Correlation Between High-sensitivity C Reactive Protein and Local Arterial Stiffness Measured by Radio Frequency Echotracking System in Type 2 Diabetic Patients

Azri Nurizal, Dono Antono, Ika Prasetya Wijaya, Hamzah Shatri

Department of Internal Medicine, Faculty of Medicine Universitas Indonesia – Cipto Mangunkusumo Hospital, Jakarta, Indonesia.

Correspondence mail:
Division of Cardiology, Department of Internal Medicine, Faculty of Medicine Universitas Indonesia – Cipto Mangunkusumo Hospital. Jl. Diponegoro no. 71, Jakarta 10430, Indonesia. email: azri.nurizal@yahoo.com.

ABSTRAK

Tujuan: *untuk mengetahui hubungan antara kadar hsCRP dan kekakuan arteri pada pasien diabetes melitus tipe 2.* ***Metode:*** *melalui studi potong lintang, dilakukan pemeriksaan kadar hsCRP dan derajat kekakuan arteri karotis pada 40 pasien dengan diabetes melitus tipe 2. Kekakuan arteri karotis kommunis diperiksa berdasarkan radio frequency echotracking system untuk menentukan pulse wave velocity (PWV) atau kekakuan arteri karotis lokal (carotid-PWV).* ***Hasil:*** *nilai median hsCRP pada penelitian ini adalah 4,5 (0,2-18,9) mg/L dan nilai rata-rata kekakuan arteri karotis adalah 8,8±1,7 m/detik. hsCRP berkorelasi kuat dengan karotid-PWV (r=0,503, P=0,001). Korelasi hsCRP dengan karotid-PWV ini tetap terlihat setelah dilakukan koreksi terhadap umur, indeks masa tubuh dan mean arterial pressure (r = 0,450, P = 0,005).* ***Kesimpulan:*** *hsCRP berkorelasi positif sedang dengan kekakuan arteri pada pasien diabetes melitus tipe 2.*

Kata kunci: *kekakuan arteri, C-reaktif protein.*

ABSTRACT

Aim: *to identify the correlation between hsCRP and arterial stiffness in type-2 diabetic patients.* ***Methods:*** *a cross-sectional study was conducted to assess the plasma levels of hsCRP and carotid arterial stiffness in 40 patients with type-2 diabetes mellitus. The common carotid artery stiffness was evaluated using radio frequency echo-tracking system to determine the local carotid pulse wave velocity (carotid-PWV).* ***Results:*** *median value of hsCRP in this study was 4.5 (0.2 to 18.9) mg/L and the average value of local carotid stiffness was 8.8±1.7 m/sec. High sensitive CRP showed a strong correlation with carotid-PWV (r=0.503, P=0.001). hsCRP level was independently associated with carotid-PWV after adjustment for age, body mass index, and mean arterial pressure (r=0.450, P=0.005).* ***Conclusion:*** *hs-CRP has moderate positive correlation with arterial stiffness in patients with type-2 diabets mellitus.*

Key words: *arterial stiffness, C-reactive protein.*

INTRODUCTION

Cardiovascular disease (CVD) is the main cause of morbidity and mortality in patients with type-2 diabetes mellitus (T2DM).[1] Diabetes mellitus is a disease that can activate an acute-phase reactant, such as C-reactive protein (CRP), which is considered as a predictor of risk for type-2 DM.[2] It has been reported that assessment of inflammatory markers such as high-sensitivity C-reactive protein (hsCRP) is an essential method to identify individuals at risk for developing cardiovascular events.[3]

Increase in arterial stiffness (AS) is demonstrated by increased arterial pulse wave velocity (PWV). Some epidemiological studies suggest that the increased AS can predict cardiovascular (CV) mortality and morbidity independent of other CV risk factors.[4] The Framingham Heart Study demonstrates that measurement of PWV may be helpful to achieve target of preventing the development of CV events in those who have some risk factors.[5]

