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Association of Periodontal Diseases with Elevation of Serum C-reactive Protein and Body Mass Index

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Abstract

Background and aims. C-reactive protein (CRP) is a well-known acute-phase reactant produced by the liver in response to inflammation caused by various stimuli. Periodontal disease is a chronic infection of tooth-supporting structures characterized by attachment loss and alveolar bone loss. The aim of this study was to assess the relationship between serum C-reactive protein levels and periodontal diseases.

Materials and methods. The study was conducted on 166 patients referring to Tabriz Faculty of Dentistry. The age range was between 35 and 59 years. 83 subjects with periodontitis according to NHANES III index as test group and 83 healthy individuals as controls participated in this study. Body mass index (BMI), waist circumference (WC), probing depth, attachment loss and CRP levels were measured in both test and control groups. Data was analyzed with Student's *t*-test, odds ratio (OR), Chi-square test and Spearman's correlation coefficient, using SPSS 13.0 software.

Results. The results revealed a statistically significant difference between all of the analyzed variables in test and control groups ($P < 0.05$). Classifying the test subjects into two subgroups (subjects with CRP $\geq$ 3 mg/l and subjects with CRP < 3 mg/l), the highest OR in females belonged to WC (OR = 6.4; 95% CI: 1.18-35.2, P = 0.02) and in males to obesity (OR = 4.8; 95% CI: 0.65-35.19, P = 0.05). Considering the correlation between obesity, overweight, WC and CRP with probing depth and attachment loss denoted that obesity presented the highest ($r = 1$, $P = 0.00$) and overweight the lowest ($r = 0.4$, $P = 0.07$) association. In females, CRP was related to the severity of periodontitis and attachment loss ($r = 0.662$, $P = 0.00$). Excluding overweight, the association between all the variables was statistically significant ($P < 0.05$).

Conclusion. Our findings indicate that periodontal disease is correlated with CRP elevation and diseases associated with obesity.

Key words: **BMI, C-reactive protein, obesity, periodontal disease.**

Introduction

Periodontal disease is a Gram-negative chronic infection of the tooth-supporting structures in which long-lasting periodontitis ultimately is followed by alveolar bone loss.[1] Historically, it was believed that all individuals were uniformly susceptible to developing periodontal disease and that accumulation of plaque, poor oral hygiene and perhaps occlusal trauma were sufficient to initiate periodontitis. However, recently it has become accepted that periodontal disease is caused by specific bacterial infections and individuals are not uniformly susceptible to these infections.[2] In addition, an association between periodontal diseases and systemic conditions such as coronary heart disease has been revealed in the past decades and several hypotheses have been proposed to explain this relationship, one of which is that of systemic inflammation and increased circulating cytokines.[3] Based on epidemiological and experimental evidences, it is suggested that inflammation plays an important role in the pathogenesis of atherosclerosis, which may lead to cardiovascular diseases.[4] C-reactive protein (CRP) is a well-known acute-phase reactant produced by the liver in response to inflammation due to various stimuli. In acute inflammation, serum CRP levels exceed 100 mg/l, and the level decreases in chronic inflammation.[5]

The cause of small elevations in serum CRP levels in healthy people is not clear. Systemic or localized infections caused by microorganisms such as Chlamydia pneumonia, Helicobacter pylori, and cytomegalovirus have been suggested as causes of CRP elevation which leads to atherosclerotic diseases.[5]

Periodontitis as a common chronic inflammatory disease caused by Gram-negative pathogens such as Porphyromonas gingivalis and Tannerella forsythensis elevate serum CRP levels. Serum CRP level is a significant predictor of cardiovascular disease.[6] Moreover, obesity, the most common nutritional disorder increasing world wide, is a significant risk factor for numerous adult diseases including cardiovascular problems.[7]

Adipose tissue excretes pro-inflammatory cytokines such as interleukin-6 (IL-6) and tumor necrosis factor α (TNF-α).[8] TNF-α induces IL-6 synthesis, a prime regulator of CRP synthesis.[9] Among inflammatory mediators found in gingival crevicular fluid, TNF-α plays an integral role in the pathogenesis of periodontitis. It has also been reported that CRP is related to body mass index (BMI) and waist circumference (WC).[10] On the other hand, an association between obesity and periodontal disease has been indicated.[11]

Genetic background, lifestyle, and specially diet vary between countries. Therefore, the aim of this study was to answer the question whether there is any association between BMI, serum CRP and periodontal disease.

