

Full Length Research Paper

Peripheral blood and C-reactive protein levels (CRP) in chronic periodontitis

Balwant Rai¹, Jasdeep Kaur², Simmi Kharb², Rajnish Jain², S. C. Anand² and Jaipaul Singh²

¹Prabhu Dayal Memorial (PDM) Dental College and Research Institute - Haryana - Bahadurgarh-India.

²BJS dental collage, India

³Post Graduate Institute of Medical Science, Rohtak, Haryana, India.

⁴UCLan, UK

Accepted 4 December, 2016

Evidence for a potential link between periodontal disease and coronary heart disease (CHD) has accumulated in recent years. C-reactive protein is potential marker of cardiovascular risk and associated with periodontal disease. CRP levels were analyzed in 26 periodontitis patients and 23 healthy controls along with hemoglobin, red blood cell count (RBC), leukocyte cell count (total and differential WBC), platelet count. Total WBC, neutrophil and platelet counts and CRP levels were raised significantly in periodontitis patients as compared to controls ($p < 0.01$) while RBC count and hemoglobin were significantly lowered in periodontitis as compared to controls ($p < 0.01$). These findings suggest an important role of CRP in development of periodontitis. Routinely screening for CRP in periodontitis patients might be an important tool to prevent heart disease.

Key words: C-reactive protein, WBC, RBC, thrombocyte, periodontitis, coronary heart disease.

INTRODUCTION

Periodontal disease, a common chronic oral inflammatory disease, is characterized by destruction of soft tissue and bone. Atherosclerosis starts early in life, however, since disease progression is usually slow, clinical symptoms or hospitalization are rare before 40 years of age. Epidemiological associations between periodontitis and cardiovascular disease have been reported (Morrison et al., 1999; Slade et al., 2003). Periodontitis and atherosclerosis have complex aetiologies, genetic and gender predispositions and might share pathogenic mechanisms as well as common risk factors. It is becoming increasingly clear that infections and chronic inflammatory conditions such as periodontitis may influence the atherosclerotic process. They may increase haemostatic variable which pronate haemostatic plugs and thrombi and rheological variables, both of which play important roles in pathogenesis of vascular disease (Fredriksson et al., 1999; Herzberg et al., 1998). The crucial casual relation might be established by prospective treatment studies, which elucidate the connection between treatment of poor dental

health and systemic inflammatory markers (Iwamoto et al., 2003; D'Aiuto et al., 2004). Several short-term intervention studies have been reported that treatment of periodontitis reduces the serum concentrations of inflammatory markers, such as C-reactive protein (CRP), TNF- α , IL-6, which are thought to be initiating factor cardiovascular disease. The aim of this study was to determine whether patients with severe periodontitis have higher plasma concentrations of established risk markers of atherosclerosis such as CRP and peripheral blood markers.

MATERIALS AND METHODS

Twenty six subjects ranging 30 - 65 years (Table 1) who had at least 6 mm loss of clinical attachment and had been referred to the Department of Periodontology, Government Dental College, PGIMS, Rohtak were selected for the study due to their severe periodontitis and were undergoing treatment for this condition. On clinical examination there is a horizontal loss of supporting tissue by more than 1/3 of root length with bleeding on probing, furcation (the area between the multiroot tooth in relation to bone height) involvements of the multi-rooted teeth and/or angular bony defects. Probing depth and attachment level measurements were performed at six sites on tooth. Patients were excluded from the study if they had alcoholic or chronic smoker. The healthy, non periodontal con-

*Corresponding author. E-mail: drbalwantraissct@rediffmail.com. Tel.: 091-9812185855.

Table 1. Clinical loss of attachment and numbers of sites involved in periodontitis assess of periodontal disease of the control and periodontal patients.

Age (in years)	No. of sites				Clinical loss of attachment (in mm)			
	Male		Female		Male		Female	
	P	N	P	N	P	N	P	N
30-47.5	(n=14)	(n=11)	(n=12)	(n=13)	(n=14)	(n=11)	(n=12)	(n=13)
	9.0±1.2	1.3±1.2*	9.2±1.3	1.4±1.2*	8.2±1.4	1.4±1.3*	8.7±1.3	1.7±1.4*
47.6-65	(n=7)	(n=5)	(n=7)	(n=5)	(n=7)	(n=5)	(n=6)	(n=7)
	9.2±1.3	1.6±1.2*	10.4±1.4	1.8±1.3*	8.4±1.2	1.7±1.3*	9.3±1.2	2.1±1.7*
	(n=7)	(n=6)	(n=5)	(n=8)	(n=7)	(n=6)	(n=6)	(n=6)

Value = Mean ± S.D.

P = periodontal patients

N = normal control

*Significant difference (p<0.05) between normal and periodontal patients.

Table 2. Total number of peripheral blood; leukocytes, neutrophil counts, total RBC counts, Hb level and C-reactive protein plasma levels in periodontitis patients and healthy controls.

