

# Periodontitis: A Future Risk of Acute Coronary Syndrome? A Follow-Up Study Over 3 Years

Stefan Renvert,\*† Ola Ohlsson,\*‡ Thomas Pettersson,‡ and G. Rutger Persson§||

**Background:** Periodontitis has been associated with cardiovascular disease. We assess if the recurrence of acute coronary syndrome (ACS) could be predicted by preceding medical and periodontal conditions.

**Methods:** A total of 165 consecutive subjects with ACS and 159 medically healthy, matched control subjects were examined and followed for 3 years. Periodontitis was defined by alveolar bone loss. Subgingival microbial samples were studied by the checkerboard DNA–DNA hybridization method.

**Results:** The recurrence of ACS was found in 66 of 165 (40.0%) subjects, and a first ACS event was found in seven of 159 (4.4%) subjects among baseline control subjects. Subjects who later had a second ACS event were older ( $P < 0.001$ ). Significantly higher serum levels of high-density lipoprotein ( $P < 0.05$ ), creatinine ( $P < 0.01$ ), and white blood cell (WBC) counts ( $P < 0.001$ ) were found in subjects with future ACS. Periodontitis was associated with a first event of ACS (crude odds ratio [OR]: 10.3:1; 95% confidence interval [CI]: 6.1 to 17.4;  $P < 0.001$ ) and the recurrence of ACS (crude OR: 3.6:1; 95% CI: 2.0 to 6.6;  $P < 0.001$ ). General linear modeling multivariate analysis, controlling for age and the prediction of a future ACS event, identified that WBC counts ( $F = 20.6$ ;  $P < 0.001$ ), periodontitis ( $F = 17.6$ ;  $P < 0.001$ ), and serum creatinine counts ( $F = 4.5$ ;  $P < 0.05$ ) were explanatory of a future ACS event.

**Conclusions:** The results of this study indicate that recurrent ACS events are predicted by serum WBC counts, serum creatinine levels, and a diagnosis of periodontitis. Significantly higher counts of putative pathogens are found in subjects with ACS, but these counts do not predict future ACS events. *J Periodontol* 2010;81:992-1000.

## KEY WORDS

Inflammation; myocardial infarction; periodontitis; recurrence.

Cardiovascular diseases comprise a variety of heart and vascular conditions.<sup>1</sup> Over the last 30 years, greater life expectancy and changes in diet and exercise habits resulted in a higher prevalence of obesity, elevated levels of blood cholesterol, hypertension, and diabetes mellitus, which are all cardiovascular risk factors.<sup>2</sup> Although medical advances have resulted in an increased survival rate of patients with acute coronary syndrome (ACS), the risks for the recurrence of ACS remain very high.

A large number of surrogate measures to assess the risks of future cardiovascular diseases exists. These include assessments of carotid intima–media thickness, flow-mediated dilatation of the brachial artery, and assessments of many serum markers (i.e., high-density lipoprotein [HDL], low-density lipoprotein [LDL], fibrinogen, triglycerides, high-sensitivity C-reactive protein [hsCRP], hemoglobin A1c [HbA1c], and systolic/diastolic blood pressure). However, data on such measures only partly account for the occurrence of future cardiovascular diseases.<sup>3</sup>

A relationship between periodontitis, a common chronic oral disease in adults, and cardiovascular diseases was identified in several studies.<sup>4-6</sup> However, this association is not strong and may be partly explained by the fact that several studies assessing this association have been performed without medically

\* Department of Health Sciences, University of Kristianstad, Kristianstad, Sweden.

† School of Dental Sciences, Trinity College, Dublin, Ireland.

‡ Department of Medicine, Kristianstad Central Hospital, Kristianstad, Sweden.

§ Department of Periodontology, University of Bern, Bern, Switzerland.

|| Departments of Periodontics and Oral Medicine, University of Washington, Seattle, WA.

confirmed clinical status. Another factor that makes it difficult to interpret data is the different criteria used to define periodontitis in studies assessing the relationship between periodontitis and cardiovascular diseases.<sup>4</sup>

Recent dental studies<sup>7-12</sup> suggested that comprehensive non-surgical periodontal therapy may have positive effects on the intima-media thickness and brachial artery flow rate. However, the immediate effects of periodontal intervention may induce an immediate exaggeration of serum hsCRP and initially reduce the brachial artery flow rate in subjects without evidence of cardiovascular disease.<sup>12</sup> Limited data are available on the effects of such surrogate markers as risk factors for outcomes of periodontal therapy in subjects with a confirmed preceding history of ACS.

In one recent case-control, longitudinal intervention study,<sup>13</sup> the authors concluded that the extent of severe cardiovascular adverse events in subjects treated for chronic periodontitis in specialist periodontal clinics or receiving routine dental care in a general dental practice did not differ. The findings also suggested that non-surgical periodontal intervention does not increase the risk for adverse cardiovascular events in subjects with a history of cardiovascular disease. Thus, non-surgical periodontal therapy in cardiovascular-risk patients is safe and does not induce serious adverse events.

In a previous case-control study report, we identified that severe periodontitis, defined by the extent of alveolar bone loss, was strongly associated with ACS.<sup>14,15</sup> However, current clinical assessments of periodontal conditions using probing depths (PDs) and the bleeding index failed to demonstrate a risk association with ACS. Similar results were obtained by others.<sup>16-18</sup> Our data<sup>15</sup> also suggested that subjects with ACS have an increased pathogenic bacterial burden in periodontal pockets.

The objective of the present case-control follow-up study over 3 years is to study if a one-time assessment of periodontal and medical conditions could predict a future event of ACS in subjects who had not received organized periodontal therapy during the time period.

