J Clin Periodontol 2014; 41: 1048–1054 doi: 10.1111/jcpe.12284

Journal of Clinical Periodontology

Periodontitis in coronary heart disease patients: strong association between bleeding on probing and systemic biomarkers

Syed Akhtar H. Bokhari[1], Ayyaz A. Khan[2], Arshad K. Butt[2], Mohammad Hanif[3], Mateen Izhar[2], Dimitris N. Tatakis[4] and Mohammad Ashfaq[1]

[1]The University of Faisalabad, Faisalabad, Pakistan; [2]Sheikh Zayed Federal Postgraduate Medical Institute & Hospital, Lahore, Pakistan; [3]Department of Statistics, National College of Business Administration & Economics, Lahore, Pakistan; [4]Division of Periodontology, College of Dentistry, Ohio State University, Columbus, OH, USA

Bokhari SAH, Khan AA, Butt AK, Hanif M, Izhar M, Tatakis DN, Ashfaq M. Periodontitis in coronary heart disease patients: strong association between bleeding on probing and systemic biomarkers. J Clin Periodontol 2014; 41: 1048–1054. doi: 10.1111/jcpe.12284.

Abstract

Aim: Few studies have examined the relationship of individual periodontal parameters with individual systemic biomarkers. This study assessed the possible association between specific clinical parameters of periodontitis and systemic biomarkers of coronary heart disease risk in coronary heart disease patients with periodontitis.

Materials and Methods: Angiographically proven coronary heart disease patients with periodontitis (n = 317), aged >30 years and without other systemic illness were examined. Periodontal clinical parameters of bleeding on probing (BOP), probing depth (PD), and clinical attachment level (CAL) and systemic levels of high-sensitivity C-reactive protein (CRP), fibrinogen (FIB) and white blood cells (WBC) were noted and analyzed to identify associations through linear and stepwise multiple regression analyses.

Results: Unadjusted linear regression showed significant associations between periodontal and systemic parameters; the strongest association (r = 0.629; $p < 0.001$) was found between BOP and CRP levels, the periodontal and systemic inflammation marker, respectively. Stepwise regression analysis models revealed that BOP was a predictor of systemic CRP levels ($p < 0.0001$). BOP was the only periodontal parameter significantly associated with each systemic parameter (CRP, FIB, and WBC).

Conclusion: In coronary heart disease patients with periodontitis, BOP is strongly associated with systemic CRP levels; this association possibly reflects the potential significance of the local periodontal inflammatory burden for systemic inflammation.

Key words: coronary heart disease; C-reactive protein; fibrinogen; periodontitis; white blood cells

Accepted for publication 15 June 2014

Conflict of interest and source of funding statement

Authors report no conflict of interest. This study was supported by Grant No: 20-685, R & D 06/1959, Higher Education Commission, Government of Pakistan.

Periodontitis, a chronic inflammatory disease of infectious nature (Williams 1990, Tatakis & Kumar 2005), has been strongly associated with elevation of systemic markers, such as C-reactive protein (CRP; Paraskevas et al. 2008, Fisher et al. 2010), fibrinogen (FIB) and white blood cells (WBC); (Kweider et al. 1993). These three biomarkers have been significantly associated with coronary heart disease (CHD); (Wang et al. 2002) and CRP is considered an independent predictor of CHD (Wang et al. 2002).

Abundant evidence supports a strong association between periodontitis and CHD (Humphrey et al.

2008, Dietrich et al. 2013, Tonetti & Van Dyke 2013) an association potentially explained by diverse biological mechanisms (Meurman et al. 2004, Kebschull et al. 2010). Such mechanisms include the inflammatory burden (Noack et al. 2001, Montebugnoli et al. 2004, Schenkein & Loos 2013), the oral microbial burden (Noack et al. 2001, Reyes et al. 2013), and haemostatic factors (Montebugnoli et al. 2004, Schenkein & Loos 2013). Interventional trials suggest that periodontal therapy, in general, results in significant reduction of systemic marker levels (D'Aiuto et al. 2004, 2006, Seinost et al. 2005, Hussain Bokhari et al. 2009, Offenbacher et al. 2009, Bokhari et al. 2012, Teeuw et al. 2014), especially in patients with systemic diseases such as CHD (Teeuw et al. 2014). Despite the evidence linking periodontitis and CHD, there are limited data on the potential association of specific periodontal clinical parameters with any individual systemic biomarker, such as CRP (Beck & Offenbacher 2002, Ebersole et al. 2002, Slade et al. 2003, Montebugnoli et al. 2004).

The purpose of the present study was to examine the possible associations between individual clinical parameters of periodontitis, that is, bleeding on probing (BOP), a marker of periodontal inflammation (Cooper et al. 1983), and probing depth (PD) and clinical attachment level (CAL), two markers of tissue destruction, and biomarkers of CHD risk (CRP, Fibrinogen, WBCs) by performing secondary analyses of the baseline data from a randomized controlled periodontal intervention trial in CHD patients with periodontitis (Bokhari et al. 2012).