Materials and Methods

In this analytical case-control study, we evaluated a population of 166 patients, aged 35 to 59, referring to Tabriz University of Medical Sciences Faculty of Dentistry. The study design and the applied methods were accepted by the Ethics Committee of Tabriz University of Medical Sciences. The test group consisted of 83 patients with periodontal disease (44 females, 39 males) and control group comprised of 83 healthy subjects.

Samples were selected based on the following criteria:

1. No systemic diseases
2. No smoking, alcohol or drug use
3. No use of oral contraceptives or other medicine
4. No pregnancy
5. No use of antibiotics during the past 3 months

Diagnosis of periodontal disease was established when one or more regions had attachment loss of 3 mm or more and pocket depth of more than 4 mm, according to NHANES III index.

One quadrant of the upper jaw and one quadrant of the lower jaw were randomly chosen and the teeth were examined. The distance between gingival margin and the sulcus base (probing depth) and the distance between the gingival margin and CEJ in distofacial, mesiofacial and midfacial surfaces of each tooth were measured.

BMI and WC, two indicators of obesity, were considered in this study. BMI is defined as the individuals' body weight divided by the square of their height and measured in kg/m^2. To obtain BMI, the heights of all subjects were

measured and they were accurately weighed with a digital balance. WC was measured in centimeter.

Subjects were categorized in 4 groups based on the WHO classification of BMI: One is considered underweight with BMI lower than 18.5 kg/m^2, normal with 18.5 < BMI < 24.5, overweight with 25 < BMI < 29.5 and obese with BMI higher than 30 kg/m^2. WC is an indicator of intra-abdominal fat tissue. Measurements higher than 89 cm for women and 102 cm for men are considered as obesity.[12]

CRP was measured using a high sensitive ELISA kit (IBL, Hamburg, Germany). Patients were fasted overnight. Blood samples were collected in the morning, and centrifuged to extract serum. According to the manufacturer's instructions, the wells of the microtitre plate were coated with polyclonal antibodies directed against C-reactive protein. In a first incubation step, the CRP was bound to the coated polyclonal rabbit antibodies. In a second incubation step, a peroxidase-labeled CRP antibody was added. After a washing step to remove all unbound substances, the solid phase was incubated with tetramethylbenzidine substrate. An acidic stopping solution was then added which converted the color to yellow. The intensity of the yellow color is directly proportional to the concentration of CRP in the sample. A dose response curve of the absorbance (at 450 nm) unit versus concentration was generated. CRP, present in the patient samples, was determined directly from this calibration curve. The combination of two specific antibodies in this method drastically reduces the possibility of false-negative results.

Differences between the mean values were evaluated using Student's *t*-test. Odds ratios for individuals with CRP levels higher than 3 mg/l, who are in high risk for cardiovascular diseases, were calculated and Chi-square test was used to assess the differences between subjects with CRP levels higher and lower than 3 mg/l. Spearman's correlation coefficient was applied to determine the relationship between severity of periodontal disease and obesity, overweight, WC and CRP variables. SPSS 13.0 software was used for data analysis.

Results

Mean values of BMI, WC, attachment loss, probing depth, and CRP in the test group were higher than those of the control group in both genders and the differences were statistically significant (P = 0.000) (Table 1).

Table 1. Mean values of BMI, WC, AL, PD and CRP in test and control groups

	Female		Male	
Mean value	**Test**	**Control**	**Test**	**Control**
Age	44 ± 5.8	45 ± 6.11	45 ± 5.5	45 ± 5.5
BMI	29 ± 3.8	24 ± 2.5	28 ± 3.2	26 ± 1.6
WC	96 ± 10	84 ± 8.9	105 ± 9.9	88 ± 6.1
AL	4.43 ± 1	1.69 ± 0.61	4.26 ± 0.96	1.22 ± .73
PD	4.77 ± 0.7	2.1 ± 0.74	4.6 ± 0.66	1.94 ± .79
CPR	3.57 ± 1.62	1.7 ± 1.5	2.46 ± 1.67	1.4 ± .94

BMI, body mass index; WC, waist circumference; AL, attachment loss; PD, probing depth; CRP, C-reactive protein.

There were significant associations between WC and obesity variables and CRP in both genders in this study ($P < 0.05$) (Table 2). The association was more prominent in females with the strongest association seen between WC and CRP ($P = 0.02$) and the weakest between overweight and CRP ($P = 0.53$). Likewise, the weakest association in males was attributed to overweight, while obesity and WC showed approximately equal associations with CRP.