Pulse wave velocity is the speed of pulse wave going out through the aorta.[6,7] Carotid-femoral pulse wave velocity (cf-PWV) has been established as the gold standard evaluation for arterial stiffness. However, the cf-PWV measurement may not reflect the real pathophysiological condition of AS. The distance between carotid and femoral is measured manually and it may be different from the real length of arterial line due to anatomical variety.[8] Stenosis or narrowing in aortic, iliac, and proximal femoral arteries may weaken and inhibit the pressure wave. Central obesity, particularly in men and women with big-sized breasts, may cause the distance measurement to be inaccurate.[9] Advances in ultrasonography have brought a new development of AS local measuring method using the radio frequency (RF) echotracking system; therefore, the technical problems in regional measurement can be overcome. The RF echotracking system can measure the change of diameter on altered pressure and volume against the vascular wall, which allow local measurement of PWV; thus, it may overcome drawbacks and limitations of regional AS measurement.[9,10]

Available reports on arterial stiffness are hitherto always use regional measurement.[11-19] The present study is the first study observing the correlation between hsCRP and AS through local measurement using radio frequency (RF) echotracking system. The aim of the study was to identify the correlation between hsCRP levels and arterial stiffness in patients with type 2 diabetes mellitus.

METHODS

This was a cross-sectional study on 40 type-2 diabetic patients aged between 40-60 year who willing to participate in this study and visited our department between March and May 2014 using consecutive sampling. Subjects with heart rhythm disorder, heart failure, carotid arterial stenosis, cancer, infectious disease/acute inflammation, renal dysfunction (GFR<60 ml/minutes/1.73 m^2) and those who were unable to lay down or disturbing local cervical problems or with contraindication for physical examination (mass, ulcer, cervical fixation, cervical osteoarthritis, etc) were excluded from the study.

Blood samples were taken from antecubital vein after overnight fasting. All patients underwent laboratory examinations including total cholesterol, triglycerides (TG), high-density lipoprotein (HDL), low-density lipoprotein (LDL), fasting blood glucose (FBG), 2-hour post-prandial blood glucose (2hPPBG), glycosylated hemoglobin (HbA1c), uric acid and hsCRP level. Measurements of arterial stiffness were performed by automatic resolution echotracking system radio frequency based on ultrasound method using ESAOTE MyLab 70 QAS machine, which utlilized linear probe of 10-12 MHz 124 RF-lines.

Measurements were performed on left and right common carotid arteries at 1 cm proximal to the bifurcation in order to avoid the effect of complex flow in carotid sinus. Scanning was performed on common carotid arterial wall through antero-lateral approach. The results of measurements were mean of 6 taps of examination. The examination was considered valid when the SD of distension was <20 μm. Results of measurement on vascular diameters and distensions combined with data of blood pressure were subsequently processed by the ultrasound machine which gave results

about carotid arterial stiffness in the form of pulse wave velocity.[6,10] Mean values of left-right carotid arteries were used as the values representing each individual. Measurement of AS using the radio frequency echotracking system had a low intraobserver variability.[10]

All statistical tests were performed using SPSS 20 for Windows. Shapiro-Wilk normality test was performed to identify the normality of data distribution. Data with normal distribution was expressed in mean value and standard deviation; while data with abnormal distribution was expressed in median, maximal and minimal value. Data transformation was conducted before performing correlation test in order to normalize the data distribution. Correlation test were performed by using Pearson or Spearman correlation test, as appropriate. In order to find a pure correlation between hsCRP level and AS, a partial correlation analysis was performed. The significance level of 95% was used.

RESULTS

Basic characteristics of participants are shown in **Table 1**. Mean value of arterial stiffness in the subjects was 8.8±1.7 m/second.

The mean value of hsCRP level in this study was 5.8 mg/L with a median of 4.5 mg/L (range: 0.2–18.9).

Bivariate analysis showed a strong correlation between hsCRP level and AS with r=0.503 and p=0.001) (**Figure 1)**. In addition, the correlation between various associated variables and arterial stiffness can be seen in **Table 2**.