Materials and Methods

Study design and study population

The present study is cross-sectional investigation of periodontal clinical parameters and inflammatory biomarkers of CHD risk performed on the baseline data of 317 subjects recruited for a randomized controlled trial (Bokhari et al. 2012). Information on study population, clinical periodontal parameters, and systemic biomarkers has been recently reported in detail (Bokhari et al. 2012) and is only summarized here. Patients with angiographically defined stable CHD (>50% stenosis of ≥1 coronary vessel) and periodontitis (whole mouth BOP > 20% of sites; ≥4 teeth with ≥1 site with periodontal pocket depth (PPD) ≥4 mm and CAL ≥3 mm at same site) were recruited and examined between July 2008 and February 2009. Amongst screened CHD patients, 354 met the inclusion criteria and 317 agreed to participate. The study was approved by the Ethics Committee of Punjab Institute of Cardiology (Lahore, Pakistan) and eligible patients provided written informed consent prior to enrolment into the study.

Eligibility criteria

General and medical eligibility criteria were as follows: aged >30 years; CHD case (according to definition above), having coronary angiography, or myocardial infarction, or coronary artery bypass grafting, or angioplasty >3 months prior to entry into study, clinically stable in the absence of any potentially confounding inflammatory condition; free of other acute or chronic systemic conditions; non-smokers (never smoker) or former smokers (did not smoke at inclusion time and had not smoked at all for at least the last 12 consecutive months); able and willing (through informed written consent) to comply with study procedures. Patients who had received medications (during last 3 months) known to affect systemic inflammatory markers (systemic steroids; non-steroidal anti-inflammatory drugs; immunosuppressants; hormone replacement therapy; contraceptives; systemic antibiotics) were excluded (Bokhari et al. 2012). Cut-off level for hypertension was defined at ≥140/≥90 mm Hg for systolic/diastolic blood pressure, respectively (Sesso et al. 2007). Oral and periodontal inclusion criteria were as follows: at least 14 natural teeth present (excluding third molars) that can be evaluated periodontally; periodontitis diagnosis (according to definition above); no mechanical/surgical periodontal therapy in the last 6 months; no acute oral diseases (mucosal lesions), oral infections, or need for immediate dental/periodontal care (e.g., necrotizing ulcerative gingivitis, extraction, endodontic therapy, etc.); no oral surgical treatment (including any non-surgical extractions) in the last 2 months. General screening was done by a trained staff nurse and systemic conditions were evaluated by two hospital cardiologists. At inclusion, patients were using medications that included anti-platelets, β-blockers, angiotensin receptor blockers, calcium channel antagonists, lipid-lowering agents, diuretics, and nitrates; medication use was recorded.

Clinical periodontal measures (Exposures)

Periodontal status was assessed through clinical parameters of BOP, PD and CAL (six sites per tooth, for all teeth present, excluding 3rd molars). Periodontal examinations were carried out by a single examiner (SAHB). Detailed descriptions for recording BOP, PD and CAL have been previously reported (Bokhari et al. 2012). Briefly, BOP was noted within 30 s of probing, while PD and CAL were measured using a UNC15 probe (Hu-Friedy, Chicago, IL, USA) and recorded to the nearest mm.

Systemic biomarkers of CHD risk (Outcome measures)

Levels of high-sensitivity CRP, FIB and WBC were analyzed from peripheral blood. Non-fasting blood samples were obtained and prepared by the hospital phlebotomist. Manufacturer instructions were followed up for blood sample storage and analysis. CRP levels (mg/l) were analyzed using a high-sensitivity assay (Cardiophase® CCRP Flex reagent kit; Dade Behring Ltd, Atterbury, Milton Keynes MK, UK); Fibrinogen (mg/l) was analyzed on a semi-automated analyzer (KC4 Delta; Trinity Biotech GmbH, Lemgo, Germany) and WBC counts ($\times 10^9$/l) were obtained on an automated haematology analyzer (KX-2; Sysmex Corp, Singapore).

Statistical analysis

Statistical analysis (SPSS v.15; IBM, Chicago, IL, USA) was performed and significance level was set at 95% ($p \leq 0.05$). All parameters (overall patient mean) were analyzed for each individual. Continuous variables were described in mean ± SD

and categorical characteristics in frequency. Subjects were categorized into tertiles of mean BOP%, PD and CAL and mean CRP, FIB and WBCs for comparison purposes. Percent (%) sites of PD (≥4 mm, ≥5 mm, ≥6 mm) and CAL (≥3 mm, ≥4 mm, ≥5 mm) for subjects were calculated to report corresponding associations.

On the basis of tertiles for each of the three periodontal parameters, subjects were grouped into (i) high periodontitis ($n = 37$): defined as any patient having BOP ≥ 45.37% and PD ≥ 3.58 mm and CAL ≥ 3.91 mm; (ii) moderate periodontitis ($n = 19$): defined as any patient having BOP = 32.80–45.36% and PD = 2.95–3.57 mm and CAL = 2.67–3.90 mm; and (iii) low periodontitis ($n = 20$): defined as any patient having BOP ≤ 32.79% and PD ≤ 2.94 mm and CAL ≤ 2.66 mm.