Evaluation of the relationship between pocket depth and attachment loss with obesity, overweight, WC and CRP revealed that obesity owned the strongest association in both genders. Except for the overweight, the correlation of variables was statistically significant ($P < 0.05$). In addition, the results showed that the association between the noted variables and probing depth was identical to that of attachment loss (Table3).

Table 2. Association between overweight, obesity, waist circumference and CRP in females and males

	Female		Male	
Variable	**OR (95% CI)**	**P-value**	**OR (95% CI)**	**P-value**
Overwight	0.75 (0.14-3.9)	0.53	0.57 (0.061-2.52)	0.60
Obesity	5.62 (1.05-0.13)	0.036	4.8 (0.65-35.19)	0.05
Waist circunference	6.4 (1.18- 35.2)	0.02	4.33 (0.95-19.57)	0.05

OR, odds ratio; CI, confidence interval.

Table 3. Correlation of severity of periodontal disease with BMI, WC and CPR in females and males

Variable	**Female**				**Male**			
	AL		**PD**		**AL**		**PD**	
	r	**P-value**	**r**	**P-value**	**r**	**P-value**	**r**	**P-value**
Overwieght	0.200	0.21	0.259	0.17	0.153	0.26	0.277	0.18
Obesity	1	0.000	1	0.000	1	0.000	1	0.000
WC	0.622	0.000	0.622	0.000	0.327	0.04	0.422	0.04
CRP	0.662	0.000	0.587	0.000	0.470	0.04	0.453	0.04

PD, probing depth; AL, attachment loss; BMI, body mass index; WC, waist circumference; CRP, C-reactive protein.

Discussion

The results of the present study indicate that obesity, WC and overweight are associated with CRP levels which is in accordance with the results of a previous study.[11]

Furthermore, the association of WC and higher CRP levels has been shown to be more significant than those of obesity and overweight which could be due to the fat accumulation in abdominal region among females. Fat accumulation in upper body is associated with more health problems than lower body obesity.[10]

Development of cardiovascular diseases is affected by many factors, among which chronic inflammation is of critical importance.[4] It has been suggested that CRP is an important risk factor for coronary heart disease.[13] It facilitates the formation of atherosclerotic lesions via activating complement system and production of foam cells.[14] A study investigating the direct and indirect effects of periodontal pathogens on cardiovascular system suggested elevation of CRP levels in periodontitis.[15]

Correlation coefficients of attachment loss and probing depth with obesity, overweight, WC and CRP were determined in the present study. The results demonstrated that, except for the overweight, these variables were significantly associated with attachment loss and probing depth, with obesity presenting the strongest correlation. Considering that obesity may lead to periodontal disease through various mechanisms and obesity-associated diseases e.g. diabetes may exacerbate periodontitis,[10] the achieved results were not unexpected. It has been reported that adipose tissue excretes pro-inflammatory cytokines such as IL-6 and TNF-α.[8] The latter, found in gingival crevical fluid, plays an important role in pathogenesis of periodontitis.[10]

The weak association between overweight and periodontal disease, on the other hand, may be attributed to the relatively broad range of "overweight" (25-29 kg/m^2) in BMI definition. Dalla Vecchia et al[16] using the criteria "30% of the teeth or more presenting CAL > 5 mm" to determine the presence of periodontitis found that obese women had 1.65 times more chances of having periodontitis after adjustment for age and smoking.

The results of the present study indicate a significant correlation between attachment loss, probing depth and CRP. Noack et al[17] observed statistically significant increase in CRP levels in 174 subjects with periodontal disease and a positive correlation between elevated levels of CRP, attachment loss and probing depth. They found increased CRP levels in deeper pockets which could be due to the presence of periodontal gram-negative pathogens like Porphyromonas gingivalis in subgingival region.[17] Yamazaki et al[18] indicated an association between increasing of CRP and periodontal disease; however, they did not find any relationship between the severity of periodontitis and CRP elevation in a sample size of 24 patients ($P = 0.056$). It seems that increasing the sample size might lead to a statistically significant difference between the test and control groups, as Mattila et al[19] reported a significant difference between the test and control groups with a sample group of 30 individuals which is consistent with the results of the present study. In addition, Slade et al[20] indicated that extensive periodontal disease and BMI are jointly associated with increased CRP levels.

As true with all analytical case-control studies, it was not feasible to demonstrate which variable was affected by the other in the present study. Also, the relationship between periodontal disease and atherosclerosis was not directly observed in the present study.

In conclusion, our findings demonstrated correlations between obesity, waist circumference, elevated CRP levels and severity of periodontitis.

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