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Original Article

The association of plasma high-sensitivity C-reactive protein level with rheumatic heart disease: The possible role of inflammation

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ABSTRACT

Background: Currently, it is not clear whether recurrent traumatic events lead to progression of rheumatic heart disease (RHD) after the incident of acute rheumatic fever or a persistent inflammatory state at the site of the valves. The aim of this study was to assess the possible association between plasma high sensitive C Reactive Protein (hs-CRP) level as an indicator of inflammation and RHD.
Materials & methods: Ninety patients with RHD and 90 healthy controls who had undergone complete echocardiographic examination were enrolled in this cross-sectional study. A score was given to each patient according to the severity of valvular involvement. Plasma hs-CRP level was checked for each patient by ELISA method twice with two-week interval, and the mean hs-CRP was calculated.
Results: The mean plasma hs-CRP level in the case group was significantly higher compared to its level in the control group (2.59 ± 4.82 and 0.55 ± 0.43 in the case and control groups respectively, $p < 0.001$). There was also a strong association between the level of plasma hs-CRP and the severity of rheumatic valvular involvement ($p < 0.001$).
Conclusion: The mean plasma hs-CRP level seems to have a significant association with RHD and its severity. Further studies are needed to determine the cause and effect relationship.

1. Introduction

Rheumatic heart disease (RHD) is a chronic progressive valvular disease which is considered as a major public health concern and is associated with 2,33,000 annual deaths in developing countries.[1] It is a delayed sequel of rheumatic fever (RF), an acute febrile inflammatory disease which affects the susceptible children after untreated streptococcus pharyngitis.[2] Nearly 30–45% of children with RF develop carditis during acute attacks with endocardial, myocardial and/or pericardial inflammation. Later in life, about half of these patients will progress to RHD with permanent valvular lesions including valve stenosis and/or regurgitation.[3] Involvement of the mitral valve is present in approximately 90% of individuals with RHD and is characterized by leaflet thickening and calcification, commissural fusion, and also chordal thickening, fusion and shortenin.[4]

It is thought that cardiac involvement during acute attack of RF is an inflammatory process, which is induced by the cross-reaction between streptococcal antigens and the valve tissue.[5] However there is an ongoing debate about the etiology of valve damage in patients with RHD.[6] These changes may be induced primarily due to the progressive valve damage as the result of constant trauma induced by the turbulent flows to a valve, which is deformed by the initial episode of RF (hemodynamic theory).

On the other hand, it may be the result of a persistent hidden valvular inflammation initiated by acute attack of RF which can lead to valve thickening and deformity, over the course of time (inflammatory theory). Assuming the credibility of the latter theory in the pathogenesis of RHD, it would be safe to expect the elevated levels of some inflammatory markers in these patients. The aim of the present study was to evaluate the possible association between plasma level of hs-CRP as a marker of inflammation and the presence of RHD and its severity.

2. Materials & methods

All patients who referred to Al-Zahra Heart Center affiliated to Shiraz University of Medical Sciences (from February 2014 to September 2015) were enrolled in the study. The case group consisted of patients who were referred to echocardiography unit due to the presence of cardiac murmurs. Some of these patients

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Table 1
Scoring protocol for grading the severity of valvular damage.

Severity	Valve stenosis	Valve Insufficiency
Mild	1 points	1 points
Moderate	2 points	2 points
Severe	3 points	3 points

were already diagnosed as having RHD and/or had a history of previous RF attack. The diagnosis of RHD was confirmed in all patients by echocardiographic findings. The control group consisted of subjects who referred to the hospital for routine checkup and in whom no organic cardiac problem was noted during the clinical, para-clinical, and echocardiographic evaluations. Patients with reactivation of RF, acute infection, inflammatory diseases, malignancy, autoimmune disorders, coronary artery disease, and trauma were excluded. In addition, patients who used acetyl salicylic acid (ASA) or other drugs, which influence CRP serum level, were also excluded from the study. Simple sampling was used for selecting the cases and the controls. All the patients filled the consent forms. The research ethics committee of Shiraz University of Medical Sciences approved the study. After the initial evaluation, 90 patients with RHD and 90 healthy subjects, as the control group, were included for further evaluations.