Forward selection procedure regression analyses (cutoff level: $p = 0.01$) were performed using systemic biomarkers (CRP, FIB and WBC) as dependent variables, each analyzed under 13 independent variables, that is, BOP, PD, CAL, age, gender, body mass index (BMI), smoking, hypertension, socioeconomic status, cardiac medications in four groups (anti-platelets, beta blockers, lipid-lowering agents, calcium channel antagonists). Skewedness of all variables was assessed, and skewed variables (CRP, PD) were log-transformed prior to regression analysis.

Results

Three hundred and seventeen ($n = 317$) eligible CHD patients with periodontitis were included in the study. The demographical, medical and periodontal and systemic parameters of the study participants are presented in Table 1.

Table 1. Characteristics of study population

Age (years)	49.3 ± 9.3[a] (31–79)
Age-groups (%):	
30–39	42 (13)
40–49	122 (38)
50–59	97 (31)
≥60	56 (18)
Gender (%)	
Male	273 (86)
Female	44 (14)
Education (%)	
No education	63 (20)
Secondary	217 (68)
University	37 (12)
Income (per month) (%)	
Rs. <10,000 (<US$ 101)	274 (86)
Rs. 10–20,000 (US$ 101–202)	34 (11)
Rs. >20,000.00 (>US$ 202)	9 (3)
Occupation (%)	
Unskilled	165 (52)
Skilled	75 (24)
Professional	77 (24)
Body mass index	28.3 ± 6.8[a] (18–58)
≤Desirable (≤25 Kg/m^2; %)	115 (36)
≥Overweight (>25 Kg/m^2; %)	202 (64)
Smoking (%):	
Non-smoker	191 (60)
Former smoker	126 (40)
Coronary heart disease extent (%):	
1 vessel	89 (28)
2 vessels	137 (43)
≥3 vessels	91 (29)
Hypertension:	
Systolic blood pressure	123.6 ± 10.9[a] (90–170)
Diastolic blood pressure	84.1 ± 7.6[a] (60–110)
Hypertensive (%)	168 (53)
Non-hypertensive (%)	149 (47)
Periodontal and systemic parameters	
BOP%	42.3 ± 15.5[a] (20.8–100.0)
PPD mm	3.4 ± 0.9[a] (1.3–7.3)
PPD ≥ 4 mm%	39.4 ± 16.9[a] (6.4–89.4)
PPD ≥ 5 mm%	21.8 ± 14.5[a] (0.0–80.4)
PPD ≥ 6 mm%	9.8 ± 9.3[a] (0.0–70.2)
CAL mm	3.5 ± 1.6[a] (0.09–11.2)
CAL ≥ 3 mm%	55.9 ± 19.7[a] (9.00–100.0)
CAL ≥ 4 mm%	40.8 ± 20.8[a] (3.0–99.0)
CAL ≥ 5 mm%	27.7 ± 19.4[a] (0.6–97.0)
C-reactive protein (mg/l)	4.6 ± 3.6[a] (0.1–24.2)
Fibrinogen (mg/l)	382.6 ± 146.5[a] (94.0–1093.0)
White blood cells (×10^9/l)	8.1 ± 2.0[a] (3.5–15.3)

Data presented in *n* (%) or [a]Mean ± SD (Range).

Unadjusted linear associations between periodontal and systemic parameters are presented in Fig. 1. A significant positive relationship between mean BOP% and serum levels of CRP ($r = 0.629$; $p < 0.001$), FIB ($r = 0.424$; $p < 0.001$) and WBC ($r = 0.493$; $p < 0.001$) was noted. The associations between PD or CAL and the systemic parameters were also significant ($p < 0.001$), but less strong than BOP (Fig. 1).

A linear regression model was fit with BOP%, PD and CAL as independent and CRP as dependent variables. There was a significant ($p = 0.001$) relationship between these variables. BOP showed a significant effect (or prediction) on CRP levels, with lowest standard error (SE = 0.011). When PD and CAL remain constant, 1 unit change (1% increase) in BOP corresponds to a 0.136 mg/l increase in CRP levels. PD also had an effect on CRP level prediction (SE = 0.165); when BOP and CAL remain constant, 1 unit change (1 mm increase) in PD corresponds to 0.572 mg/l increase in CRP levels.