## MATERIALS AND METHODS

The Ethical Committee at Lund University, Lund, Sweden (institutional review board [IRB] #LU556-00) approved the study. The study was performed between 2002 and 2007. The participating subjects signed IRB-approved informed-consent forms. The original study protocol was described elsewhere.<sup>14</sup> Since the previous report,<sup>14</sup> we continued to enroll subjects until 165 consecutively surviving subjects with ACS and 159 control subjects (matched for age, gender, smoking, and socioeconomic status) were enrolled. A total of 287 males and 37 females

were included in the study. Medical study data from the enrolled and hospital-admitted subjects with a diagnosis of ACS were obtained.

All ACS subjects underwent a standard-of-care cardiovascular examination at the time of hospital admission. The medical diagnosis of baseline ACS status was based on chest pain associated with typical electrocardiogram (ECG) changes. The initial ECG was considered diagnostic for myocardial infarction if there was an ST segment elevation  $\geq 2$  mm in a chest lead or an ST segment elevation  $\geq 1$  mm in a limb lead. ST depression and/or T-wave inversion changes combined with typical serial pattern of cardiac markers (i.e., creatinine kinase isoenzyme MB [CK-MB] and troponin T) according to local laboratory standards were also considered diagnostic for myocardial infarction. Left bundle block was considered diagnostic for myocardial infarction if chest pain associated with typical serial patterns of cardiac markers were present. Medical data including an ECG, serum assessments for troponin T, lipids (cholesterol, HDLs, and LDLs), hsCRP, white blood cell (WBC) count, HbA1c, sodium, potassium, and creatinine levels were collected.

All subjects who were admitted and treated for ACS received standard-of-care cardiovascular procedures and medications as deemed appropriate for each subject. Thus, a variety of interventions, drugs, and recalls were used for the purpose of stabilizing the subjects and to prevent recurrence of ACS. In this process, all subjects were compliant. All subjects who were readmitted to the hospital within the follow-up period of 3 years underwent the described medical examinations to confirm the new diagnosis of ACS.

Once the subjects with ACS were released from the hospital, a calibrated, experienced examiner (Christel Lindahl, Region Skåne, Specialty Clinic for Periodontology, and Kristianstad University, Kristianstad, Sweden) and a supervising periodontist (SR) at the Dental Clinic of Health Sciences at the University of Kristianstad performed the dental examinations.

A routine cardiology examination was performed in all control subjects to rule out a condition of preexisting ACS or a potential risk for a future ACS event. Serum lipids (cholesterol, HDL and LDL, hsCRP, WBC count, HbA1c, sodium, potassium, and creatinine levels) were assessed in control subjects. The control subjects did not undergo the troponin-T assessments but were otherwise examined to confirm that they did not have cardiovascular disease.

Routine clinical periodontal assessments were also performed in all subjects. The examiner (SR) was unaware of whether study subjects belonged to the ACS or control groups. Data on full-mouth PDs, gingival bleeding, oral hygiene, and the number of remaining teeth were collected. Full-mouth intraoral

radiographs were taken. Dental radiographs were analyzed for the extent of alveolar bone loss. Thus, definitive periodontitis was diagnosed based on the assessment of alveolar bone loss. The extent of alveolar bone loss was defined as the distance  $\geq 4.0$  mm between interproximal bone levels and the cemento-enamel junction (CEJ) as described elsewhere.<sup>14</sup> In summary, bone loss in different categories was expressed as having 10%, 20%, 30%, 40%, or  $>40\%$  of measurable sites with distances  $\geq 4.0$  mm between CEJ and bone level. An examiner (GRP) calibrated as part of another study and unaware of the medical diagnosis analyzed the radiographs.<sup>14,19</sup> In the previous publication<sup>15</sup> on periodontitis and ACS, we reported that the extent of alveolar bone loss was a robust marker of the association between periodontitis and ACS.

Although the great majority of the cases with ACS were older men, in our previous report,<sup>14</sup> we continued to enroll consecutive cases with ACS but enrolled only women (independent of age) and men who were  $<60$  years of age. We also continued to enroll control subjects who were chosen to meet the age, gender, smoking, and socioeconomic conditions of the additionally enrolled ACS cases.<sup>14</sup> Because of the fact that it is difficult to control for the impact of many different confounding factors in cardiovascular diseases, healthy control subjects, without any medical or subjective symptoms of cardiovascular disease, were included as subjects without ACS. The control subjects were balanced to the subjects with ACS according to gender, smoking status, and marital and socioeconomic factors to compensate for potential confounders. None of the subjects in either group received qualified periodontal therapy within the previous 3 years before the baseline examination or during the following 3-year period.

During the following 3-year period (2003 to 2006), medical records at the regional primary care hospital in Kristianstad, Sweden, and at affiliated clinics were screened to assess if and when study subjects had experienced an additional ACS event. Similarly, records were screened to identify control subjects who had been diagnosed with a first event of ACS. Records of the dental clinic and the regional periodontal specialty clinic confirmed that none of the subjects had received dental treatments other than routine dental care performed in a general dental practice. The original medical and dental data obtained were used to assess the utility of such information to predict a future ACS event.<sup>14</sup>

### Microbiologic Assessments

The sequence of dental examinations was as follows: 1) full-mouth radiographs were taken from which the four worst interproximal periodontal sites were identified, 2) subgingival microbial samples were collected

and pooled from these selected sites, and 3) other periodontal examination procedures were performed. The bacterial samples were processed by the checkerboard DNA-DNA hybridization method<sup>15,20-22</sup> and processed at the Oral Microbiology Laboratory, University of Bern. Subgingival bacterial samples were harvested with sterile endodontic paper points.

### Statistical Methods

The data were analyzed with a statistical program.<sup>¶</sup> Independent *t* (equal variance was not assumed) and Mann-Whitney U tests were used to assess group differences. Cochran-Mantel-Haenszel statistics for estimation of odds ratios (ORs) (crude values were non-adjusted) were used to assess the risks between study parameters and ACS. General linear modeling (GLM) multivariate analysis was performed to assess explanatory variables related to a new event of ACS.

