Proinflammatory cytokines predict the incidence of diabetic peripheral neuropathy over 5 years in Chinese type 2 diabetes patients: A prospective cohort study

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ABSTRACT

Background: Inflammation has been implicated in the pathogenesis of diabetic peripheral neuropathy (DPN) as suggested in various cross-sectional studies. However, more convincing prospective studies in diabetes patients are scarce. Therefore, we aimed to evaluate whether proinflammatory cytokines could predict the incidence of DPN through a prospective study with a five-year follow-up.

Methods: We followed up 315 patients with diabetes who did not have DPN, recruited from five community health centers in Shanghai in 2014, for an average of 5.06 years. Based on the integrity of blood samples, 106 patients were selected to obtain the proinflammatory cytokines. Plasma markers of proinflammatory cytokines at baseline included interleukin-6 (IL-6), tumor necrosis factor-α (TNF-α), vascular endothelial growth factor (VEGF), and intercellular adhesion molecule 1 (ICAM-1). Neuropathy was assessed by MSNI at baseline and during follow-up.

Findings: Among the 106 chosen patients, 63 developed DPN after 5.06±1.14 years of follow-up. The baseline plasma levels of TNF-α, IL-6, and ICAM-1 were higher in the neuropathic group ($p<0.05$). In multivariate models, increased plasma levels of TNF-α (hazard ratio, HR: 8.74 [95% confidence interval, CI: 1.05−72.68]; $p <0.05$) and ICAM-1 (HR 23.74 [95% CI:1.47−383.81]; $p<0.05$) were both associated with incident DPN, after adjusting for known DPN risk factors.

Interpretation: Increased plasma levels of proinflammatory factors, especially TNF-α and ICAM-1, predicted the incidence of DPN over 5 years in Chinese diabetes patients, but larger longitudinal studies are required for confirmation.

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1. Introduction

Diabetic peripheral neuropathy (DPN) is the most common diabetic microvascular complication, although the incidence and prevalence of DPN vary greatly among epidemiological studies [1,2]. The most frequent form, distal sensorimotor polyneuropathy (DSPN), affects approximately 30% of people with diabetes, while the incidence of DSPN is nearly 2% per year [3]. As the prevalence of diabetes increases, the prevalence and morbidity of DPN will also increase [4]. Despite the high disability and mortality rates of DPN, only few efficient therapeutic agents are available. Thus, early prediction and

Research in context

Evidence before this study

Evidence that systemic inflammation plays an essential role in the pathogenesis of diabetic peripheral neuropathy (DPN) is emerging from both experimental and clinical studies. However, most clinical studies available to date were cross-sectional and data from Chinese population was still lacking.

Added value of this study

This follow-up study was conducted with an aim to analyze whether plasma proinflammatory cytokines could predict the incidence of DPN over five years. We found that incidence of DPN was significantly higher in patients with higher levels of proinflammatory cytokines. Additionally, TNF-α and ICAM-1 levels were significantly associated with the incidence of DPN.

Implications of all the available evidence

Based on the results of the present study, early detection of plasma levels of proinflammatory cytokines, especially TNF-α and ICAM-1, might help predict the earliest stages of DPN, enabling early intervention.

diagnosis, enabling early intervention to delay or prevent negative consequences related to DPN, are especially important.

Evidence that systemic inflammation plays an essential role in the pathogenesis of DPN is emerging from both experimental and clinical studies. Immune cells including CD4+ T cells, CD8+ T cells, macrophages, and mast cells have already been detected in injured nerves of patients with DPN [5, 6]. Our previous study showed that interleukin-1 receptor antagonist (IL-1RA), which could inhibit inflammation, delayed the development of DPN and had a protective effect on DPN in animal models [7]. We also reported that the neutrophil-to-lymphocyte ratio (NLR), an indicator of systemic inflammation, was associated with the status of DPN even after adjusting for potential related factors [8]. However, most clinical studies available to date have been cross-sectional [9], and data from the Chinese population are still lacking. Therefore, this follow-up study was conducted with an aim to analyze whether plasma proinflammatory cytokines could predict the incidence of DPN over five years.