A linear model was fit with BOP %, PD and CAL as independent and FIB as dependent variable. There was a significant ($p < 0.0001$) relationship between these variables, with 1 unit change in BOP resulting in 0.522 mg/l increase in FIB levels. When a linear model was fit with BOP%, PD and CAL as independent and WBC as dependent variables, their relationship was also

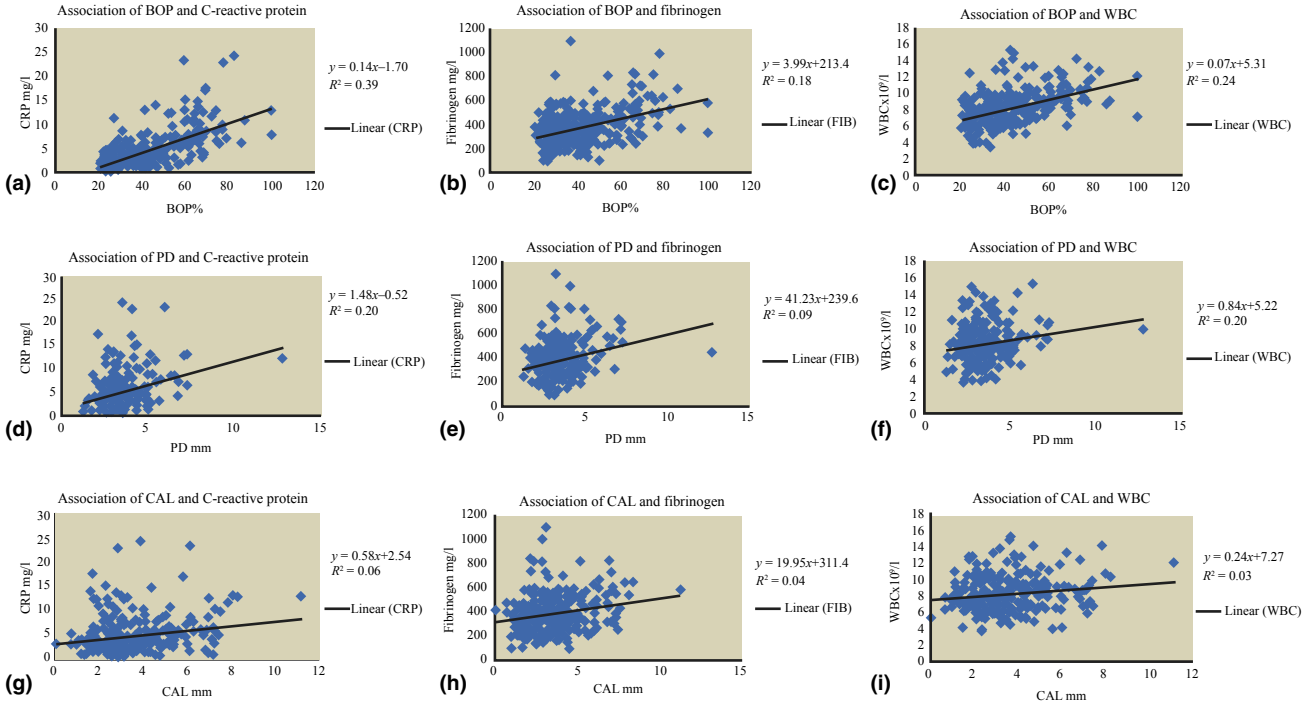

Fig. 1. Unadjusted linear associations between periodontal and systemic parameters (A. Bleeding on Probing (BOP) and C-Reactive Protein; B. Bleeding on Probing (BOP) and Fibrinogen; C. Bleeding on Probing (BOP) and WBCs; D. Probing Depth (PD) and C-Reactive protein; E. Probing Depth and Fibrinogen; F. Probing Depth and WBCs; G. Clinical Attachment Loss (CAL) and C-Reactive Protein; H. Clinical Attachment Loss (CAL) and Fibrinogen; I. Clinical Attachment Loss (CAL) and WBCs

significant ($p < 0.0001$). One unit change in BOP resulted in 0.064 units increase in WBC levels.

Stepwise regression analysis models revealed significant association of CRP with BOP, PD, and income. FIB showed a significant association with BOP, PD, and age, while WBC was significantly associated only with BOP (Table 2).

When patients were distributed separately into tertiles of mean BOP (High: ≥45.37%; Moderate: 32.80–45.36%; Low: ≤32.79%), PD (High: ≥3.58 mm; Moderate: 2.95–3.57 mm; Low: ≤2.94 mm), and CAL (High: ≥3.91 mm; Moderate: 2.67–3.90 mm; Low: ≤2.66 mm), the distribution of tertiles of CRP levels (High: ≥4.69 mg/l; Moderate: 3.01–4.68 mg/l; Low: ≤3.00 mg/l) was significantly different within each periodontal parameter distribution (BOP: $p < 0.001$; PD: $p = 0.002$; CAL: $p = 0.011$; Fig. 2).

When patients were distributed into tertiles of mean CRP levels, the distribution of tertiles of periodontitis (High: $n = 37$; Moderate: $n = 19$; Low: $n = 20$; see Methods for definition) were significantly different, as

Table 2. Stepwise regression analysis. Dependent variables: CRP, fibrinogen and WBC

Variable	β	p	Adj R^2
CRP[a]			
BOP	0.129	<0.001	0.358
PD	0.129	0.006	0.372
Income	0.118	0.008	0.384
Fibrinogen[b]			
BOP	0.366	<0.001	0.177
PD	0.153	0.004	0.193
Age	0.105	0.038	0.202
WBC[c]			
BOP	0.491	<0.001	0.238

[a]Age, gender, education, work type, BMI, hypertension, and CAL are not independently associated with CRP.
[b]Gender, education, income, work type, BMI, hypertension, and CAL are not independently associated with Fibrinogen.
[c]Age, gender, education, income, work type, BMI, hypertension, PD and CAL are not independently associated with WBCs.
BMI, body mass index; BOP, bleeding on probing; CAL, clinical attachment level; CRP, serum C-reactive protein levels; PD, probing depth; WBC, white blood cells.