## RESULTS

In the present study, the ages of subjects varied between 31 and 87 years, and 46% of subjects were  $<60$  years of age. Among the cohort of 165 subjects with ACS, 66 (40.0%) subjects were readmitted within 3 years after the first ACS event with a new ACS diagnosis. In the control group and within the same time period, seven of 159 (4.4%) subjects were admitted to the hospital with a confirmed first event of ACS. Analysis of the baseline data demonstrated that the crude OR of being admitted to a hospital with ACS and having periodontitis (all subjects included in the analysis), defined by the extent of alveolar bone loss at 30% of sites, was 10.3:1 (95% confidence interval [CI]: 6.1 to 17.4;  $P < 0.001$ ). The crude OR to predict a future event of ACS (all subjects included in the analysis) was 3.6:1 (95% CI: 2.0 to 6.6;  $P < 0.001$ ).

### Analyses of Baseline Medical and Periodontal Conditions in Association With a Future ACS Event (test group with a history of ACS but stable and those with a future ACS event)

Data regarding characteristics of subjects in the ACS group with either stable conditions or recurrent ACS are presented in Table 1. Subjects with a recurrent ACS event were older than stable ACS subjects ( $P < 0.001$ ). At the time of first admission with a diagnosis of ACS, higher serum creatinine values were identified in those subjects who, within 3 years, had a new event of ACS ( $P < 0.05$ ; Table 1). The distribution of serum creatinine values by periodontal diagnosis is presented in a boxplot diagram for subjects with stable ACS or recurrence of ACS in Figure 1. In difference, serum troponin-T values at the time of the first ACS admission were higher among those with stable ACS conditions than in those with recurrent ACS

¶ SPSS 17.0 for MAC, SPSS, Chicago, IL.

Table 1.

## Subject-Characteristic Values of Disease Surrogate Markers in Subjects With Stable and Recurrent ACS

Variable	ACS		Significance	
	Stable	Recurrent	95% CI	P
Male gender (%)	88.9	86.4	–	0.63 (NS)
Age (years; mean ± SD)	59.1 ± 8.5	63.7 ± 8.8	1.8 to 7.2	<0.001
Smoking (years; mean ± SD)	21.8 ± 17.8	22.7 ± 18.1	–6.8 to 5.0	0.77 (NS)
Creatinine (μmol/l; mean ± SD)	76.7 ± 16.2	84.0 ± 22.1	–13.7 to –0.95	<0.05
Serum hsCRP (mg/l; mean ± SD)	17.0 ± 31.7	14.4 ± 23.5	–6.33 to 12.81	0.66 (NS)
HDL (mg/dl; mean ± SD)	1.18 ± 0.26	1.23 ± 0.37	–0.21 to 0.22	0.37 (NS)
LDL (mg/dl; mean ± SD)	2.99 ± 1.08	2.94 ± 0.99	–0.28 to 0.38	0.76 (NS)
Triglycerides (mg/dl; mean ± SD)	1.69 ± 0.88	1.86 ± 1.18	–0.50 to 0.18	0.35 (NS)
Cholesterol (mg/dl; mean ± SD)	4.91 ± 1.15	4.98 ± 1.01	–0.41 to 0.27	0.70 (NS)
WBC count (×10 <sup>9</sup> /l; mean ± SD)	8.61 ± 2.75	9.04 ± 2.92	–1.34 to 0.47	0.35 (NS)
Serum HbA1c (%; mean ± SD)	5.25 ± 1.47	4.97 ± 1.03	–0.24 to 0.79	0.29 (NS)
Blood glucose (%; mean ± SD)	6.07 ± 3.04	6.41 ± 2.52	–1.49 to 0.90	0.56 (NS)
Bone loss at 30% cutoff (periodontitis) (%; mean ± SD)	73.7 ± 44.2	76.2 ± 44.3	–	0.73 (NS)
BOP (gingivitis) (%; mean ± SD)	57.1 ± 22.0	50.5 ± 21.0	–36.4 to 13.4	0.06
BOP at >20% of sites (%; mean ± SD)	95.9 ± 20.9	96.7 ± 17.6	–	0.76 (NS)
Sites with PD ≥6 mm (%; mean ± SD)	3.9 ± 5.6	3.3 ± 5.1	–1.5 to 2.5	0.81 (NS)

– = not applicable; NS = not statistically significant; BOP = bleeding on probing.

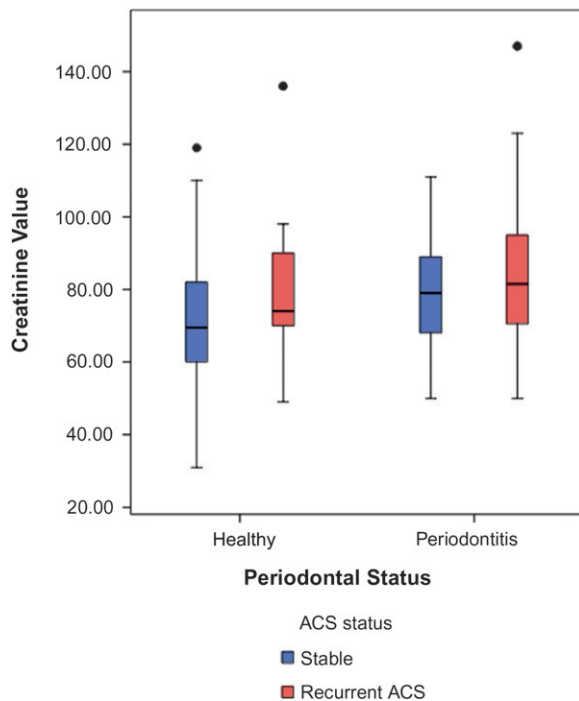
(mean difference: 0.12 ng/ml; SE of the difference: 0.05; 95% CI: 0.02 to 0.23;  $P < 0.01$ ) and at 18 hours after the first ACS admission ( $P < 0.05$ ). Statistical analyses failed to demonstrate that CKMB values over the assessed time points during the first 24 hours after admission differed between those subjects with stable or recurrent ACS conditions. Statistical analyses also failed to demonstrate that serum hsCRP levels differed by alveolar bone loss status in both subgroups. Furthermore, no correlation was found between the proportion of PDs ≥6 mm, 4 to 5 mm, <4 mm and hsCRP.

