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Effects of *Chlamydia pneumonia* infection on progression of coronary heart disease in elderly patients.

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Abstract

This study explored the effects of *Chlamydia pneumoniae* (Cpn) infection in elderly patients with Coronary Heart Disease (CHD), as well as related treatment strategies. Serum levels of Endothelin-1 (ET-1), Interleukin-6 (IL-6), C-reactive protein, fibrinogen, soluble vascular cell adhesion molecule-1, and cardiac troponin-1 were determined in 81 elderly CHD patients with Cpn infection (group A) and 82 elderly CHD patients without Cpn infection (group B). Patients in group A were treated with oral azithromycin; serum Cpn-immunoglobulin (Ig) M and (Ig) G levels in group A were measured before and after treatment. Levels of various indicators in group A were higher than those in group B (P<0.05), while the titres of Cpn-IgA and Cpn-IgG in group A were lower than those in group B (P<0.05). Cpn infection may result in increases in inflammatory factors. Azithromycin treatment may reduce the antibody titer, which may reduce the incidence of adverse events in elderly patients with coronary heart disease.

Keywords: *Chlamydia pneumonia* infection, Elderly patients, Coronary heart disease.

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Introduction

Coronary Heart Disease (CHD) is common in the elderly and results from atherosclerosis. Inflammatory reactions play a major role in CHD [1], and plaque inflammation is a key factor in plaque instability. The activation of inflammation and immune cells play key roles in the loss and rupture of collagen fiber caps [2,3], and plaque rupture could lead to adverse cardiovascular events [4]. *Chlamydia pneumoniae* (Cpn) infection can cause an increase in inflammatory factors such as C-Reactive Protein (CRP) and others [5], which would promote the occurrence and development of plaque inflammation through inflammatory cytokines or pneumonia-associated phospholipase D [6,7]. Cpn can cause pneumonia, bronchitis, myocarditis [8], endocarditis [9], and pericarditis [10], which can result in vascular inflammation [11], and may lead to acute cardiac and cerebrovascular events [12]. However, the correlation between Cpn and CHD in the elderly remains unclear. We retrospectively analysed clinical data of 163 elderly patients with CHD, 81 of who received azithromycin for Cpn infection, and evaluated multiple inflammatory factors to determine the effects of Cpn infection on CHD in the elderly.

Materials and Methods

Clinical data

From April 2011 to June 2013, 163 elderly patients with CHD who were admitted to our hospital were recruited for the study. All patients had undergone coronary angiography or coronary computed tomography angiography. CHD was diagnosed if at least one large coronary branch had stenosis>50%. All patients had cardiac function grade II or above, left ventricular ejection fraction ≥ 50%, and left ventricular end diastolic diameter ≤ 50 mm. Patients with malignancy, other bacterial or viral infections, toxic diseases, liver and kidney dysfunction, or immune system diseases were excluded.

Of the 163 patients with CHD, 81 with Cpn infection were placed in group A, while the other 82 patients without Cpn infection were recruited as the control group. Patients in group A were administered azithromycin (500 mg/d) orally for a total of 3 days.

Biochemical testing

Fasting venous blood samples were collected in the morning, and serum was isolated by centrifugation at 1,500 rpm for 15 min and stored at-70°C. Radioimmunoassay was used to determine serum levels of Interleukin-6 (IL-6); plasma Fibrinogen (Fg) and serum CRP levels were detected using immunonephelometry. Enzyme-Linked Immunosorbent Assay (ELISA) was used to measure levels of Endothelin-1 (ET-1) (Biotech Co., Ltd, Shanghai, China) and soluble Vascular Cell Adhesion Molecule-1 (sVCAM-1) (Fangcheng Biotechnology Co. Ltd. Beijing), according to manufacturer's instructions. Cardiac troponin-1 (c-TnI) levels were measured routinely in the clinical laboratory of our hospital. Cpn-Immunoglobulin (Ig) M and (Ig) G titers were measured in all patients the day

after admission to determine whether Cpn infection was present; Cpn-IgM and IgG titers were also measured before and 1 month after azithromycin treatment for infection in group A.