High-Periodontitis subjects were most frequently found in the High-CRP tertile ($p = 0.015$).

Discussion

The purpose of this study was to examine the possible associations between individual clinical parameters of periodontitis (BOP, PD, and CAL) and systemic biomarkers of CHD risk (CRP, fibrinogen, WBC) in a population of CHD patients with periodontitis. The results indicate that BOP, the clinical parameter that provides a measure of periodontal tissue inflammation (Greenstein et al. 1981, Engelberger et al. 1983), is strongly associated with CRP, a systemic inflammatory biomarker and CHD risk factor (Wang et al. 2002).

A strong association between periodontitis and CHD has been established from epidemiological studies (Humphrey et al. 2008, Dietrich et al. 2013, Tonetti & Van Dyke 2013). This association is indirectly supported by intervention trials that examined the effects of periodontal therapy on CHD risk markers (D'Aiuto et al. 2004, 2006, Seinost et al. 2005, Hussain Bokhari et al. 2009, Offenbacher et al. 2009, Bokhari et al. 2012, Teeuw et al.

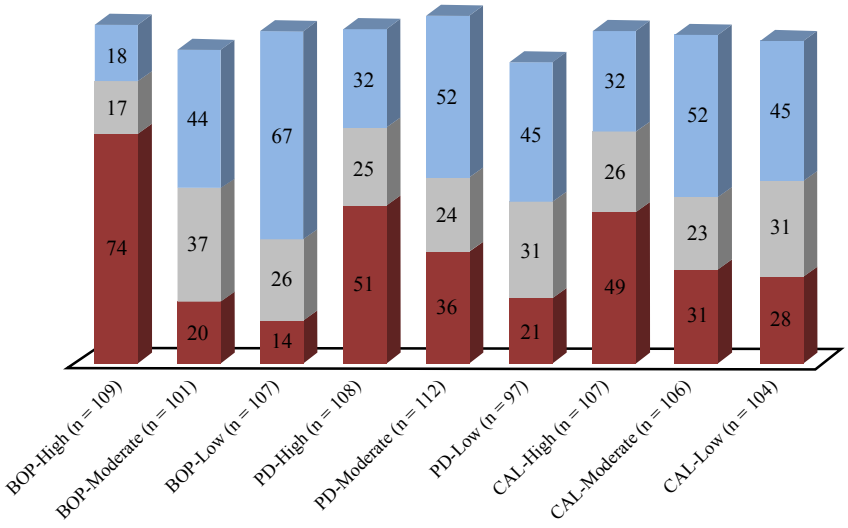

Fig. 2. Distribution of subjects (*n*) by High-Moderate-Low tertiles of mean BOP%, PD and CAL (see text for definitions) in corresponding tertiles of CRP levels [■ CRPHigh (≥4.69 mg/l); ■ CRP Moderate (3.0–4.68 mg/l); ■ CRPLow (<3.0 mg/l)].

2014). Periodontitis has been strongly associated with systemic CRP levels (Paraskevas et al. 2008, Fisher et al. 2010) and other systemic CHD risk indicators (Kweider et al. 1993, Danesh et al. 1998). The results of this study, where participants with high periodontitis were much more likely to belong to the high-CRP subgroup, are congruent with reports of a dose–response relationship between periodontitis and CHD (Joshipura et al. 1996, Arbes et al. 1999) and provide further support for the association between periodontitis and systemic CRP levels.

In this study, BOP was the periodontal parameter most strongly associated with CRP levels in 317 CHD patients with periodontitis; this result is consistent with the reported statistically significant positive association between high percentage of BOP^+ sites and CRP levels in 159 systemically healthy adolescents (Lopez et al. 2011), as well as the positive association between extent of BOP and CRP levels in 5063 community-dwelling adults from the Atherosclerosis Risk in Communities (ARIC) study (Beck & Offenbacher 2002). In periodontitis patients, sites with BOP are histologically characterized by a threefold increase in the volumetric density of inflammatory cells (lymphocytes, macrophages and monocytes), compared to sites without BOP (Cooper et al. 1983). The significant association between BOP, a clinical marker of localized tissue inflammation, and a marker of systemic inflammation (CRP) is consistent with the recently reported strong association between periodontal tissue metabolic activities, a possible surrogate for periodontal inflammation, and histologically assessed atherosclerotic plaque inflammation in patients who underwent carotid endarterectomy (Fifer et al. 2011). The results of this study have potential implications for future studies (inclusion criteria, periodontal therapy endpoints) investigating the association between periodontal and cardiovascular disease.