At the time of the periodontal examination, significantly higher subgingival bacterial counts were found among subjects with ACS than in the control subjects for 27 of 40 species studied ( $P < 0.001$ ). This included species such as *Tannerella forsythia* (previously *T. forsythensis*), *Treponema denticola*, *Fusobacterium nucleatum*, *Prevotella intermedia*, *Streptococcus mitis*, *Streptococcus sanguinis*, and *Streptococcus anginosus* but not *Porphyromonas gingivalis*. However, none of the counts of the 40 species differed between

subjects with recurrent ACS and those who were in remission. The distribution of *T. forsythia* in subjects with a baseline diagnosis of ACS or recurrent ACS is presented in a boxplot diagram in Figure 2.

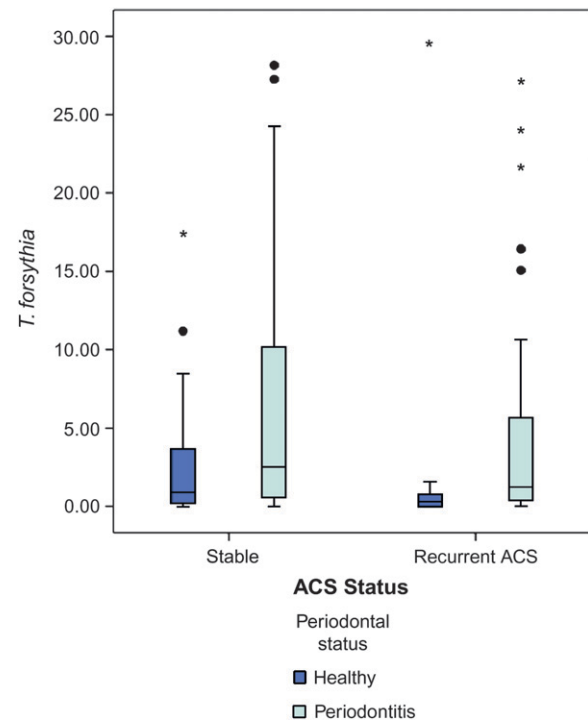
Analysis by independent *t*-test (equal variance was not assumed) failed to identify that neither the proportion of sites with PDs in the categories (<4 mm, 4 to 5 mm, ≥6 mm) or bacterial counts differed between those with stable ACS conditions, and those with a recurrent ACS. Statistical analyses demonstrated a trend of higher counts of bleeding on probing among subjects with a recurrence of ACS ( $P = 0.06$ ). Statistical analyses failed to identify a correlation between serum hsCRP and the percentage of sites with bleeding on probing in both subgroups.

Crude OR calculations for dichotomous data for serum creatinine levels >100 μmol/l demonstrated that subjects with creatinine scores >100 μmol/l were at a significantly higher risk for a recurrence of ACS with a crude OR of 2.9:1 (95% CI: 1.1 to 7.0;  $P < 0.005$ ). Statistical analyses failed to identify a significant



**Figure 1.**

Distribution of serum creatinine values by periodontal diagnosis for subjects with stable or recurrent ACS. ● = outlier value. The bottom and top of boxes are 25<sup>th</sup> and 75<sup>th</sup> percentiles and the line in the middle of the box is the 50<sup>th</sup> percentile. The ends of the whiskers represent one SD above and below the mean of the data.



**Figure 2.**

Distribution of *T. forsythia* counts in subjects with stable or recurrent ACS by periodontal diagnosis. ● = outlier value; \* = extreme outlier values. The bottom and top of boxes are 25<sup>th</sup> and 75<sup>th</sup> percentiles and the line in the middle of the box is the 50<sup>th</sup> percentile. The ends of the whiskers represent one SD above and below the mean of the data.

association between bone-loss score data and the risk of a recurrence of ACS (crude OR: 1.1; 95% CI: 0.55 to 2.37;  $P=0.55$ ). The crude OR for bleeding on probing and a recurrence of ACS was 1.0:1 (95% CI: 0.37 to 2.79;  $P=0.99$ ).

#### **Analyses of Baseline Medical and Periodontal Conditions in Association With Future ACS (control group with or without a future ACS included)**

Data regarding characteristics of subjects in the original ACS group remaining stable over the following 3 years, and those in the ACS group with a recurrent ACS event in the following 3 years are presented in Table 1. Trends of higher serum hsCRP levels ( $P=0.06$ ) and a higher proportion of sites with PD  $\geq 6$  mm ( $P=0.06$ ) were found among subjects with stable conditions. Statistical analyses failed to demonstrate any other differences in variable values between stable control subjects and those with a first event of ACS. Within the stable subjects of the control group, statistical analyses failed to demonstrate differences in serum hsCRP levels between subjects with or without an evidence of alveolar bone loss. However, significantly higher serum creatinine levels were found in subjects with alveolar bone loss at the 10% and

20% levels ( $P<0.001$ ) and at the 30% to 50% levels ( $P<0.01$ ). Consistent trends of differences among subjects in the control group who later developed ACS had more evidence of alveolar bone loss (43% versus 21% in healthy controls), and higher serum creatinine values (mean difference: 4.0  $\mu\text{mol/l}$ ) were found.