There was a significant correlation between AK and age, BMI, as well as MAP. Furthermore, after a correction was done for variables of age, BMI and MAP, there was still a moderate correlation between hsCRP level and AS (r=0.450, P=0.005), with R Square value (R2) of 20.3%.

DISCUSSION

In the present study, manifestation of arterial stiffness was evaluated using carotid pulse wave velocity by local measurement method on carotid arterial wall. The mean value of arterial stiffness in the study was 8.8±1.7 m/second.

Table 1. Subject characteristics

Variables	n (%)	Mean (SD)	Median (range)
Age			53.3 (40-60)
Sex:			
- Male	14 (35)		
- Female	26 (65)		
hsCRP level (mg/L)			4.5 (0.2–18.9)
AS (PWV) (m/second)		8.8 (1.7)	
BMI (kg/m^2)		26.3 (4.7)	
SBP (mmHg)		137.7(19.2)	
DBP (mmHg)		79.0 (10.0)	
MAP (mmHg)			98.0 (81.6-131.3)
Total cholesterol (mg/dl)		197.9(55.8)	
LDL (mg/dl)			112.5 (60-268)
HDL (mg/dl)		45.3 (11.3)	
TG (mg/dl)			132.5 (30–686)
UA (mg/dl)		6.1 (2.1)	
FBG (mg/dl)			119.5 (83–383)
2hPPBG (mg/dl)			202.0 (45–521)
HbA1c (%)		8.6 (2.1)	

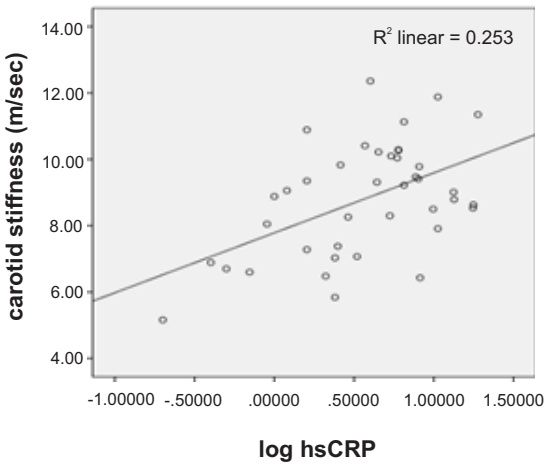

Figure 1. Correlation between log hsCRP level and arterial stiffness (r =0.503, P=0.001)

Gasner et al[20] reported that carotid PWV in CHD patients was significantly higher than the control group, i.e. 7.4±1.3 m/second vs 6.5 ±1.1 m/second (p<0.01).

Table 2. Correlation between associated variables and arterial stiffness

Risk Factors	AS (PWV)	
	r	p
Age	0.422	0.007
BMI	0.332	0.036
MAP	0.380	0.016
LDL	0.130	0.423
HbA1c	0.116	0.484

HbA1c, glycated hemoglobini; AS, arterial stiffness; LDL, Low-density lipoprotein; BMI, body mass index; MAP, mean arterial pressure; PWV, pulse wave velocity

Mean hsCRP level in this study was 5.8 mg/L with median of 4.5 (0.2–18.9). These results are similar to a report by Nandyala et al.[21] that found hsCRP level in male diabetic patients of 4.88±2.02 mg/L and 5.12±2.14 mg/L in female patients; while for the non-diabetic patients, the hsCRP level was 1.24±0.56 mg/L. Likewise on a report of Mojahedi et al.[22] hsCRP level in diabetic patients with microalbuminuria was 4.98±1.45 (0.6–7.3) mg/L, a lower level was found in diabetic patients without albuminuria, i.e. 2.82±2.10 (0–7.0) mg/L. The hsCRP levels in the present study was higher than a study conducted Mahajan et al[23] who reported hsCRP level in T2DM patients of Indian ethnicity was 1.58 mg/L (0.65–3.49) in male subjects and 2.68 mg/L (1.28–4.94) in female subjects. Previous studies in subjects with metabolic syndrome found that the mean value of hsCRP level ranged between 2.0 and 2.7 mg/L.[24-26] While Oh et al.[11] reported that the mean value of hsCRP level in female subjects with metabolic syndrome was 1.2 mg/L.