The results of stepwise regression analyses indicated that BOP was also strongly associated with fibrinogen and the only clinical periodontal parameter associated with WBC (Table 2). This finding is consistent with earlier reports of a positive association between periodontitis and increased fibrinogen or WBC levels (Loos et al. 2000, Sahingur et al. 2003, Joshipura et al. 2004, Bizzarro et al. 2007, Amabile et al. 2008, Buhlin et al. 2009). Fibrinogen, the main coagulation protein and important determinant of blood viscosity, interacts with bacteria and bacterial surface components that may promote colonization and adhesion (Sahingur et al. 2003), and is significantly associated with CHD (Danesh et al. 1998). Although WBC elevations in periodontitis are rather modest, with leukocytosis ($>10 \times 10^9$/l) being sporadic amongst periodontitis patients (Loos 2005), it is notable that modestly elevated WBC counts are associated with an increased risk for cardiovascular diseases (Danesh et al. 1998, Loos et al. 2000, Dave & Van Dyke 2008). With respect to the systemic biomarkers examined in this study, the use of non-fasting blood samples represents a potential limitation.

Established risk factors for atherosclerotic diseases include diabetes mellitus, smoking, hypercholesterolaemia, dyslipidemia, obesity, and hypertension (Wood 2001). In the present study population, diabetes mellitus and current smoking were excluded; however, hypertension and obesity (noted through BMI) were recorded and accounted for in the statistical analyses performed. The study population was selected to be free from acute or chronic inflammatory conditions; however, subclinical or unknown inflammatory conditions might have had an effect on the observed systemic inflammatory biomarker levels. Although this study population was receiving lipid-lowering medications, the lack of information on the subjects' lipid profile represents a limitation of the study; however, periodontal intervention studies have failed to indicate an effect of periodontal therapy on lipid profiles (Tonetti & Van Dyke 2013).

Despite this study finding that PD and CAL were also significantly associated with the systemic biomarkers, these associations were weaker compared to BOP. In addition, the stepwise regression analyses indicated that only PD had modest impact on the BOP associations with CRP and fibrinogen, while CAL did not reach statistical significance for any systemic parameter. The association of PD with CRP reported herein is consistent with the findings of the ARIC study (Beck & Offenbacher 2002, Slade et al. 2003) and of a small-scale study (47 patients, 18 with periodontitis) on Greek acute myocardial infarction patients (Kodovazenitis et al. 2011). The latter study did not identify an association between BOP and CRP (Kodovazenitis et al. 2011).

In conclusion, in CHD patients with periodontitis, BOP is strongly

associated with systemic CRP levels; this association possibly reflects the potential significance of the local periodontal inflammatory burden for systemic inflammation.

Acknowledgements

The kind assistance of staff of Punjab Institute of Cardiology and Sheikh Zayed Medical Complex during this study is gratefully acknowledged.

References

Amabile, N., Susini, G., Pettenati-Soubayroux, I., Bonello, L., Gil, J. M., Arques, S., Bonfil, J. J. & Paganelli, F. (2008) Severity of periodontal disease correlates to inflammatory systemic status and independently predicts the presence and angiographic extent of stable coronary artery disease. *Journal of Internal Medicine* **263**, 644–652.

Arbes, S. J. Jr, Slade, G. D. & Beck, J. D. (1999) Association between extent of periodontal attachment loss and self-reported history of heart attack: an analysis of NHANES III data. *Journal of Dental Research* **78**, 1777–1782.

Beck, J. D. & Offenbacher, S. (2002) Relationship among clinical measure of periodontal disease and their associations with systemic markers. *Annals of Periodontology* **7**, 79–89.

Bizzarro, S., van der Velden, U., ten Heggeler, J. M., Leivadaros, E., Hoek, F. J., Gerdes, V. E., Bakker, S. J., Gans, R. O., Ten Cate, H. & Loos, B. G. (2007) Periodontitis is characterized by elevated PAI-1 activity. *Journal of Clinical Periodontology* **34**, 574–580.

Bokhari, S. A. H., Khan, A. A., Butt, A. K., Azhar, M., Hanif, M., Izhar, M. & Tatakis, D. N. (2012) Non-surgical periodontal therapy reduces coronary heart disease risk markers: a randomized controlled trial. *Journal of Clinical Periodontology* **39**, 1065–1074.

Buhlin, K., Hultin, M., Norderyd, O., Persson, L., Pockley A. G.Rabe, P., Klinge, B. & Gustafsson, A. (2009) Risk factors for atherosclerosis in cases with severe periodontitis. *Journal of Clinical Periodontology* **36**, 541–549.

Cooper, P. G., Caton, J. G. & Polson, A. M. (1983) Cell populations associated with gingival bleeding. *Journal of Periodontology* **54**, 497–502.

D'Aiuto, F., Parkar, M., Andreou, G., Suvan, J., Brett, P. M., Ready, D. & Tonetti, M. S. (2004) Periodontitis and systemic inflammation: control of the local infection is associated with a reduction in serum inflammatory markers. *Journal of Dental Research* **83**, 156–160.

D'Aiuto, F., Parkar, M., Nibali, L., Suvan, J., Lessem, J. & Tonetti, M. S. (2006) Periodontal infections cause changes in traditional and novel cardiovascular risk factors: results from a randomized controlled clinical trial. *American Heart Journal* **151**, 977–984.