Statistical analysis

Continuous variables are presented as mean ± standard deviation and were compared by one way Analysis of Variance (ANOVA). Categorical data are presented as percentages and were compared with the chi-square or Fisher's exact test. All data were analysed using SPSS 13.0 software (SPSS, Inc., Chicago, IL, USA). P<0.05 was considered statistically significant.

Results

Study population

A total of 163 elderly patients with CHD were recruited for the study. Group A comprised 81 patients with Cpn infection. Group B comprised 82 patients without Cpn infection, who were matched for sex, age, and history of diabetes, hypertension, smoking, and low-density lipoprotein cholesterol level. Basic data are shown in Table 1, and no significant differences were detected between the groups (P>0.05, respectively).

Table 1. *Comparisons of baseline characteristics, age, disease history, diabetes, hypertension, smoking, LDL-c of the patients group and the control group.*

	Group A	Group B	Value	P
	(n=81)	(n=82)		
Male (%)	54 (66.7%)	54 (65.8%)	χ^2=0.01	>0.05
Age (year)	71.38 ± 5.28	72.09 ± 5.32	t=0.86	>0.05
CHD history (years)	5-24 (17.38 ± 5.29)	5-25 (18.03 ± 5.22)	t=0.79	>0.05
Diabetes	38 (47%)	42 (51%)	χ^2=0.30	>0.05
Hypertension	58 (72%)	54 (66%)	χ^2=0.63	>0.05
Smoking	32 (40%)	35 (43%)	χ^2=0.17	>0.05
LDL-c (mmol/l)	3.30 ± 1.14	3.18 ± 1.55	t=0.45	>0.05

Data are present as mean ± S.D. or n (%): Baseline characteristics.

Table 2. *Comparisons of ET-1, IL-6, CRP, Fg, sVCAM-1, CK, CK-MB and CTnI levels of the patients group and the control group.*

	Group A	Group B	Value	P
	(n=81)	(n=82)		
ET-1 (pg/ml)	51.23 ± 20.54	31.22 ± 10.08	t=3.526	<0.001
IL-6 (ng/L)	10.61 ± 3.46	6.72 ± 2.93	t=2.647	<0.01
CRP (mg/L)	3.54 ± 1.72	1.92 ± 1.03	t=2.161	<0.05
Fg (g/L)	2.98 ± 0.84	1.87 ± 0.92	t=2.624	0.01
sVCAM-1	27.33 ± 11.78	6.20 ± 1.38	t=3.668	<0.001
CK (IU/L)	159.67 ± 27.36	131.25 ± 30.69	t=3.258	<0.002
CK-MB (IU/L)	17.05 ± 6.32	13.68 ± 4.61	t=2.997	<0.005
CTnI (ng/ml)	0.26 ± 0.08	0.12 ± 0.03	t=2.648	<0.01

Data are shown as mean ± SD. ET-1: Endothelial-1; IL-6: Interleukin-6; CRP: C-Reactive Protein; Fg: Fibrinogen; sVCAM-1: Soluble intercellular Cell Adhesion Molecule-I, CK: Creatine Kinase, CK-MB: Creatine Kinase-MB; CTnI: Cardiac Troponin I.

Comparison of ET-1, IL-6, CRP, Fg, and sVCAM-1 levels

ET-1, IL-6, CRP, Fg, and sVCAM-1 levels in group A were significantly higher than those in group B (P<0.05, respectively) (Table 2).

Comparison of myocardial biomarkers

Creatine Kinase (CK), CK-MB, c-TnT, and c-TnI levels were significantly higher in group A than in group B (P<0.05, respectively) (Table 2).

Treatment efficacy

The titers of Cpn-IgM and Cpn-IgG in group A decreased significantly after treatment (P<0.05, respectively; Table 3).