2.1. Data sampling

A questionnaire was designed in order to collect the subjects' data. This questionnaire contained the participants' demographic data, duration of RHD, and history of smoking. Blood pressure was measured in every patient using a mercury sphygmomanometer with a standard technique described elsewhere. Fasting blood sugar (FBS), total cholesterol, and serum triglyceride levels were also measured for each patient by nephrometry method. Standard enzyme-linked immunosorbent assay (ELISA) was used to assess the levels of hs-CRP (IBL, DB52181, Hamburg, Germany). The test was performed for each individual twice (except for missing cases); the first one at the time of echocardiographic examination and the second one two weeks later. The average of the two tests was used for data analysis. An electrocardiogram was obtained for each patient and previous medical files were checked to detect confirmed episodes of atrial fibrillation (AF).

Transthoracic echocardiography was performed in all patients in order to evaluate valvular disease (GE echo machine vivid 6). Several modalities of echocardiography including M-Mode, 2D, Doppler, and color flow mapping were used. Mitral valve (MV), aortic valve (AV) and tricuspid valve (TV) were evaluated for the presence of stenosis or regurgitation. The severity of valve stenosis and/or regurgitation was determined[7] and a score was given to every individual valvular dysfunction according to severity, as shown in Table 1. The total valvular dysfunction score for each patient was calculated by summation of all the three cardiac scores including MV, AV and TV valves' scores.

2.2. Statistical analysis

The statistical package for social science, SPSS (version 17.0, SPSS Inc., Chicago, IL, USA) was used for data analysis. Differences between mean values were analyzed by Student's unpaired *t*-test. Univariate comparisons between the groups were made with non-parametric tests: Kruskal–Wallis tests for multi-group comparisons and Mann–Whitney tests for two-group comparisons. The X^2 test and Fischer's probability test were used to compare the proportions. Differences were considered significant at $p < 0.05$.

3. Results

3.1. Patients characteristics

Ninety patients constituted the case group, which included 24 males and 66 females with the mean age of 38.32 ± 7 years. The control group included 49 males and 41 females with a mean age of 37.48 ± 8.3 years. There was no significant difference between the case and control groups in terms of age, gender, history of smoking, mean blood pressure, FBS, triglyceride, total cholesterol. The demographic and laboratory findings in the case and control groups are summarized in Table 2.

3.2. Comparing the hs-CPR levels

The mean hs-CRP was 2.59 ± 4.82 and 0.55 ± 0.43 in the case and control groups respectively with the mean hs-CRP being significantly higher in the case group ($p < 0.001$, Fig. 1).

3.3. Association of hs-CPR with severity of rheumatic valvular disease

The distributions of the rheumatic valvular scores and mean hs-CRP levels in the case group are shown in Table 3. The result showed that hs-CRP level was strongly associated with the severity of rheumatic valvular involvement ($p < 0.001$). We also compared the levels of hs-CRP between the two subgroups of patients with a score ≥ 4 and patients with a score <4. The mean hs-CRP was 2.59 ± 3.28 and 1.6 ± 1.07 in patients with a score ≥ 4 and patients with a score < 4, respectively. Mean hs-CRP level was significantly higher in the patients with score ≥ 4 ($P = 0.03$).

In patients with combined mitral stenosis and mitral regurgitation, higher scores of combined mitral stenosis and mitral regurgitation were associated with higher levels of hs-CRP ($p = 0.008$). There was also a significant association between the level of hs-CRP and severity of mitral stenosis ($p = 0.037$).