Danesh, J., Collins, R., Appleby, P. & Peto, R. (1998) Association of fibrinogen, C-reactive protein, albumin, or leukocyte count with coronary heart disease: meta-analyses of prospective studies. *Journal of the American Medical Association* **279**, 1477–1482.

Dave, S. & Van Dyke, T. (2008) The link between periodontal disease and cardiovascular disease is probably inflammation. *Oral Diseases* **14**, 95–101.

Dietrich, T., Sharma, P., Walter, C., Weston, P. & Beck, J. (2013) The epidemiological evidence behind the association between periodontitis and incident atherosclerotic cardiovascular disease. *Journal of Clinical Periodontology* **40** (Suppl. 14), S70–S84. Erratum in: *J Clin Periodontol*. 2013; 40(Suppl. 14), S210–5.

Ebersole, J. L., Cappelli, D., Mathys, E. C., Steffen, M. J., Singer, R. E., Montgomery, M., Mott, G. E. & Novak, M. J. (2002) Periodontitis in humans and non-human primates: oral-systemic linkage inducing acute phase proteins. *Annals of Periodontology* **7**, 102–111.

Engelberger, T., Hefti, T., Kallenberger, A. & Rateitschak, K. H. (1983) Correlations among papilla bleeding index, other clinical indices and histologically determined inflammation of gingival papilla. *Journal of Clinical Periodontology* **10**, 579–589.

Fifer, K. M., Qadir, S., Subraminian, S., Vijakumar, J., Figueroa, A. L. & Truong, Q. A. (2011) Positron emission tomography measurement of periodontal 18F-fluorodeoxyglucose uptake Is associated with histologically determined carotid plaque inflammation. *Journal of the American College of Cardiology* **57**, 971–976.

Fisher, M. A., Borgnakke, W. S. & Taylor, G. W. (2010) Periodontal disease as a risk marker in coronary heart disease and chronic kidney disease. *Current Opinion in Nephrology and Hypertension* **19**, 519–526.

Greenstein, G., Caton, J. & Polson, A. M. (1981) Histologic characteristics associated with bleeding after probing and visual signs of inflammation. *Journal of Periodontology* **52**, 420–425.

Humphrey, L. L., Fu, R., Buckley, D. I., Freeman, M. & Helfand, M. (2008) Periodontal disease and coronary heart disease incidence: a systematic review and meta-analysis. *Journal of General Internal Medicine* **23**, 2079–2086.

Hussain Bokhari, S. A., Khan, A. A., Tatakis, D. N., Azhar, M., Hanif, M. & Izhar, M. (2009) Non-surgical periodontal therapy lowers serum inflammatory markers: a pilot study. *Journal of Periodontology* **80**, 1574–1580.

Joshipura, K. J., Rimm, E. B., Douglass, C. W., Trichopoulos, D., Ascherio, A. & Willett, W. C. (1996) Poor oral health and coronary heart disease. *Journal of Dental Research* **75**, 1631–1636.

Joshipura, K. J., Wand, H. C., Merchant, A. T. & Rimm, E. B. (2004) Periodontal disease and biomarkers related to cardiovascular disease. *Journal of Dental Research* **83**, 151–155.

Kebschull, M., Demmer, R. T. & Papapanou, P. N. (2010) "Gum bug, leave my heart alone!"–epidemiologic and mechanistic evidence linking periodontal infections and atherosclerosis. *Journal of Dental Research* **89**, 879–902.

Kodovazenitis, G., Pitsavos, C., Papadimitriou, L., Deliargyris, E. N., Vrotsos, I., Stefanadis, C. & Madianos, P. N. (2011) Periodontal disease is associated with higher levels of C-reactive protein in non-diabetic, non-smoking acute myocardial infarction patients. *Journal of Dentistry* **39**, 849–854.

Kweider, M., Lowe, G. D., Murray, G. D., Kinane, D. F. & McGowan, D. A. (1993) Dental disease, fibrinogen and white cell count; links with myocardial infarction? *Scottish Medical Journal* **38**, 73–74.

Loos, B. (2005) Systemic markers of inflammation in periodontitis. *Journal of Periodontology* **76**, 2106–2115.

Loos, B. G., Craandijk, J., Hoek, F. J., Wertheim-van Dillen, P. M. & Van der Velden, U. V. (2000) Elevation of systemic markers related to cardiovascular diseases in the peripheral blood of periodontitis patients. *Journal of Periodontology* **71**, 1528–1534.

Lopez, R., Baelum, V., Hedegaard, C. J. & Bendtzen, K. (2011) Serum levels of C-reactive protein in adolescents with periodontitis. *Journal of Periodontology* **82**, 543–549.

Meurman, J. H., Sanz, M. & Janket, S. J. (2004) Oral health, atherosclerosis, and cardiovascular disease. *Critical Reviews in Oral Biology and Medicine* **15**, 403–413.