2. Methods

2.1. Study design and patients

In this prospective follow-up study, we aimed to examine whether proinflammatory cytokines were associated with the incidence of DPN. All participants were recruited from five community hospitals in Shanghai, People's Republic of China, in 2014 and followed up until 2019. Inclusion criteria included (1) willingness to participate in the study, (2) diagnosis of type 2 diabetes according to the 1999 World Health Organization diagnostic criteria, (3) age ≥18 years, and (4) valid Michigan Neuropathy Screening Instrument (MNSI) score. Exclusion criteria included (1) peripheral neuropathy other than diabetic origin, (2) pregnancy, (3) coincident major psychiatric disorders, (4) neurological diseases, and (5) diagnosis of DPN.

All examinations were performed by trained doctors and nurses.

2.2. Ethics

The study was approved by the Human Investigation Ethics Committee of Huashan Hospital. With a full understanding of the study, each participant signed the informed consent form voluntarily.

2.3. Demographic and clinical data

All participants completed a questionnaire to obtain information on demographic and clinical data, including age, sex, duration of diabetes, current medications, and smoking and alcohol history. Questionnaire completion was followed by a brief but standardized clinical examination involving height, weight, waist circumference, hip circumference, and blood pressure. Waist-to-hip ratio (WHR) was calculated as waist circumference divided by hip circumference. Body mass index (BMI) was calculated as weight divided by height squared (kg/m^2).

2.4. Laboratory data

A fasting venous blood sample was collected from each participant. Levels of total cholesterol (TC), triacylglycerol (TG), high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), serum creatinine (Scr), alanine aminotransferase (ALT), and aspartate aminotransferase (AST) were analyzed using an automatic analyzer (Hitachi 7600 chemical analyzer). Levels of glycated hemoglobin (HbA1c) were estimated by high-pressure liquid chromatography using an analyzer (HLC-723G7, Tosoh Corporation, Japan). The estimated glomerular filtration rate (eGFR) was calculated using the Modification of Diet in Renal Disease (MDRD) equation developed for the Chinese population. The levels of glucose, C-peptide, and insulin were measured after overnight fasting and 120 min after a 100-g steamed bread meal test, to avoid severe glucose fluctuation in patients with type 2 diabetes. Fasting plasma glucose (FPG) and postprandial plasma glucose (PPG) were analyzed using the hexokinase method and an automatic analyzer (Hitachi 7600 chemical analyzer). C-peptide and insulin levels were measured using the chemiluminescence method with an ADVIA Centaur XP automatic analyzer (Siemens Healthcare Diagnostics). The urinary albumin-to-creatinine ratio (ACR) was measured by an enzymatic method on the Roche/Hitachi cobas c system (Roche Diagnostic GmbH, Mannheim, Germany).

2.5. Proinflammatory cytokines

Proinflammatory cytokines including TNF-α, IL-6, ICAM-1 as well as VEGF were quantified using a Luminex 200 apparatus (Luminex Corp.) and Millipore multiplex immunoassay panels (Millipore).

2.6. Assessment of DPN

Neurological examination was performed using the MNSI assessment. The assessment evaluated each abnormality revealed in foot appearance (deformities, dry skin, callus, infection, or fissure), foot ulceration, ankle reflex presence, and vibratory threshold by tuning fork. Responses were assigned scores of 1 (absent), 0.5 (decreased), or 0 (normal) for each foot (maximum score = 8). The standard score for the diagnosis of DPN was >2 points as previously suggested in literature [10, 11].

2.7. Statistical analysis

All statistical analyses were performed using SAS version 9.3 (SAS Institute Inc., Cary, NC, USA). All data were checked for normality before analysis, and they are presented as means ± SD for normally distributed variables and median, 25th percentile, and 75th percentile (Median [P25, P75]) for nonnormally distributed and continuous variables. Both anthropometry and biochemical measurements were compared between groups, using Student's t-test for normally distributed continuous data and nonparametric tests for variables without a normal distribution. Categorical variables were presented as frequencies and proportions and analyzed using the χ^2 test or

Table 1

Baseline demographics and diabetic characteristics of patients grouped according to status of DPN at follow up (N = 315).