#### **Analyses of Associations Between Future ACS and Baseline Medical and Periodontal Conditions Among Subjects Considered Healthy at Baseline Who Remained Healthy, and Among Subjects in this Group Presenting With ACS Within the Following 3 Years**

In these analyses, all subjects with future ACS were included in the future ACS group ( $n=73$ ) and all stable former ACS and stable control subjects were included in the new control group ( $n=251$ ). Analyses by independent  $t$  tests (equal variance was not assumed) identified significantly higher counts of HDL ( $P<0.05$ ), creatinine ( $P<0.01$ ), and WBCs ( $P<0.001$ ) among subjects with future ACS (Table 2). In addition, analyses by Mann-Whitney  $U$  tests demonstrated that the alveolar bone loss as defined by the 10% to 50% level was more common among subjects with future ACS ( $P<0.001$ ). Analysis by stepwise binary forward

**Table 2.****Subject-Characteristic Values of Disease Surrogate Markers in Control Subjects With Stable, Healthy Conditions and Subjects With a First Event of ACS in 3 Years**

Variable	Status		Significance	
	Healthy	ACS in 3 Years	95% CI	P
Male gender (%)	89.3	85.6	–	0.75 (NS)
Age (years; mean $\pm$ SD)	60.1 $\pm$ 8.8	64.1 $\pm$ 7.9	–11.3 to 6.6	0.23 (NS)
Smoking (years; mean $\pm$ SD)	14.9 $\pm$ 16.1	18.3 $\pm$ 22.3	–26.7 to 20.0	0.73 (NS)
Creatinine ( $\mu$ mol/l; mean $\pm$ SD)	76.5 $\pm$ 14.1	80.6 $\pm$ 8.7	–12.2 to 4.1	0.29 (NS)
Serum hsCRP (mg/l; mean $\pm$ SD)	2.7 $\pm$ 3.7	1.8 $\pm$ 0.8	–6.33 to 12.81	0.06
HDL (mg/dl; mean $\pm$ SD)	1.51 $\pm$ 0.46	1.51 $\pm$ 0.26	–0.24 to 0.24	0.99 (NS)
LDL (mg/dl; mean $\pm$ SD)	3.24 $\pm$ 0.92	3.46 $\pm$ 1.00	–1.10 to 0.74	0.66 (NS)
Triglycerides (mg/dl; mean $\pm$ SD)	1.71 $\pm$ 1.70	1.17 $\pm$ 0.07	0.03 to 1.04	0.05
Cholesterol (mg/dl; mean $\pm$ SD)	5.49 $\pm$ 1.00	5.47 $\pm$ 1.26	–1.14 to 1.19	0.96 (NS)
WBC count ( $\times 10^9$ /l; mean $\pm$ SD)	6.34 $\pm$ 1.64	6.54 $\pm$ 0.72	–0.84 to 0.54	0.72 (NS)
Serum HbA1c (%; mean $\pm$ SD)	4.69 $\pm$ 1.14	4.87 $\pm$ 0.87	–0.84 to 0.47	0.55 (NS)
Bone loss at 30% cutoff (periodontitis) (%; mean $\pm$ SD)	21.1 $\pm$ 44.1	42.9 $\pm$ 53.4	–	0.17 (NS)
BOP (gingivitis) (%; mean $\pm$ SD)	49.5 $\pm$ 20.3	59.0 $\pm$ 23.1	–30.8 to 12.0	0.33 (NS)
BOP at >20% of sites (%; mean $\pm$ SD)	89.6 $\pm$ 30.5	100.0 $\pm$ 0.0	–	0.37 (NS)
Sites with PD $\geq$ 6 mm (%; mean $\pm$ SD)	2.3 $\pm$ 4.3	0.85 $\pm$ 1.2	–0.1 to 2.9	0.06

– = not applicable; NS = not statistically significant; BOP = bleeding on probing.

regression (Wald statistics) identified WBC counts ( $\beta = 0.21$ ;  $P < 0.001$ ) and alveolar bone loss at the 30% cutoff level ( $\beta = 1.26$ ;  $P < 0.001$ ) as explanatory variables of a future ACS event.

#### **Periodontitis Defined by Alveolar Bone Loss at the 30% Cutoff Level and Serum Variables**

**Baseline subjects with ACS.** Statistically higher HDL values were found among subjects who did not have a diagnosis of periodontitis by the bone loss definition (mean difference: 0.14 mg/dl; 95% CI: 0.01 to 0.25;  $P < 0.05$ ). Subjects with periodontitis were also older (mean difference: 6.4 years; 95% CI: 3.2 to 9.5;  $P < 0.001$ ).

**Baseline control subjects.** Significantly higher serum values were found among subjects with periodontitis defined as having  $\geq 30\%$  of surfaces with defined bone loss and creatinine values (mean difference: 6.5  $\mu$ mol/l; 95% CI: 1.6 to 11.5;  $P < 0.01$ ), LDL with higher scores among subjects with bone loss (mean difference: 0.44 mg/dl; 95% CI: 0.02 to 0.85;  $P < 0.05$ ), and HDL with higher scores (mean dif-

ference: 0.14 mg/dl; 95% CI: 0.01 to 0.27;  $P < 0.05$ ). Subjects with periodontitis were also older (mean difference: 5.4 years; 95% CI: 2.4 to 8.3;  $P < 0.001$ ). Statistical analyses failed to demonstrate that serum hsCRP in subjects with periodontitis defined by the various alveolar bone loss cutoff levels differed from those who did not have periodontitis (mean CRP scores in both groups: 2.6 ml/l).

**All subjects included.** Significantly higher serum values were found in subjects with a diagnosis of periodontitis for hsCRP (mean difference: 7.7 mg/l; 95% CI: 2.2 to 13.1;  $P < 0.001$ ), creatinine (mean difference: 6.0  $\mu$ mol/l; 95% CI: 2.2 to 9.8;  $P < 0.001$ ), and WBCs (mean difference: 1.8  $\times 10^9$ /l; 95% CI: 1.2 to 2.3;  $P < 0.001$ ). Subjects with periodontitis were also older (mean difference: 4.6 years; 95% CI: 2.7 to 6.5;  $P < 0.001$ ).

**Explanatory factors predicting a future ACS event and independent of previous ACS or not.** Analysis by GLM multivariate analysis, controlling for age and in the prediction of a future ACS event, identified that WBC counts ( $F = 20.6$ ;  $P < 0.001$ ), alveolar bone

loss at the 30% cutoff level ( $F = 17.6$ ;  $P < 0.001$ ), and serum creatinine counts ( $F = 4.5$ ,  $P < 0.05$ ) were explanatory of a future ACS event.