On bivariate analysis, there were a significant correlation between hsCRP levels and AS (r=0.503, P=0.001), age (r=0.422, P=0.007), BMI (r=0.332, P=0.036) and MAP (r=0.380, P=0.016). There was no significant correlation between AS and HbA1c as well as with LDL. The correlation between hsCRP levels and AS was still significant after an adjustment on variables of age, BMI and MAP (r=0.450, P=0.005).

Previously, Oh et al.[11] conducted a study in women with metabolic syndrome and found that there is a correlation between AS (measured by BaPWV) and hsCRP levels (r=0.341, P=0.016) after being adjusted for age. Yasmin et al.[12] reported that aortic PWV in healthy individuals is independently correlated with age, male sex, MAP and CRP. Likewise, brachial PWV is correlated with age, MAP, CRP, male sex, TG, HDL and smoking. Anan et al[13] who conducted a study in T2DM Japanese patients reported that in the bivariate and multivariate analysis, there were a correlation between BaPWV and hsCRP level. Tomiyama et al[14] reported that BaPWV is significantly correlated to hsCRP level in healthy Japanese individuals. Kim et al.[15] reported that hsCRP level is significantly correlated to Heart to femoral PWV (hfPWV) and brachial to ankle PWV (baPWV) in patients with non-diabetic hypertension. While London et al[16] found that there is a correlation between aortic PWV and CRP in patients with end-stage renal failure and there is an inverse correlation between CRP level and the effectiveness of antihypertension treatment. Gomez-Marcos et al[17] reported that in patients with hypertension there is a significant correlation between hsCRP and PWV after adjustment with age, cardiovascular risk factors and the use of antihypertension and anti-lipid agents. Moreover, Llaurado et al[18] showed a significant correlation between aortic PWV and hsCRP in type-1 DM patients without cardiovascular disease.

Different results are reported by Sung et al[19] who did not find a significant correlation between hsCRP and baPWV in common Chinese population. Important findings found in this study shows that in T2DM patients, there is a significant correlation between AS and hsCRP level after an adjustment with age and cardiovascular risk factors. These data indicate that AS is correlated to systemic inflammation and the inflammation plays an important role in the development of arterial wall stiffeness.

CRP may induce atherosclerosis through various ways. It activates platelet complement and induces cytokines expressions. Activated complement platelet-activating factor cytokines will stimulate leukocytes to release radical oxygen. CRP also increases generation of free-radical oxygen by monocytes and neutrophils. Radical oxygen has been known to be involved

in pathophysiology of atherosclerosis. The hypothesis of oxidative atherosclerosis depends on radical oxygen. Adhesion cellular molecules are involved in atherogenesis. CRP induces expressions of adhesion molecules in endothelial cells and monocyte chemotactic protein-1 (MCP-1). Radical oxygen produced by CRP may also increase the expression of adhesion molecules, which is modulated by free radical and oxidative stress and suppressed by antioxidants.[27] CRP works on smooth muscle cells in blood vessels and performs upregulating type of angiotensin I receptor. It also stimulates migration and proliferation of smooth muscle cells and produces reactive oxygen species (ROS).[28] Since such endothelial dysregulation may cause arterial vasoconstricition, proliferation of smooth muscle cells and vascular inflammation, increased CRP level may increase vascular stiffness and aggrevate further vascular inflammation. Therefore, this acute-phase reactant protein is not only an inflammatory marker, but may also have modulating function, which contributes to the development and evolution of atherosclerosis and increases arterial vascular stiffness.

CONCLUSION

From this study, it can be concluded that high-sensitivity C-reactive protein has a positive moderate correlation with arterial stiffness in T2DM patients.

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