	non-DPN(n = 152)	DPN(n = 163)	P value
Age (years)	63.71±9.68	66.94±7.32	**0.0073**
DM duration(years)	12.35±8.97	15.04±10.26	**0.0144**
Male N (%)	74(52.11%)	79(48.47%)	0.9691
BMI (kg/m2)	25.01±3.29	25.30±3.40	0.4531
Waist circumference(cm)	87.17±8.63	90.40±9.90	**0.0052**
WHR	0.91±0.06	0.92±0.07	0.1237
SBP (mmHg)	129.91±15.40	133.11±15.06	**0.0350**
DBP (mmHg)	78.60±8.37	80.24±8.70	0.1233
TG (mmol/L)	1.96±1.29	1.97±1.96	0.1425
TC (mmol/L)	5.23±1.08	5.15±1.07	0.6368
LDL-C (mmol/L)	3.00±0.81	2.94±0.85	0.6588
HDL-C (mmol/L)	1.31±0.30	1.33±0.29	0.4073
FPG(mmol/L)	8.08±2.40	8.88±2.93	**0.0021**
PPG(mmol/L)	14.23±5.40	15.56±5.11	**0.0440**
HbA1c (%)	7.45±1.66	7.82±1.49	**0.0022**
AST (U/L)	24.48±8.99	24.29±8.95	0.7914
ALT (U/L)	29.56±17.45	28.45±14.43	0.9752
C-peptide 0 min (U/L)	1.84 [1.15, 2.22]	1.89 [1.06, 2.46]	0.2787
C-peptide 120 min (U/L)	6.17 [3.30, 8.13]	5.33 [3.08, 7.33]	0.3316
Insulin 0 min (U/L)	12.41 [4.60, 14.54]	14.94 [5.00, 13.23]	0.9044
Insulin 120 min (U/L)	61.90 [9.50, 65.30]	93.16 [14.83, 66.72]	0.2661
eGFR (ml/min.1.73 m2)	96.34±34.26	88.81±24.86	0.2891
ACR	35.75 [17.12, 71.00]	18.21 [8.84, 32.37]	0.0858
HOMA-IR	38.49 [18.67, 60.91]	32.14 [16.61, 57.74]	0.7022
Smoking N (%)	15 (9.87%)	18 (11.04%)	0.7338
Alcohol intake N (%)	7 (4.73%)	11 (6.75%)	0.4129

*P<0.05 between non-DPN and DPN.

Table 2

Baseline demographics and diabetic characteristics of patients grouped according to status of DPN at follow up (N = 106).

	non-DPN(n = 43)	DPN(n = 63)	P value
Age (years)	61.33±13.79	66.52±7.38	**0.0200**
DM duration(years)	12.63±7.33	14.80±10.11	0.2305
Male N (%)	22(38.60%)	35(61.40%)	0.6560
BMI (kg/m2)	25.25±3.31	25.65±3.58	0.5749
Waist circumference(cm)	87.10±11.53	89.47±10.48	0.2811
WHR	0.92±0.05	0.91±0.06	0.6522
SBP (mmHg)	131.53±13.72	134.21±12.96	0.3112
DBP (mmHg)	78.56±7.77	81.13±7.40	0.0884
TG (mmol/L)	1.68±0.79	2.11±2.77	0.5233
TC (mmol/L)	5.16±1.21	5.14±0.93	0.9617
LDL-C (mmol/L)	2.97±0.89	3.04±0.83	0.7559
HDL-C (mmol/L)	1.24±0.33	1.30±0.26	0.4728
FPG(mmol/L)	8.53±2.82	9.11±3.13	0.3289
PPG(mmol/L)	15.93±5.13	16.29±5.22	0.7393
HbA1c (%)	7.15 [6.50, 9.10]	7.30 [6.60, 8.80]	0.98
AST (U/L)	7.51 [5.22, 9.90]	8.14 [4.95, 11.16]	0.9308
ALT (U/L)	36.25 [16.30, 72.90]	28.96 [10.75, 62.70]	0.9752
C-peptide 0 min (U/L)	1.55 [1.19, 1.97]	1.92 [1.06, 2.51]	0.5375
C-peptide 120 min (U/L)	5.39 [2.55, 7.38]	5.83 [3.22, 7.33]	0.7406
eGFR (ml/min.1.73 m2)	96.34±34.26	88.81±24.86	0.2891
ACR	35.75 [17.12, 71.00]	18.21 [8.84, 32.37]	0.0858
HOMA-IR	38.49 [18.67, 60.91]	32.14 [16.61, 57.74]	0.7022

*P<0.05 between non-DPN and DPN.