## DISCUSSION

All subjects in this 3-year follow-up study with a previous history of ACS received postemergency care consistent with known treatment methods to reduce future risks of ACS.<sup>23</sup> During the first 3-year period, the medical interventions prevented a recurrence of ACS in 60% of the cases.

Several studies<sup>24-27</sup> reported similar rates of ACS recurrence. In the present study, the recurrence of ACS was higher than the rate reported elsewhere<sup>13</sup> of serious adverse cardiovascular events in subjects with periodontitis. This may be explained by differences in the definitions of ACS and periodontitis, treatments, and study-population risk factors.

The socioeconomic burden may contribute significantly to the risk of ACS such that the infectious burden may be of less importance as an explanatory factor in cardiovascular disease.<sup>28</sup> For this reason, we designed the study so that socioeconomic conditions, including education and working and living conditions, were matched such that the control subjects and subjects with ACS presented with similar conditions.

Due to the inclusion criteria used for the control subjects, we were unable to assess the impact of smoking, gender, marital status, and socioeconomic status as risk factors in ACS and periodontitis. Smoking has been considered a risk factor for periodontitis and confounder for cardiovascular disease.<sup>29,30</sup> The present study failed to demonstrate that smoking habits had a significant impact on the recurrence of ACS. This is most likely an effect of the enrollment strategy used in the present study.

It was proposed that periodontal intervention has effects on some surrogate measures of cardiovascular disease risk.<sup>7-12</sup> For example, data suggest that immediately after periodontal intervention, an acute response to non-surgical periodontal debridement and immediate elevation of serum hsCRP levels may occur.<sup>12,31-33</sup> The present study demonstrated that the serum hsCRP levels at the time of original hospital admission were higher in subjects with ACS than in control subjects and were higher in subjects with periodontitis. However, serum hsCRP was not included in our model for future ACS events. This is most likely explained by the fact that the medical intervention included therapy to reduce the extent of systemic inflammation by prescription of statins.<sup>34,35</sup> Because hsCRP, as an acute-phase reactant, is elevated in individuals with ACS, an added impact of periodontitis may not be sufficient to result in a statistical difference by peri-

odontal status. In the systemically healthy control group, the present data consistently identified that subjects in the control group with periodontitis had higher serum WBC counts, serum creatinine levels, and HDL levels and lower LDL levels than control subjects with periodontitis. This is in agreement with others<sup>4,12,15,31,32,36,37</sup> who identified that periodontitis is reflected by such serum markers.

A report suggested<sup>38</sup> that the infectious burden is a factor in cardiovascular diseases. Other reports<sup>39-42</sup> suggested that bacteria specifically associated with periodontitis may be associated with cardiovascular disease. We previously reported that subgingival levels of *T. forsythia* and several *Streptococci* species were significantly higher in subjects with a diagnosis of ACS.<sup>15</sup> In the present study, analyses of factors predicting the recurrence of ACS or a new first event of ACS failed to include all of the bacterial species studied.

Age is an important factor associated with periodontitis and cardiovascular diseases. Older patients have many diseases, and many geriatric subjects with ACS have multiorgan failures.<sup>43</sup> Studies<sup>44-48</sup> demonstrated that the prevalence of periodontitis among older subjects is high. When controlling for age, three factors (WBC counts, creatinine levels, and periodontitis defined by bone loss) remained as explanatory of a future ACS event. Serum creatinine values are associated with kidney functions and predictive of serious ACS.<sup>49-51</sup> The findings in the present study are consistent with others<sup>52</sup> and suggested a close relationship between systemic inflammatory effects of periodontal infections, serum creatinine values, and cardiovascular disease.

Our findings on the relationship between WBC counts and ACS and periodontitis are consistent with what we previously reported for a subset of the subjects.<sup>53</sup> This relationship between WBC counts and periodontitis is consistent with a systemic inflammatory impact as the result of periodontitis.<sup>54,55</sup> Data suggest that serum WBC counts are independent predictors of left-ventricular systolic dysfunction after an ACS event.<sup>56</sup> High WBC counts in middle-aged men have also been associated with an increased long-term incidence of hospitalizations as a result of heart failure.<sup>57</sup> Serum creatinine values reflect kidney function and are linked to serious adverse outcomes in individuals with ACS.<sup>58</sup>

The fact that the statistical analyses failed to demonstrate differences in values by study parameters between remaining healthy control subjects and those who later developed ACS is most likely an effect of lacking statistical power. The trends of differences indicated that subjects in the control group who later developed ACS had more evidence of periodontitis defined by alveolar bone loss at  $\geq 30\%$  of interproximal

surfaces  $\geq 4.0$  mm (43% in baseline healthy controls but who had an ACS event in 3 years versus 21% in healthy controls).

## CONCLUSIONS

Recurrent ACS was suggested to be predicted by serum WBC counts, serum creatinine levels, and a diagnosis of periodontitis. Significantly higher counts of putative pathogens were found in subjects with ACS but these do not predict the risk of a future ACS event.