Total No. of patients	Sex	Total WBC counts (x10 ⁹ /L)	Total thrombocytes counts (x10 ⁹ /L)	Neutrophil counts (x10 ⁹ /L)	Total RBC counts (x10 ¹² /L)	Hb level (g/dl)	C-reactive plasma level (mg/L)
Periodontitis group (n=26)	M (n=14)	8.3 ± 1.2a	2.5 ± 1.7a	6.5 ± 1.3a	4.4 ± 1.3a	10 ± 1.7a	4.28 ± 1.72a
		8.5 – 8.2	2.7 – 2.3	6.8 – 6.4	4.7 – 4.2	12 – 9	4.93 – 4.12
	F (n=12)	8.1 ± 1.4b	2.4 ± 1.5b	6.4 ± 1.7b	4.2 ± 1.7b	8.5 ± 1.2b	4.22 ± 1.74b
		8.3 – 7.9	2.6 – 2.1	6.6 – 6.3	4.5 – 4.1	8.7 – 8.1	4.53 – 4.12
	MF(n=26)	8.2 ± 1.5c	2.3 ± 1.5c	6.4 ± 1.8c	4.3 ± 1.6c	9.5 ± 1.7c	4.25 ± 1.73c
		7.9 – 8.5	2.1 – 2.7	6.3 – 6.8	4.1 – 4.7	8.1 – 12	4.12 – 4.93
Healthy group (n=23)	M (n=11)	6.1 ± 1.3d	2.1 ± 1.2d	6.1 ± 1.3d	4.8 ± 1.3d	11.5 ± 1.2d	1.70 ± 1.82d
		6.7 – 6.2	2.5 – 1.9	4.7 – 4.1	4.9 – 4.3	12.3 – 11.6	1.92 – 1.32
	F (n=13)	5.8 ± 1.2e	1.9 ± 1.3e	5.8 ± 1.2e	4.7 ± 1.2e	9.8 ± 1.3e	1.68 ± 1.83e
		6.3 – 5.4	2.3 – 1.7	4.3 – 3.9	4.9 – 4.2	10.2 – 8.9	1.93 – 1.52
	MF(n=24)	5.9 ± 1.3f	2.0 ± 1.2f	4.2 ± 1.3f	4.7 ± 1.7f	10.7 ± 1.3f	1.69 ± 1.82f
		5.4 – 6.7	1.7 – 2.5	3.9 – 4.7	4.2 – 4.9	8.3 – 12.3	1.32 – 1.93

Value = Mean ± S.D.

a, b, c: significant difference (p<0.05) as compared to the healthy group (d, e, f) respectively.

control group comprised 23 subjects (Table 1; range 20 - 63 years). None of the control subjects exhibited clinical signs over 5 mm or any clinical attachment loss. None of the participants was cardiovascular disease or any other ongoing general disease or infections diagnosed. Informed consent was taken from each subject. The study was proved by PDM ethical society of our collage.

In all these cases, the peripheral blood were taken drawn for routine investigation, that is, total white blood cells count, red blood cell counts, thrombocytes count and Hb level. Plasma was obtained after centrifugation at 1500x g for 10 min and stored at -4°C until analysis of C-reactive protein (Eckersall et al., 2004). Student t- test was performed using SPSS software package (version 7.0) and the significance level was set at 95 percentage confidence interval.

RESULTS AND DISCUSSION

The clinical loss of attachment and number of sites involvement were significantly higher in periodontitis as compared to normal patients and also with increasing the

age clinical loss of attachment and numbers of sites involvement were increased (Table 1) as reported in previous studies (Rai, unpublished data; Ushida et al., 2008). Total WBC, neutrophil, total thrombocytes counts and C-reactive protein (CRP) levels were increase significantly in periodontitis patients in comparison to healthy controls while RBC counts and Hb level were decreased in periodontitis in comparison to healthy controls (Table 2 and Figure 1, p<0.01). The chronic infections, such as periodontitis, are associated with increased risk for cardiovascular disease. It has been reported that higher number of leukocytes in periodontitis (Kweider et al., 1993). Subsequent studies have reported slight elevations of leukocytes in periodontitis in comparison to controls, although not always statistically significant (Fredriksson, 1999). In present study the total leukocytes counts significantly increased in periodontitis as compared to