Table 3

Baseline proinflammatory cytokines of patients grouped according to status of DPN at follow up (N = 106).

	non-DPN(n = 43)	DPN(n = 63)	P value
TNF-α(pg/ml)	2.45 [1.86, 3.57]	3.15 [2.73, 10.68]	**0.0161**
IL-6 (pg/ml)	0.96 [0.83, 1.31]	1.38 [0.83, 5.27]	**0.0373**
VEGF (pg/ml)	37.47 [18.23, 95.65]	54.54 [27.93, 106.80]	0.0661
ICAM-1 (pg/ml)	117,338.0 [68,574.50, 186,394.0]	156,337.0 [105,337.0, 308,943.0]	**0.0097**

*P<0.05 between non-DPN and DPN.

Fisher's exact test. Multivariate logistic regression analysis was performed to evaluate the association between DPN and proinflammatory cytokines after adjusting for other variables. P values of less than 0.05 were considered to indicate statistical significance for all analyses.

2.8. Role of the funding

3. Results

3.1. Study participants

A total of 476 diabetic patients were evaluated for eligibility in 2014, and 161 patients were excluded due to a diagnosis of DPN. After follow-up for 5.06±1.14 years, the remaining 315 participants were divided into the non-DPN group (N = 152) and the DPN group (N = 163) according to their MNSI examination scores, which were evaluated in 2019 at follow-up (Table 1). Among 315 patients without DPN, the blood samples were collected based on participants' informed preference. 209 patients' blood samples at baseline were not available. Finally, 106 patients were included in this study.

3.2. Demographics and diabetic characteristics

Demographic and diabetic characteristics of 315 patients at baseline and at follow-up, according to the DPN status, are shown in Table 1. Compared with the non-DPN group, the patients who had developed DPN at follow-up were much older (66.94±7.32 vs. 63.71±9.68, p = 0.0073), with longer durations of diabetes (15.04± 10.26 vs. 12.35±8.97, p = 0.0144) and higher levels of systolic blood pressure (SBP) (133.11±15.06 vs. 129.91±15.40, p = 0.0350). They also had higher FPG (8.88±2.93 vs. 8.08±2.40, p = 0.0021), PPG (15.56±5.11 vs. 14.23±5.40, p = 0.0440), and HbA1c (7.82±1.49 vs. 7.45±1.66, p = 0.0022) than patients without DPN. No significant differences were observed between these two groups in BMI, WHR, diastolic blood pressure (DBP), lipid levels, HOMA-IR, ALT, AST, C-peptide, insulin, HOMR-IR, eGFR, and ACR (Table 1).

The baseline characteristics of the 106 selected patients are shown in Table 2. Compared with patients in the non-DPN group, the patients who had developed DPN at follow-up were much older (66.52±7.38 vs. 61.33±13.79, p = 0.020) than patients without DPN. No significant differences were observed between these two groups in the duration of diabetes, BMI, WHR, SBP, DBP, FPG, PPG, HbA1c, lipid levels, HOMA-IR, ALT, AST, C-peptide, insulin, HOMR-IR, eGFR, and ACR (Table 2). Additionally, plasma levels of TNF-α (3.15 [2.73, 10.68] vs. 2.45 [1.86, 3.57], p = 0.0161), IL-6 (1.38 [0.83, 5.27] vs. 0.96 [0.83, 1.31], p = 0.0373), and ICAM-1 (156,337.0 [105,337.0, 308,943.0] vs. 117,338.0 [68,574.50, 186,394.0], p = 0.0097) at baseline were significantly higher in patients with DPN (Table 3). However, no significant differences were observed between the two groups in VEGF (54.54 [27.93, 106.80] vs. 37.47 [18.23, 95.65], p = 0.0661) (Table 3).