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## REFERENCES

- Rosamond W, Flegal K, Friday G, et al. Heart disease and stroke statistics – 2007 update: A report from the American Heart Association Statistics Committee and Stroke Statistics Subcommittee. *Circulation* 2007; 115:e69-e171.
- Kuller LH. Nutrition, lipids, and cardiovascular disease. *Nutr Rev* 2006;64:S15-S26.
- EUROASPIRE study group. A European Society of Cardiology survey of secondary prevention of coronary heart disease: Principal results. European action on secondary prevention through intervention to reduce events. *Eur Heart J* 1997;18:1569-1582.
- Persson GR, Persson RE. Cardiovascular disease and periodontitis. An update on the associations and risk. *J Clin Periodontol* 2008;35(Suppl. 8):362-379.
- Bahekar AA, Singh S, Saha S, Molnar J, Arora R. The prevalence and incidence of coronary heart disease is significantly increased in periodontitis: A meta-analysis. *Am Heart J* 2007;154:830-837.
- Mustapha IZ, Debrey S, Oladubu M, Ugarte R. Markers of systemic bacterial exposure in periodontal disease and cardiovascular disease risk: A systematic review and meta-analysis. *J Periodontol* 2007;78:2289-2302.
- Blum A, Front E, Peleg A. Periodontal care may improve systemic inflammation. *Clin Invest Med* 2007; 30:E114-E117.
- Elter JR, Hinderliter AL, Offenbacher S, et al. The effects of periodontal therapy on vascular endothelial function: A pilot trial. *Am Heart J* 2006;151:47e1-47e6.
- Mercanoglu F, Oflaz H, Öz O, et al. Endothelial dysfunction in patients with chronic periodontitis and its improvement after initial periodontal therapy. *J Periodontol* 2004;75:1694-1700.
- Montebugnoli L, Servidio D, Miaton RA, et al. Periodontal health improves systemic inflammatory and haemostatic status in subjects with coronary heart disease. *J Clin Periodontol* 2005;32:188-192.
- Seinost G, Wimmer G, Skerget M, et al. Periodontal treatment improves endothelial dysfunction in patients with severe periodontitis. *Am Heart J* 2005;149: 1050-1054.
- Tonetti MS, D'Aiuto F, Nibali L, et al. Treatment of periodontitis and endothelial function. *N Engl J Med* 2007;356:911-920.
- Beck JD, Couper DJ, Falkner KL, et al. The Periodontitis and Vascular Events (PAVE) pilot study: Adverse events. *J Periodontol* 2008;79:90-96.
- Persson GR, Ohlsson O, Pettersson T, Renvert S. Chronic periodontitis, a significant relationship with acute myocardial infarction. *Eur Heart J* 2003;24: 2108-2115.
- Renvert S, Pettersson T, Ohlsson O, Persson GR. Bacterial profile and burden of periodontal infection in subjects with a diagnosis of acute coronary syndrome. *J Periodontol* 2006;77:1110-1119.
- Engelbrecht SP, Lamster IB, Elkind MS, et al. Radiographic measures of chronic periodontitis and carotid artery plaque. *Stroke* 2005;36:561-566.
- Geismar K, Stoltze K, Sigurd B, Gyntelberg F, Holmstrup P. Periodontal disease and coronary heart disease. *J Periodontol* 2006;77:1547-1554.
- Rech RL, Nurkin N, da Cruz I, et al. Association between periodontal disease and acute coronary syndrome. *Arq Bras Cardiol* 2007;88:185-190.
- Persson RE, Hollender LG, Persson GR. Assessment of alveolar bone levels from intraoral radiographs in subjects between ages 15 and 94 years seeking dental care. *J Clin Periodontol* 1998;25:647-654.
- Socransky SS, Smith C, Martin L, Paster BJ, Dewhirst FE, Levin AE. "Checkerboard" DNA-DNA hybridization. *Biotechniques* 1994;17:788-792.
- Socransky SS, Haffajee AD, Smith C, et al. Use of checkerboard DNA-DNA hybridization to study complex microbial ecosystems. *Oral Microbiol Immunol* 2004;19:352-362.
- Katsoulis J, Lang NP, Persson GR. Proportional distribution of the red complex and its individual pathogens after sample storage using the checkerboard DNA-DNA hybridization technique. *J Clin Periodontol* 2005; 32:628-633.
- Bassand JP, Hamm CW, Ardissino D, et al. Guidelines for the diagnosis and treatment of non-ST-segment elevation acute coronary syndromes: The task force for the diagnosis and treatment of non-ST-segment elevation acute coronary syndromes of the European Society of Cardiology. *Eur Heart J* 2007;28:1598-1660.
- Chew DP, Huynh LT, Liew D, et al. Potential survival gains in the treatment of myocardial infarction. *Heart* 2009;95:1844-1850.
- Jiménez-Navarro MF, Muñoz-García A, Ramirez-Marrero MA, et al. Preinfarction angina prior to first myocardial infarction does not influence long-term prognosis: A retrospective study with subgroup analysis in elderly and diabetic patients. *Clin Cardiol* 2009; 32:E62-E65.
- Furtado MV, Rossini AP, Campani RB, et al. Interleukin-18: An independent predictor of cardiovascular events in patients with acute coronary syndrome after 6 months of follow-up. *Coron Artery Dis* 2009;20:327-331.
- Motivala AA, Tamhane U, Ramanath VS, et al. A prior myocardial infarction: How does it affect management and outcomes in recurrent acute coronary syndromes? *Clin Cardiol* 2008;31:590-596.
- Steptoe A, Shamaei-Tousi A, Gylfe A, Henderson B, Bergstrom S, Marmot MM. Socioeconomic status,