3.4. Mean hs-CRP and atrial fibrillation

Atrial fibrillation was detected in 50 (55.5%) patients, among whom 31 (34.4%) with a history of paroxysmal and 19 (21.1%) were permanent. There was no significant difference between the

Table 2
Demographic and laboratory finding of the participants.

factors	RHD group	Control group	p value	Patients with AF	Patients without AF	p value
Age	38.32 ± 7	37.48 ± 8.3	0.665	35.56 ± 6.9	39.81 ± 6.7	0.061
Female (%)	73	45	0.046	78	65	0.339
Smoking (%)	26.6	23.3	0.567	30	27.5	0.643
Hypertension (%)	17.7	20	0.487	10	10	0.89
FBS	98.52 ± 19.51	99.8 ± 21.14	0.662	94.9 ± 17.1	101.3 ± 20.8	0.087
Triglyceride	159.4 ± 61.19	157.2 ± 70.6	0.807	165.3 ± 57.2	154.8 ± 64.1	0.358
Total Cholesterol	149 ± 59.3	147.1 ± 62.2	0.821	138.4 ± 58.5	157.3 ± 59.1	0.092
HDL	35.58 ± 11.4	39.75 ± 12.41	0.86	32.76 ± 9.87	39.34 ± 12.45	0.123
LDL	124.54 ± 36.5	112.87 ± 34.75	0.25	135.65 ± 40.23	118.87 ± 35.5	0.098

RHD: Rheumatic Heart Disease; FBS: Fasting Blood Sugar.

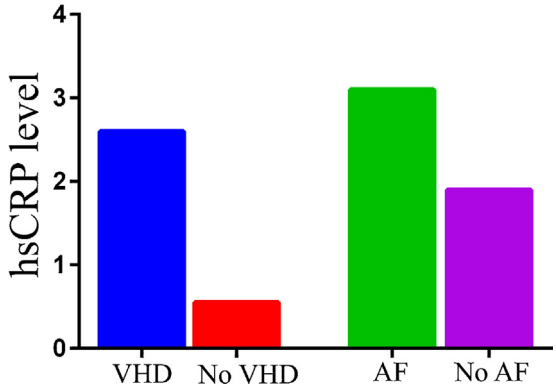

Fig. 1. Mean hs-CRP in the study groups.
Left in patients with and without rheumatic valvular heart disease (VHD) and right in rheumatic VHD patients with and without atrial fibrillation (AF).

Table 3
The frequency of valvular rheumatic score and mean hs-CPR in the case group.

Type	RHD Score	Mean hsCPR level	P Value
MS severity	1	1.02 ± 0.28	0.037
	2	1.92 ± 0.39	
	3	2.83 ± 0.66	
Combined MS and MR	1	1.91 ± 0.56	0.008
	2	2.69 ± 0.87	
	3	2.92 ± 0.48	
	4	2.25	
	5	1.96	

patients with and without AF regarding age, gender, history of smoking, FBS, triglyceride and total cholesterol (Table 2).

The mean hs-CRP level in patients with and without atrial fibrillation was 3.1 ± 6.2 and 1.9 ± 1.5, respectively. The mean hs-CRP was significantly higher in patients with AF (P = 0.042, Fig. 1). Correlation analysis revealed a moderate association between hs-CRP levels and presence of AF (p = 0.013).

4. Discussion & conclusion

Here, we have shown that in rheumatic valvular disease there is an elevated level of hs-CRP and there is a positive correlation between hs-CRP level and severity of valvular dysfunction. Also, AF was noted to be more common in patients with higher levels of hs-CRP.

The inciting event of RF is acute tonsillopharyngitis caused by group A streptococci and the responsible pathogenic mechanism is known as autoimmune reaction due to antigen mimicry between M protein epitopes of group A Streptococci and heart proteins. Auto-reactive antibodies are assumed to activate complement proteins and cause inflammation, leading to valve injury in susceptible individuals.[5] However, the pathophysiologic mechanism of progressive valvular damage in RHD is still speculative. It is hypothesized that chronic valvular damage may be the result of hemodynamic stresses applied on the injured valves or the result of an ongoing low-grade inflammatory process.[8] The role of inflammation promoting aggressions on cardiac valves is not well established, and clarification of this issue may help to decrease cardiovascular morbidity and mortality, which are associated with rheumatic valvular involvement.