3.3. The incidence of DPN was significantly higher in patients with higher levels of proinflammatory cytokines

To further examine the association between the incidence of DPN and proinflammatory cytokines, we divided the 106 selected patients into three groups according to tertiles of tested cytokines. Compared to individuals with low levels of TNF-α (Fig. 1), IL-6 (Fig. 2), and ICAM-1 (Fig. 3), the incidence of DPN in those with high levels of the
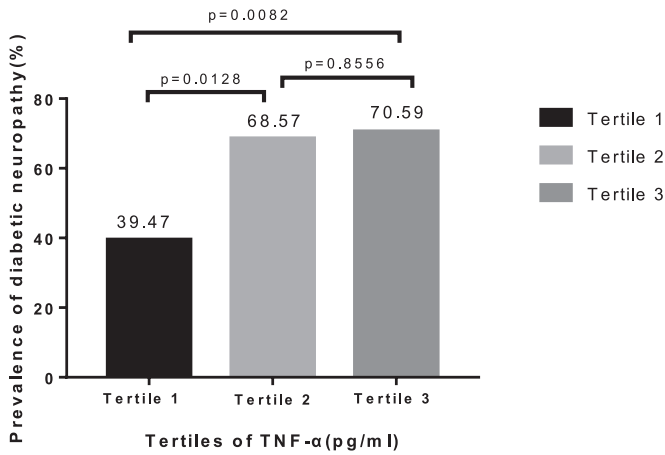
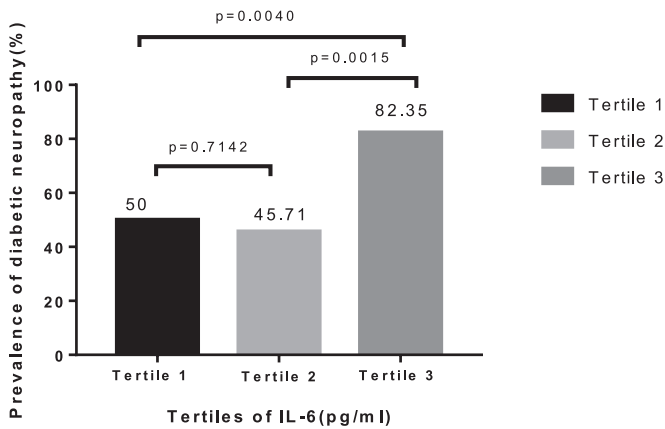**Fig 1.** Prevalence of diabetic neuropathy according to tertiles of TNF-α(%).

Fig 2. Prevalence of diabetic neuropathy according to tertiles of IL-6(%).
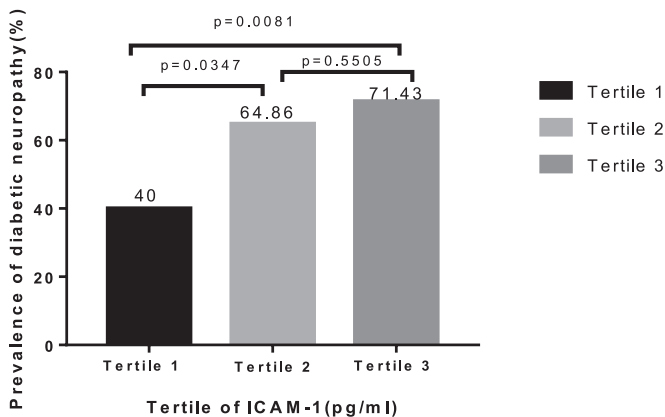

Fig 3. Prevalence of diabetic neuropathy according to tertiles of ICAM-1(%).

three cytokines was significantly higher. However, there was no significant difference in the incidence of DPN between groups graded according to the tertiles of VEGF (Fig. 4).

3.4. TNF-α and ICAM-1 levels were significantly associated with the incidence of DPN

The multiple logistic regression model shown in Table 4 demonstrates that plasma levels of TNF-α and ICAM-1 at baseline were significantly associated with the incidence of DPN at follow-up (first tertile vs. second-third tertile), even after adjusting for age, sex, BMI, duration of diabetes, HbA1c, status of dyslipidemia, and
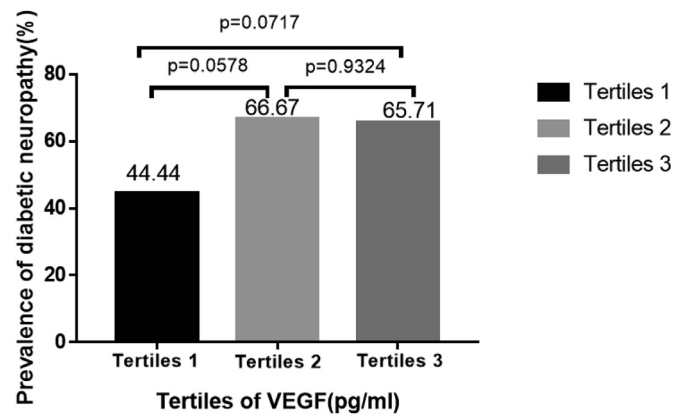

Fig 4. Prevalence of diabetic neuropathy according to tertiles of VEGF(%).

hypertension, status of smoking or drinking, physical activity and used of nonsteroidal anti-inflammatory agents at baseline.

