; HOMA-IR, homeostasis model assessment of insulin resistance; LDL-C, low-density lipoprotein cholesterol; SBP, systolic blood pressure; T2DM, type 2 diabetes mellitus; TC, total cholesterol; TG, triglycerides; UAE, urinary albumin excretion.

knockout mice after 4 hours of reperfusion.[19] Lech *et al*[20] reported that postischemic acute kidney injury was significantly aggravated in postischemic microvessels of PTX3-deficient mice. Furthermore, renal leukocyte recruitment and postischemic kidney injury were improved with PTX3 treatment after reperfusion.[20] In another study performed in mice, PTX3 treatment inhibited acute renal injury-induced interstitial fibrosis, and resulted in decreased serum creatinine level and reduced expression of collagen and smooth muscle actin.[21] In addition, PTX3 was found to attenuate renal damage by promoting M2 macrophage differentiation in mice model of hyperglycemia-induced nephropathy.[10] These results point to the important role of PTX3 in protecting or preventing the development and progression of DN. However, our study showed that patients with DN had higher serum PTX3 concentrations. In addition, serum PTX3 concentrations were positively correlated with BUN and Cr. This seems contradictory to the protective role of PTX3 in DN. This may be explained that in DN condition, more PTX3 is required to protect renal function. Therefore, serum PTX3 concentrations were higher in patients with DN than the controls.

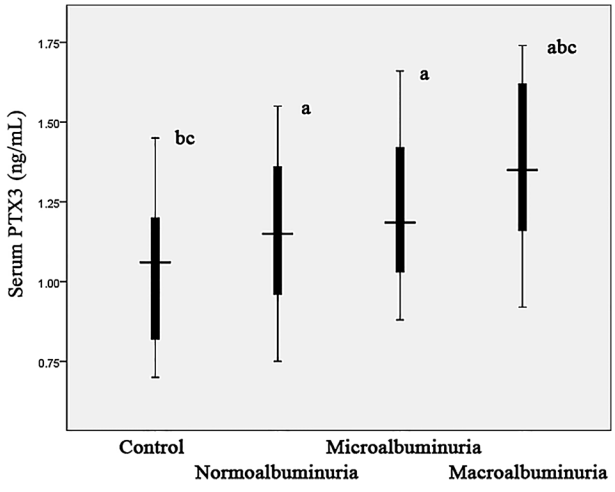

Figure 1 Serum pentraxin 3 (PTX3) concentrations in the case and control groups. [a] Significant compared with controls. [b]Significant compared with normoalbuminuria group. [c]Significant compared with microalbuminuria group.

Table 2 Simple regression analyses between serum PTX3 concentrations and various parameters

Parameters	r	p Value
Age (years)	0.049	0.536
Gender (male/female)	0.070	0.379
BMI (kg/m^2)	−0.206	0.009
SBP (mm Hg)	0.093	0.241
DBP (mm Hg)	−0.040	0.613
FPG (mmol/L)	0.052	0.514
HOMA-IR	−0.106	0.182
TG (mmol/L)	0.055	0.486
TC (mmol/L)	0.024	0.763
HDL-C (mmol/L)	0.076	0.342
LDL-C (mmol/L)	0.042	0.600
BUN (nmol/L)	0.240	0.002
Cr (μmol/L)	0.236	0.003
UAE (mg/24 hours)	0.314	<0.001

BMI, body mass index; BUN, blood urea nitrogen; Cr, creatinine; DBP, diastolic blood pressure; FPG, fasting plasma glucose; HDL-C, high-density lipoprotein cholesterol; HOMA-IR, homeostasis model assessment of insulin resistance; LDL-C, low-density lipoprotein cholesterol; SBP, systolic blood pressure; T2DM, type 2 diabetes mellitus; TC, total cholesterol; TG, triglycerides; UAE, urinary albumin excretion.

The current result demonstrated that PTX3 was negatively correlated with BMI. This is consistent with other investigations which also showed a negative correlation between PTX3 and BMI.[17 22 23] PTX3 was positively correlated with glucose transport protein 4 levels in the skeletal muscle of diabetic, obese mice.[7] This suggests that PTX3 may play a role in obesity development by promoting insulin sensitivity or glucose mechanism.

This study has several potential limitations. First, this cross-sectional study had a relatively small sample size. Further longitudinal studies with great numbers are needed. Second, the various groups appeared to have serum PTX3 levels that are all crowded around the relatively same boundaries. And the range of serum PTX3 for microalbuminuria and macroalbuminuria groups is almost identical. Therefore, our positive results should be cautioned. Third, patients with diabetes were recruited from patients admitted to the hospital; while the controls were healthy individuals participating in routine outpatient follow-up. Although not all patients with diabetes admitting to the endocrinology unit are for acute medical illnesses in China, there is still some bias about the population included. This bias may have an effect on the differences of serum PTX3 concentrations.

In short, serum PTX3 concentrations are correlated with the development of DN.

Contributors WH researched the literature and conceived the study. RW and JZ were involved in protocol development, gaining ethical approval, patient recruitment and data analysis. RW wrote the first draft of the manuscript. All authors reviewed and edited the manuscript and approved the final version of the manuscript.