- pathogen burden, and cardiovascular disease risk. *Heart* 2007;93:1567-1570.
29. Fisher S, Kells L, Picard JP, et al. Progression of periodontal disease in a maintenance population of smokers and non-smokers: A 3-year longitudinal study. *J Periodontol* 2008;79:461-468.
  30. Hujoel PP, Drangsholt M, Spiekerman C, DeRouen TA. Periodontal disease and coronary heart disease risk. *JAMA* 2000;284:1406-1410.
  31. D'Aiuto F, Parkar M, Andreou G, et al. Periodontitis and systemic inflammation: Control of the local infection is associated with a reduction in serum inflammatory markers. *J Dent Res* 2004;83:156-160.
  32. D'Aiuto F, Nibali L, Parkar M, Suvan J, Tonetti MS. Short-term effects of intensive periodontal therapy on serum inflammatory markers and cholesterol. *J Dent Res* 2005;84:269-273.
  33. Hussain Bokhari SA, Khan AA, Tatakis DN, Azhar M, Hanif M, Izhar M. Non-surgical periodontal therapy lowers serum inflammatory markers: A pilot study. *J Periodontol* 2009;80:1574-1580.
  34. Hara H, Nakamura M, Yokouchi I, et al. Aggressive statin therapy in multicenter and effectiveness for the reduction of intra-myocardial damage caused by non-ST elevation acute coronary syndrome: AMERICA study. *Ther Adv Cardiovasc Dis* 2009;3:357-365.
  35. Lewandowski M, Kornacewicz-Jach Z, Millo B, et al. The influence of low dose atorvastatin on inflammatory marker levels in patients with acute coronary syndrome and its potential clinical value. *Cardiol J* 2008;15:357-364.
  36. Ramirez-Tortosa MC, Quiles JL, Battino M, et al. Periodontitis is associated with altered plasma fatty acids and cardiovascular risk markers. *Nutr Metab Cardiovasc Dis* 2010;20:133-139.
  37. Buhlin K, Hultin M, Norderyd O, et al. Periodontal treatment influences risk markers for atherosclerosis in patients with severe periodontitis. *Atherosclerosis* 2009;206:518-522.
  38. Espinola-Klein C, Rupprecht HJ, Blankenberg S, et al. Impact of infectious burden on extent and long-term prognosis of atherosclerosis. *Circulation* 2002;105:15-21.
  39. Honda T, Oda T, Yoshie H, Yamazaki K. Effects of *Porphyromonas gingivalis* antigens and proinflammatory cytokines on human coronary artery endothelial cells. *Oral Microbiol Immunol* 2005;20:82-88.
  40. Nonnenmacher C, Stelzel M, Susin C, et al. Periodontal microbiota in patients with coronary artery disease measured by real-time polymerase chain reaction: A case-control study. *J Periodontol* 2007;78:1724-1730.
  41. Aimetti M, Romano F, Nessi F. Microbiologic analysis of periodontal pockets and carotid atheromatous plaques in advanced chronic periodontitis patients. *J Periodontol* 2007;78:1718-1723.
  42. Pucar A, Milasin J, Lekovic V, et al. Correlation between atherosclerosis and periodontal putative pathogenic bacterial infections in coronary and internal mammary arteries. *J Periodontol* 2007;78:677-682.
  43. Taneva E, Bogdanova V, Shtereva N. Acute coronary syndrome, comorbidity, and mortality in geriatric patients. *Ann N Y Acad Sci* 2004;1019:106-110.
  44. Borges-Yañez SA, Irigoyen-Camacho ME, Maupomé G. Risk factors and prevalence of periodontitis in community-dwelling elders in Mexico. *J Clin Periodontol* 2006;33:184-194.
  45. Holm-Pedersen P, Russell SL, Avlund K, Viitanen M, Winblad B, Katz RV. Periodontal disease in the oldest-old living in Kungsholmen, Sweden: Findings from the KEOHS project. *J Clin Periodontol* 2006;33:376-384.
  46. Krstrup U, Erik Petersen P. Periodontal conditions in 35-44 and 65-74-year-old adults in Denmark. *Acta Odontol Scand* 2006;64:65-73.
  47. Persson RE, Hollender LG, Powell VL, et al. Assessment of periodontal conditions and systemic disease in older subjects. II. Focus on cardiovascular diseases. *J Clin Periodontol* 2002;29:803-810.
  48. Phipps KR, Chan BK, Jennings-Holt M, et al. Periodontal health of older men: The MrOS dental study. *Gerodontology* 2009;26:122-129.
  49. Eagle KA, Lim MJ, Dabbous OH, et al. A validated prediction model for all forms of acute coronary syndrome: Estimating the risk of 6-month postdischarge death in an international registry. *JAMA* 2004;291:2727-2733.
  50. Mahaffey KW, Yang Q, Pieper KS, et al. Prediction of one-year survival in high-risk patients with acute coronary syndromes: Results from the SYNERGY trial. *J Gen Intern Med* 2008;23:310-316.
  51. Rutherford E, Leslie SJ, Soiza RL. Creatinine and eGFR are similarly predictive of outcome of acute coronary syndrome. *Int J Cardiol* 2010;141:118-120.
  52. Kshirsagar AV, Moss KL, Elter JR, et al. Periodontal disease is associated with renal insufficiency in the Atherosclerosis Risk In Communities (ARIC) study. *Am J Kidney Dis* 2005;45:650-657.
  53. Persson GR, Pettersson T, Ohlsson O, Renvert S. High-sensitivity serum C-reactive protein levels in subjects with or without myocardial infarction or periodontitis. *J Clin Periodontol* 2005;32:219-224.
  54. Taylor BA, Toftler GH, Carey HM, et al. Full-mouth tooth extraction lowers systemic inflammatory and thrombotic markers of cardiovascular risk. *J Dent Res* 2006;85:74-78.
  55. Nibali L, D'Aiuto F, Griffiths G, et al. Severe periodontitis is associated with systemic inflammation and a dysmetabolic status: A case-control study. *J Clin Periodontol* 2007;34:931-937.
  56. Aggelopoulos P, Chrysohoou C, Pitsavos C, et al. Comparative value of simple inflammatory markers in the prediction of left ventricular systolic dysfunction in postacute coronary syndrome patients. *Mediators Inflamm* 2009;2009:826297.
  57. Engström G, Melander O, Hedblad B. Leukocyte count and incidence of hospitalizations due to heart failure. *Circ Heart Fail* 2009;2:217-222.
  58. Goldberg A, Kogan E, Hammerman H, Markiewicz W, Aronson D. The impact of transient and persistent acute kidney injury on long-term outcomes after acute myocardial infarction. *Kidney Int* 2009;76:900-906.
- Correspondence: Dr. G. Rutger Persson, Department of Periodontology, University of Bern, Freiburgstrasse 7 CH 3010, Bern, Switzerland. Fax: 41-31-632 8608; e-mail: rutger.persson@zmk.unibe.ch.
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