4. Discussion

Multiple well-designed but cross-sectional studies have demonstrated higher systemic levels of proinflammatory biomarkers in patients with DPN than in those without DPN. Only a few prospective studies have sought to identify biomarkers that are linked with the incidence of DPN. Herder et al. conducted a population-based study in older adults of European descent, which suggested that TNF-α and IL-6 were predictors of incident distal sensorimotor polyneuropathy (DSPN), whereas IL-1RA and ICAM-1 were related to the progression of DSPN [12]. The study [12] mainly focused on the European population and included both prediabetic and diabetic patients. However, data on the Chinese population are still lacking. Another prospective study investigated the potential role of cell adhesion molecules in the pathogenesis of DPN [19]. The sample size of that study was relatively small (28 patients), and pro-inflammatory cytokines were not included. In our prospective study, in patients with type 2 diabetes, we found that elderly Chinese patients who developed DPN had higher levels of TNF-α, IL-6, and ICAM-1 at baseline. Interestingly, baseline VEGF levels in the group that had developed DPN at follow-up tended to be higher than that in the group without DPN. After adjusting for confounding factors, the association between IL-6 and the incidence of DPN was attenuated, while higher levels of TNF-α and ICAM-1 remained related to the incidence of DPN. It is worth mentioning that there were no marked between-group differences among 106 selected patients with known risk factors, such as glucose control, duration of diabetes, and blood pressure, probably due to the reduced sample size (Table 2). Even so, the differences in the pro-inflammatory cytokines between groups remained significant.

TNF-α is a monocyte-derived cytotoxin that may induce directed inflammatory responses [13]. As a critical regulator of immune function, TNF-α mainly binds to its receptor-TNF-α receptor 1 (TNFR1) [14]. TNF-α and TNF-α-related signaling can exacerbate vessel inflammation and oxidative stress occurring in type 2 diabetes [15], which can further contribute to the development of DPN [16]. In addition, a cross-sectional study based on observational data indicated that the TNF-α system was also activated in participants with type 1 diabetes and DPN, regardless of glycemic control and insulin resistance [17]. Due to the potential therapeutic value of TNF-α, several experimental studies have been performed. One study reported that the inhibition of TNF-α in rats with DPN using recombinant human TNFR-antibody fusion protein could relieve nerve injuries more effectively than that in untreated rats with DPN [18]. Thus, there is an experimental basis to support the predictive value of TNF-α over-activation in patients with DPN.

Table 4
Risk of DPN at follow up according to serum level of proinflammatory factors at baseline.

	OR 95%CI			
TNF-α	Model 1	Model 2	Model 3	Model 4
Tertile 1	1	1	1	1
Tertile 2	**3.35 (1.27−8.79)**	**6.82(1.79−26.01)**	5.16(0.91−29.23)	3.93(0.37−41.42)
Tertile 3	**3.68 (1.38−9.84)**	4.27(1.38−13.15)	**8.74(1.05−72.68)**	**17.29(1.32−227.03)**
IL-6	Model 1	Model 2	Model 3	Model 4
Tertile 1	1	1	1	1
Tertile 2	0.84 (0.34−2.11)	1.07(0.35−3.24)	0.62(0.12−3.23)	0.74(0.12−4.73)
Tertile 3	**4.67 (1.57−13.84)**	**4.28(1.21−15.17)**	5.99(0.54−66.25)	9.93(0.55−178.23)
VEGF	Model 1	Model 2	Model 3	Model 4
Tertile 1	1	1	1	1
Tertile 2	2.50 (0.96−6.50)	**3.48(1.03−11.83)**	3.97(0.70−22.31)	1.54(0.17−13.7)
Tertile 3	2.40 (0.92−6.25)	**3.25(1.04−10.18)**	2.77(0.45−16.98)	2.24(0.34−14.83)
ICAM1	Model 1	Model 2	Model 3	Model 4
Tertile 1	1	1	1	1
Tertile 2	**2.77 (1.07−7.20)**	**3.16(1.02−9.81)**	5.89(0.82−42.53)	17.17(0.83−353.51)
Tertile 3	**3.75 (1.38−10.17)**	**5.58(1.58−19.62)**	**23.74(1.47−383.81)**	**158.67(1.26−999.99)**

Data are odds ratios (95% confidence interval).
Model 1: unadjusted;.
Model 2: adjusted for age, gender and BMI at baseline;.
Model 3: model 2 further adjusted for duration of DM, HbA1c, status of dyslipidemia and hypertension at baseline.
Model 4: model 3 further adjusted for status of smoking or drinking, physical activity and used of nonsteroidal anti-inflammatory agents at baseline.