It is known that inflammatory mechanisms have an important role in progression of coronary artery disease (CAD) and atheroma formation.[9] Chronic degenerative aortic valvular stenosis is another cardiac disease that is associated with presentation of local and systemic inflammation. Inflammatory cells are present in the early stage of these valvular lesions, along with lipid deposits or oxidized-cholesterol particles, which may initiate and facilitate the valves' active calcification process.[10,11] The plasma hs-CRP has already been reported as a reliable marker for oxidative stress and systemic inflammation.[12–14] The plasma hs-CRP is an independent risk predictor for coronary events, adding important prognostic information to the Framingham risk score in subjects within the intermediate-risk categories.[15,16]

In our study, the mean plasma hs-CRP level was significantly higher in patients with rheumatic valve disease compared to the control group, higher hs-CRP levels were strongly associated with higher rheumatic valvular scores. These findings may support the hypothesis that following acute RF attack, there will be a continuous chronic inflammation of cardiac valves and patients with more intense inflammatory reaction have a more rapid progression of valvular dysfunction. It can be further postulated that an abnormal antibody response leads to an autoimmune process,[17–19] which causes progressive smoldering damage to the heart valves. An earlier study showed the prolonged persistence of group A streptococcal carbohydrate antibody after the initial attack of RF; the antibody levels were reduced after surgical removal of the involved valves.[20] On the other hand, in another study it was shown that myocarditis associated with acute rheumatic fever may remain active months after the clinical disease has entered a quiescent period.[21] This could be a similar case with progressive valvular damage in RHD. The association between recurrent infection and inflammation with disease progression has also been highlighted in another study. It was shown that in North America and Europe, where the prevalence of RHD is approximately 1 case/100,000 population, patients present with severe valve obstruction in the sixth decade of life. By contrast, in Africa, with a disease prevalence of 35/100,000 and higher risk with recurrent infections, severe disease often is seen in teenagers.[22] In a study by Polat and colleagues, Pentraxin-3 (PTX3) levels were measured as an indicator of inflammatory state. Their results showed that in patients with RHD, the plasma level of PTX3 and hsCRP were increased. However, compared to hsCRP, PTX3 was more closely related to the severity of mitral valve stenosis.[23] In another study on the role of systemic and chronic inflammatory processes in the pathophysiology of rheumatic heart valve disease, the neutrophil-to-lymphocyte ratio (NLR) was measured as an indicator of systemic inflammation.[24] In this study, Akboga and colleagues showed that NLR level was significantly increased in RHD cases. They concluded that increased NLR can also be a sign of ongoing chronic inflammation in patients with RHD.[25]

Although corticosteroids and salicylates can provide symptomatic improvement in RF, the short course treatment in acute attack will not alter the long-term outcome of RHD.[26] Currently, the treatment of RHD mainly involves the prevention of acute attacks of RF, and no specific treatment is proved to prevent the progression of valvular damage. Regarding the high level of CRP in the chronic phase of the disease, the idea of long term treatment with anti-inflammatory drugs, such as aspirin could be highlighted. In addition, it has been shown that statins may also inhibit the inflammatory and non-inflammatory processes that induce the acute-phase response and can reduce CRP levels.[27] Despite the preliminary studies reporting that statins can reduce the progression of aortic stenosis in patients with chronic degenerative aortic valve stenosis through an anti-inflammatory mechanism,[28] new clinical trials have shown that there is no such effect.[29] Nevertheless, it can be speculated that statins may be useful in the management of RHD and randomized controlled trials are required to be conducted to answer this question.

In our study, we observed that RHD patients with higher levels of hsCRP are more prone to AF. Similar findings are noticed by Selcuk and colleagues, and Ucer et al.[30,31] Also, higher CRP levels can elevate the risk of MS complications. Pulimamidi et al.[32] have shown that higher hsCRP level is associated with higher risk of left atrial thrombus formation. The same finding was noticed by Karthikeyan and colleagues.[33]

Limitation of the present study is that it could not determine the cause and effect relationship between the serum levels of hs-CRP and valvular damage in RHD. Further studies should be designed to clarify this important issue.

In conclusion, the present study showed a significant association between the mean plasma hs-CRP levels and RHD. It also depicted a significant association between the mean plasma hs-CRP levels and the severity of valvular involvement. Whether treating ongoing inflammation can change future treatment strategies is a matter of future research.

Conflict of interest

None.

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