Montebugnoli, L., Servidio, D., Miaton, R. A., Prati, C., Tricoci, P. & Melloni, C. (2004) Poor oral health is associated with coronary heart disease and elevated systemic inflammatory and haemostatic factors. *Journal of Clinical Periodontology* **31**, 25–29.

Noack, B., Genco, R. J., Trevisan, M., Grossi, S., Zambon, J. J. & De Nardin, E. (2001) Periodontal infections contribute to elevated systemic C-reactive protein level. *Journal of Periodontology* **72**, 1221–1227.

Offenbacher, S., Beck, J. D., Moss, K., Mendoza, L., Paquette, D. W., Barrow, D. A., Couper, D. J., Stewart, D. D., Falkner, K. L., Graham, S. P., Grossi, S., Gunsolley, J. C., Madden, T., Maupome, G., Trevisan, M., Van Dyke, T. E. & Genco, R. J. (2009) Results from the Periodontitis and Vascular Events (PAVE) Study: a pilot multicentered, randomized, controlled trial to study effects of periodontal therapy in a secondary prevention model of cardiovascular disease. *Journal of Periodontology* **80**, 190–201.

Paraskevas, S., Huizinga, J. D. & Loos, B. G. (2008) A systematic review and meta-analyses on C-reactive protein in relation to periodontitis. *Journal of Clinical Periodontology* **35**, 277–290.

Reyes, L., Herrera, D., Kozarov, E., Roldán, S. & Progulske-Fox, A. (2013) Periodontal bacterial invasion and infection: contribution to atherosclerotic pathology. *Journal of Clinical Periodontology* **40** (Suppl. 14), S30–S50.

Sahingur, S. E., Sharma, A., Genco, R. J. & De Nardin, E. (2003) Association of increased levels of fibrinogen and the -45 G/A fibrinogen gene polymorphism with chronic periodontitis. *Journal of Periodontology* **74**, 329–337.

Schenkein, H. A. & Loos, B. G. (2013) Inflammatory mechanisms linking periodontal diseases to cardiovascular diseases. *Journal of Clinical Periodontology* **40**(Suppl. 14), S51–S69.

Seinost, G., Wimmer, G., Skerget, M., Thaller, E., Brodmann, M., Gasser, R., Bratschko, R. O. & Pilger, E. (2005) Periodontal treatment improves endothelial dysfunction in patients with severe periodontitis. *American Heart Journal* **149**, 1050–1054.

Sesso, H. D., Wang, L., Buring, J. E., Ridker, P. M. & Gaziano, J. M. (2007) Comparison of interleukin-6 and C-reactive protein for the risk of developing hypertension in women. *Hypertension* **49**, 304–310.

Slade, G. D., Ghezzi, E. M., Heiss, G., Beck, J. D., Riche, E. & Offenbacher, S. (2003) Relationship between periodontal disease and C-reactive protein among adults in the Atherosclerosis Risk in Communities study. *Archives of Internal Medicine* **163**, 1172–1179.

Tatakis, D. N. & Kumar, P. S. (2005) Etiology and pathogenesis of periodontal diseases. *Dental Clinics of North America* **49**, 491–516.

Teeuw, W. J., Slot, D. E., Susanto, H., Gerdes, V. E., Abbas, F., D'Aiuto, F., Kastelein, J. J. & Loos, B. G. (2014) Treatment of periodontitis improves the atherosclerotic profile: a systematic review and meta-analysis. *Journal of Clinical Periodontology* **41**, 70–79.

Tonetti, M. S. & Van Dyke, T. E. (2013) Working group 1 of the joint EFP/AAP workshop. Periodontitis and atherosclerotic cardiovascular disease: consensus report of the Joint EFP/AAP Workshop on Periodontitis and Systemic Diseases. *Journal of Clinical Periodontology* **40** (Suppl. 14), S24–S29.

Wang, T. J., Larson, M. G., Levy, D., Benjamin, E. J., Kupka, M. J., Manning, W. J., Clouse, M. E., D'Agostino, R. B., Wilson, P. W. & O'Donnell, C. J. (2002) C-reactive protein is associated with subclinical epicardial coronary calcification in men and women: the Framingham Heart Study. *Circulation* **106**, 1189–1191.

Williams, R. C. (1990) Periodontal disease. *New England Journal of Medicine* **322**, 373–382.

Wood, D. (2001) Asymptomatic individuals–risk stratification in the prevention of coronary heart disease. *British Medical Bulletin* **59**, 3–16.

Address:
Syed Akhtar H. Bokhari
22-Rewaz Garden, Lahore 54000
Pakistan
E-mail: pdphr@yahoo.com

Clinical Relevance

Scientific rationale for the study: There is limited data on the possible association between specific periodontal clinical parameters and systemic biomarkers of coronary heart disease, despite the strong association between periodontitis and coronary heart disease.

Principal findings: Bleeding on probing, a clinical measure of periodontal tissue inflammation is most strongly associated with CRP, a systemic inflammatory biomarker and coronary disease risk factor.

Practical implications: In coronary disease patients with periodontitis, increased signs of periodontal inflammation may indicate raised levels of systemic coronary disease risk biomarkers.