Competing interests None declared.

Patient consent Obtained.

Ethics approval Ethics Board of Qilu Hospital, Shandong University.

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REFERENCES

1 Simon D. [Epidemiological features of type 2 diabetes]. *Rev Prat* 2010;60:469–73.
2 Ritz E, Rychlík I, Locatelli F, *et al*. End-stage renal failure in type 2 diabetes: a medical catastrophe of worldwide dimensions. *Am J Kidney Dis* 1999;34:795–808.
3 Satirapoj B. Nephropathy in diabetes. *Adv Exp Med Biol* 2012;771:107–22.
4 Navarro-González JF, Mora-Fernández C. The role of inflammatory cytokines in diabetic nephropathy. *J Am Soc Nephrol* 2008;19:433–42.
5 Bottazzi B, Garlanda C, Cotena A, *et al*. The long pentraxin PTX3 as a prototypic humoral pattern recognition receptor: interplay with cellular innateimmunity. *Immunol Rev* 2009;227:9–18.
6 Cieślik P, Hrycek A. Long pentraxin 3 (PTX3) in the light of its structure, mechanism of action and clinical implications. *Autoimmunity* 2012;45:119–28.
7 Abderrahim-Ferkoune A, Bezy O, Chiellini C, *et al*. Characterization of the long pentraxin PTX3 as a TNFalpha-induced secreted protein of adipose cells. *J Lipid Res* 2003;44:994–1000.
8 Mantovani A, Garlanda C, Bottazzi B, *et al*. The long pentraxin PTX3 in vascular pathology. *Vascul Pharmacol* 2006;45:326–30.
9 Shindo A, Tanemura H, Yata K, *et al*. Inflammatory biomarkers in atherosclerosis: pentraxin 3 can become a novel marker of plaque vulnerability. *PLoS ONE* 2014;9:e100045.
10 Sun H, Tian J, Xian W, *et al*. Pentraxin-3 attenuates renal damage in diabetic nephropathy by promoting M2 macrophage differentiation. *Inflammation* 2015;38:1739–47.
11 Hu W, Wang R, Li J, *et al*. Association of irisin concentrations with the presence of diabetic nephropathy and retinopathy. *Ann Clin Biochem* 2016;53:67–74.
12 Suliman ME, Yilmaz MI, Carrero JJ, *et al*. Novel links between the long pentraxin 3, endothelial dysfunction, and albuminuria in early and advanced chronic kidney disease. *Clin J Am Soc Nephrol* 2008;3:976–85.
13 Yilmaz MI, Axelsson J, Sonmez A, *et al*. Effect of renin angiotensin system blockade on pentraxin 3 levels in type-2 diabetic patients with proteinuria. *Clin J Am Soc Nephrol* 2009;4:535–41.
14 Katakami N, Kaneto H, Sakamoto F, *et al*. Plasma pentraxin 3 levels are associated with carotid IMT in type 1 diabetic patients. *Diabetes Res Clin Pract* 2013;99:185–91.
15 Abu Seman N, Witasp A, Wan Mohamud WN, *et al*. Evaluation of the association of plasma pentraxin 3 levels with type 2 diabetes and diabetic nephropathy in a Malay population. *J Diabetes Res* 2013;2013:298019.
16 Dubin R, Shlipak M, Li Y, *et al*. Racial differences in the association of pentraxin-3 with kidney dysfunction: the Multi-Ethnic Study of Atherosclerosis. *Nephrol Dial Transplant* 2011;26:1903–8.
17 Barazzoni R, Aleksova A, Carriere C, *et al*. Obesity and high waist circumference are associated with low circulating pentraxin-3 in acute coronary syndrome. *Cardiovasc Diabetol* 2013;12:167.
18 Outinen TK, Mäkelä S, Huhtala H, *et al*. High pentraxin-3 plasma levels associate with thrombocytopenia in acute Puumala hantavirus-induced nephropathia epidemica. *Eur J Clin Microbiol Infect Dis* 2012;31:957–63.
19 Chen J, Matzuk MM, Zhou XJ, *et al*. Endothelial pentraxin 3 contributes to murine ischemic acute kidney injury. *Kidney Int* 2012;82:1195–207.
20 Lech M, Römmele C, Gröbmayr R, *et al*. Endogenous and exogenous pentraxin-3 limits postischemic acute and chronic kidney injury. *Kidney Int* 2013;83:647–61.
21 Xiao Y, Yang N, Zhang Q, *et al*. Pentraxin 3 inhibits acute renal injury-induced interstitial fibrosis through suppression of IL-6/Stat3 pathway. *Inflammation* 2014;37:1895–901.
22 Witasp A, Carrero JJ, Michaëlsson K, *et al*. Inflammatory biomarker pentraxin 3 (PTX3) in relation to obesity, body fat depots and weight loss. *Obesity (Silver Spring)* 2014;22:1373–9.
23 Miyazaki T, Chiuve S, Sacks FM, *et al*. Plasma pentraxin 3 levels do not predict coronary events but reflect metabolic disorders in patients with coronary artery disease in the CARE trial. *PLoS ONE* 2014;9:e94073.