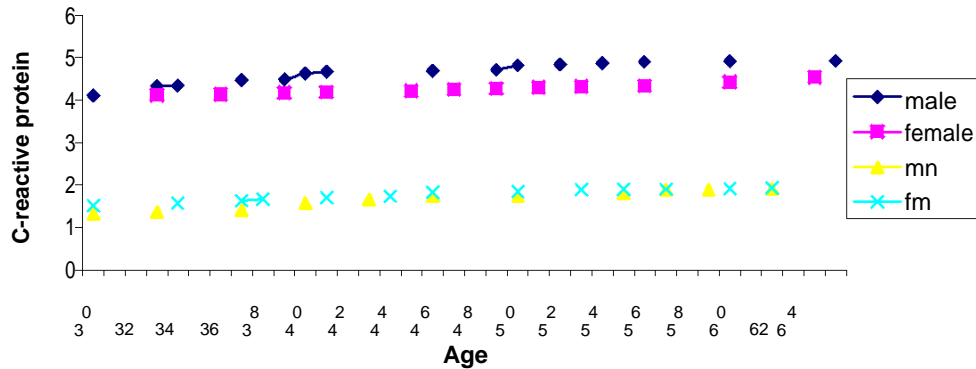


Figure 1. The level of C-reactive protein in normal and periodontitis patients in both genders in different age.

Male = periodontal patients (male)
 Female = periodontal patients (female)
 mn = normal males
 fn = normal female

healthy controls (Table 1, $p < 0.01$). A significant increase in polymorphonuclear leukocytes were also observed ($p < 0.01$, Table 1). The increase in number of leukocytes in periodontitis has been suggested to be mainly due to an increase in the number of polymorphonuclear leukocytes (Kowalik et al., 2001). Moderately elevated numbers of leukocytes have been associated with an increased risk for cardiovascular diseases (Albert and Ridker, 2004). Also, it has been suggested that the higher numbers of leukocytes increase the blood rheology. More cell numbers per unit cube made blood more viscous, and more cells may adhere to endothelial cells lining the blood vessels, therefore decrease the blood flow. Reduced blood flow could play a role in relation to cardiovascular diseases, especially in narrow or partly blocked arteries due to atherosclerotic plaque formation (Sweetnam et al., 1997).

Low hematocrit values have been reported in the periodontitis patients (Hultter et al., 2001). Lowered hemoglobin was observed in our study. Our results are consistent with those reported in literature (Fredriksson et al., 1999). Few reports are available concerning red blood cell parameters in relation to periodontitis (Salvi et al., 1997). In our study, the RBC counts were decreased significantly in periodontitis as compared to healthy ($p < 0.01$, Table 2).

Also, the platelets count were significantly increased in periodontitis as compared to healthy control (Table 2, $p < 0.01$). This increase in thrombocyte count could be due to fact that they play an integral role in innate immunity against micro-organism (Klinger et al., 2002). Certain proteins from the periodontal pathogen porphyromonas gingivalis can stimulate thrombocytes to aggregate in a similar fashion as the clotting factor thrombin (Herzberg and Weyer, 1998). It is known that inflammatory and infectious processes can result in an increase in the number of active thrombocytes; that is "reactive thrombocy-

toxis". Therefore, increase in circulating thrombocytes could occur in periodontitis patients.

The C-reactive protein (CRP) is part of the body's normal response to infection and inflammation; chronically elevated levels are associated with a heightened risk for cardiovascular disease and mortality in both, the middle aged and the elderly (Harris et al., 1999; Lower et al., 2001). CRP might also be indicator of chronic infective processes possibly correlated with risk of coronary heart disease, such as infection by chlamydia pneumonia or chronic gastric infection with helicobacter pylori (Zhu et al., 2000). The higher plasma levels of CRP were observed in the present study in case of severe periodontitis patients in comparison to healthy control (Table 2, $p < 0.01$). Recent studies (Rai, unpublished data; Ushida et al., 2008) also have reported elevated CRP levels among these with periodontitis. Conventional risk factors or conditions associated with progression of atherosclerosis include diabetes mellitus, cigarette smoking, hypertension, family history of ischaemic heart disease, hyperlipidemia, obesity, elevated plasma homocysteine, elevated CRP, menopause, male gender, nephritic syndrome and hyperthyroidism (Zhu et al., 2001). It has been well documented that CRP is a valuable marker in assessment of cardiovascular risk. And strong association of CRP with periodontal disease has been reported in literature (Zhu et al., 2000). Periodontal disease is often a chronic bacterial infection which involves inflammatory processes that also affect cardiovascular systems. Similar pathology is evident in both diseases and it is reasonable to expect some form of interaction between their pathogenic processes. Currently, American Heart Association is developing a summary on the inclusion of periodontal screening with previously established measured of risk assessment of CHD (Noack et al., 2001). There were lot of limitations of this study such important marks as CRP, alpha-TNF and interleukins (IL 6), clotting factor e.g.

prothrombin were not included in this study. Also we did not include patients with periodontitis without atherosclerosis and patients with periodontitis and atherosclerosis to prove the periodontitis is a risk factor for cardiovascular diseases. Further study is required on large scale and inflammatory factors e.g. hs-CRP, alpha-TNF and interleukins (IL 6), clotting factor e.g. prothrombin, group with periodontitis without atherosclerosis and patients with periodontitis and atherosclerosis include in study.

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