Elevated levels of VEGF and ICAM-1 were both biomarkers of endothelial dysfunction. In regard to ICAM-1, the results of the present study were consistent with those of previous studies. A five-year prospective study involving 28 diabetic patients suggested that ICAM-1 was strongly correlated with peroneal nerve conduction velocity and might be a predictor of the development of DPN [19]. Matthieu Roustit et al. launched a cross-sectional analysis and found that ICAM-1 were significantly associated with DPN, due to their higher neuropathy disability scores [20]. However, no significant difference were observed between two groups in VEGF at the present study, in line with the conclusion arrived by Doupis J [21], indicating the controversial effect of VEGF on the development of DPN.

Whether IL-6 is a friend or a foe for DPN is not yet clear. TNF-α can stimulate mononuclear cells secreting cytokines, including IL-6, and both TNF-α and IL-6 contribute to the development of DPN [22]. Nevertheless, one experimental study demonstrated that IL-6 treatment improved motor and sensory nerve conduction velocity, allodynia, and thermal hyperalgesia in experimental diabetes [23]. The intricate role of IL-6 may explain the discrepancy between the results of Herder et al. [12] and those of ours.

The main limitation of the present study is the lack of peripheral neurophysiologic tests or some other objective examinations, like thermal thresholds and corneal confocal microscopy for the diagnosis of DPN. However, the MNSI assessment is recommended to measure the nerve function of patients in cohort trials [3]. Therefore, we believe that the MNSI assessment is sufficient to satisfactorily characterize the research cohort. In addition, the extrapolation of the conclusions of our study is limited because only Chinese elderly type 2 diabetes patients were enrolled. Another limitation is the relatively small sample size. Thus, larger scale clinical trials, which include different age groups and various ethnic groups, are required to further validate the conclusions.

In conclusion, this prospective five-year study showed that elevated levels of proinflammatory cytokines could emerge as a useful indicator for predicting the incidence of DPN, which should be confirmed in larger sample studies. We previously reported that injection of IL-1RA improved ultrastructural injuries of diabetic nerves, indicating that early intervention was effective [7]. Based on the results of the present study, early detection of plasma levels of proinflammatory cytokines, especially TNF-α and ICAM-1, may help predict the earliest stages of DPN, enabling early intervention.

Data availability

The data are available on request from the authors.

Funding

Contributions

Hangping Zheng and Wanwan Sun wrote the manuscript. Yuanpin Zhang, Lijin Ji and Qi Zhang collected the data. Xiaoming Zhu and Xiaoxia Liu analyzed the data. Qian Xiong, Hongying Ye and Yiming Li revised the study and manuscript. Bin Lu and Shuo Zhang conducted the study design and quality control. All authors read and approved the final manuscript.

Declaration of Competing Interest

The authors declare that there is no conflict of interest.

Supplementary materials

Supplementary material associated with this article can be found in the online version at doi:10.1016/j.eclinm.2020.100649.

References

[1] Ziegler D, Papanas N, Vinik AI, et al. Epidemiology of polyneuropathy in diabetes and prediabetes.[J]. Handb Clin Neurol 2014;126:3–22.

[2] Bin Lu, Ji Hu, Jian Wen, et al. Determination of peripheral neuropathy prevalence and associated factors in Chinese subjects with diabetes and pre-diabetes - ShangHai diabetic neuropathy epidemiology and molecular genetics study (SH-DREAMS).[J]. PLoS ONE 2013;8:e61053.

[3] Pop-Busui R, Boulton AJ, Feldman EL, et al. Diabetic neuropathy: a position statement by the American diabetes association.[J]. Diabetes Care 2017;40(1):136–54.

[4] Chamberlain James J, Kacie Doyle-Delgado, Lacie Peterson, et al. Diabetes technology: review of the 2019 American diabetes association standards of medical care in diabetes.[J]. Ann Intern Med 2019;171:415–20.