Table 3. *Comparison of Cpn-IgM and IgG positive rates in group A.*

Group	n	Cpn-IgM (+) (%)		Cpn-IgG (+) (%)	
		Before treatment	After treatment	Before treatment	After treatment
A	81	56 (69.1)	26 (32.1)	43 (53.1)	22 (27.2)
χ^2			22.226		11.331
P			P<0.001		P=0.001

Data are shown as per centum (%). Cpn-IgM: *Chlamydia pneumoniae*-IgM; Cpn-IgG: *Chlamydia pneumoniae*-IgM.

Discussion

Cpn is an important cause of human respiratory tract infection, and the infection rate increases with age. It has been reported that Cpn infection promotes the transformation of macrophages to foam cells [13,14] and destabilizes atherosclerotic plaques [15], which can result in occlusion of coronary arteries, and induce myocardial infarction. Persistent Cpn infection may lead to arteriosclerosis [16,17] and endothelial dysfunction [18], may promote the development of atherosclerotic plaque inflammation [19], and may be involved in the pathogenesis of acute coronary syndrome [20], thus increasing the risk for CHD [21]. High levels of Heat shock protein 60 (Hsp60) and Cpn antibodies are independent risk factors for coronary atherosclerosis [22]. CHD is common in the elderly, and the morbidity associated with CHD increases with changes in life style and diet, as well as with an increase in life expectancy. Atherosclerosis, an important pathological characteristic in

CHD, is closely correlated with inflammation [23]; in other words, inflammation mediates atherosclerosis [24]. Among the inflammatory cytokines in our study, ET-1 is produced by activated macrophages and facilitates coronary plaque rapture, induces major adverse cardiac events [25], and reduces the sensitivity of coronary artery smooth muscle cells to Ca^{2+}, resulting in vascular insufficiency [26]. ET-1 recruits monocytes in the circulation and activates macrophages, leading to damage of vascular endothelial cells. ET-1 also promotes smooth muscle cell proliferation and aggravates arteriosclerosis. IL-6 can activate inflammatory cells, enhance macrophage expression [27,28], and stimulate liver cells to synthesize CRP and Fg [29]. As an independent and sensitive predictor of cardiovascular disease, IL-6 not only directly damages vascular endothelium, leading to release of abundant adhesion cells that are involved in arteriosclerosis and thrombosis, causing plaque rapture, but also induces increased sVCAM-1 production, which is associated with multiple risk factors for CHD and atherosclerotic disease [30,31]. The binding of IL-6 to monocytes or T cells can promote leukocyte adhesion and aggravate pathological damage. Fg, an acute-phase protein, can enter the artery wall via the circulation as a coagulation factor and transform into fibrin, which directly damages vascular endothelial cells, increases the incidence of thrombosis, and promotes development of atherosclerotic plaques [32]. Fg can also accumulate in the vascular wall to accelerate arteriosclerosis. In our research, we found that elderly patients with CHD complicated by Cpn infection had significantly higher levels of inflammatory factors and c-TnI than CHD patients without Cpn infection, indicating that inflammatory lesions induced by Cpn infection influence the progression of arteriosclerosis in CHD or even result in plaque rupture to cause myocardial infarction. Treatment for Cpn infection facilitates reduction in inflammatory factors, thus slowing the progression of atherosclerosis. Azithromycin is a newer macrolide, and affects protein synthesis by binding to the 50 S ribosomal subunit of pathogenic bacteria; moreover, it may improve vascular endothelial function to a certain extent [33]. Cpn-IgA and Cpn-IgG are common indicators of chronic Cpn infection. Cpn-IgA titer was markedly higher in an Acute Coronary Syndrome (ACS) group than in a non-ACS group [34]. These two indicators reflect severity of Cpn infection to some extent. In conclusion, we believe that Cpn infection may closely correlate with CHD in elderly patients; increased Cpn-induced inflammatory cytokines may promote atherosclerosis and enhance the morbidity of myocardial infarction. These patients were treated with drug therapy to control high-risk factors for atherosclerosis, and received clinical education. Treatment of chlamydial infection might improve the prognosis of elderly patients with CHD.

Conflict of Interests

No conflict of interests to declare.

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