[5] Schrøder H D, Olsson T, Solders G, et al. HLA-DR-expressing cells and T-lymphocytes in sural nerve biopsies.[J]. Muscle Nerve 1988;11:864–70.

[6] Cornblath D R, Griffin D E, Welch D, et al. Quantitative analysis of endoneurial T-cells in human sural nerve biopsies.[J]. J Neuroimmunol 1990;26:113–8.

[7] Zheng Hangping, Han Ling, Ji Lijin, et al. The preventive effect of IL-1beta antagonist on diabetic peripheral neuropathy.[J]. Endocr Metab Immune Disord Drug Targets 2020;20:753–9.

[8] Siying Liu, Hangping Zheng, Xiaoming Zhu, et al. Neutrophil-to-lymphocyte ratio is associated with diabetic peripheral neuropathy in type 2 diabetes patients.[J]. Diabetes Res Clin Pract 2017;130:90–7.

[9] Bönhof Gidon J, Christian Herder, Alexander Strom, et al. Emerging biomarkers, tools, and treatments for diabetic polyneuropathy.[J]. Endocr Rev 2019;40:153–92.

[10] Lunetta M, Le Moli R, Grasso G, et al. A simplified diagnostic test for ambulatory screening of peripheral diabetic neuropathy.[J]. Diabetes Res Clin Pract 1998;39:165–72.

[11] Feldman E L, Stevens M J, Thomas P K, et al. A practical two-step quantitative clinical and electrophysiological assessment for the diagnosis and staging of diabetic neuropathy.[J]. Diabetes Care 1994;17:1281–9.

[12] Herder C, Kannenberg JM, Huth C, et al. Proinflammatory cytokines predict the incidence and progression of distal sensorimotor polyneuropathy: KORA F4/FF4 study [J]. Diabetes Care 2017;40(4):569–76.

[13] Kriegler M, Perez C, DeFay K, et al. A novel form of TNF/cachectin is a cell surface cytotoxic transmembrane protein: ramifications for the complex physiology of TNF.[J]. Cell 1988;53:45–53.

[14] Roman Fischer, Olaf Maier. Interrelation of oxidative stress and inflammation in neurodegenerative disease: role of TNF.[J]. Oxid Med Cell Longev 2015;2015:610813.

[15] Jiyeon Yang, Yoonjung Park, Hanrui Zhang, et al. Role of MCP-1 in tumor necrosis factor-alpha-induced endothelial dysfunction in type 2 diabetic mice.[J]. Am J Physiol Heart Circ Physiol 2009;297:H1208–16.

[16] Kilo S, Berghoff M, Hilz M, et al. Neural and endothelial control of the microcirculation in diabetic peripheral neuropathy.[J]. Neurology 2000;54:1246–52.

[17] González-Clemente JM, Mauricio D, Richart C, et al. Diabetic neuropathy is associated with activation of the TNF-α system in subjects with type 1 diabetes mellitus[J]. Clin Endocrinol (Oxf) 2005;63:525–9.

[18] Xiaohong Shi, Yinghui Chen, Lubna Nadeem, et al. Beneficial effect of TNF-α inhibition on diabetic peripheral neuropathy.[J]. J Neuroinflammation 2013;10:69.

[19] Jude E B, Abbott C A, Young M J, et al. The potential role of cell adhesion molecules in the pathogenesis of diabetic neuropathy.[J]. Diabetologia 1998;41:330–6.

[20] Matthieu Roustit, Jordan Loader, Carly Deusenbery, et al. Endothelial dysfunction as a link between cardiovascular risk factors and peripheral neuropathy in diabetes.[J]. J. Clin. Endocrinol Metab 2016;101:3401–8.

[21] John Doupis, E Lyons Thomas, Szuhuei Wu, et al. Microvascular reactivity and inflammatory cytokines in painful and painless peripheral diabetic neuropathy.[J]. J Clin Endocrinol Metab 2009;94:2157–63.

[22] Xia Li, Ju Zhu, Na Liu, et al. TNF-alpha in peripheral neuropathy patients with impaired glucose regulation.[J]. J Diabetes Res 2017;2017:7024024.

[23] Cotter M A, Gibson T M, Nangle M R, et al. Effects of interleukin-6 treatment on neurovascular function, nerve perfusion and vascular endothelium in diabetic rats.[J]. Diabetes Obes Metab 2010;12:689–99.