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Periodontal disease and hypertriglyceridemia in Japanese subjects: potential association with enhanced lipolysis

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Abstract

Although periodontal disease may be associated with increased risk for atherosclerosis, the mechanism by which the disease causes atherosclerosis is still unknown. The candidates contributing to atherosclerosis in periodontal disease include low-grade inflammation such as C-reactive protein (CRP) and insulin resistance. A previous study demonstrated that periodontal therapy leads to an improvement in CRP as well as insulin resistance, indicating the relationship between periodontal disease and low-grade inflammation or insulin resistance. On the other hand, we previously demonstrated that serum triglyceride (TG) per se is independently associated with CRP or insulin resistance in Japanese populations with a body mass index (BMI) of 21.5 to 27.0 (midrange BMI). To the best of our knowledge, however, the relationship between periodontal disease and serum TG is not fully clarified. The first aim of the present study is to investigate whether periodontal disease is associated with serum TG in Japanese subjects with midrange BMI. If so, another aim of the study is to determine which mechanism is responsible for the association between periodontal disease and serum TG in these subjects. We have performed a periodontal examination in the Ogaki metabolic syndrome medical examination. One hundred sixty-two participants from 40 to 74 years old (56 men and 106 women; mean age, 66.43 ± 6.25 years) were enrolled in the study. Besides medical examination, oral panoramic radiograph was taken for all participants. Average bone score was also calculated. Periodontal bone destruction increased according to the age of the participants ($r = 0.227$, $P < .004$, Spearman correlation coefficient). Periodontal bone destruction was also associated with serum TG levels ($r = 0.299$, $P = .000$). This association was more evident in subjects with midrange BMI ($r = 0.332$, $P < .001$). In subjects with midrange BMI, TG was not correlated with BMI or waste circumstances. Furthermore, TG was not associated with age itself in the midrange BMI group. We then investigated the lipolytic activity of endotoxin in cocultures of adipocytes and macrophages. Low-dose lipopolysaccharide dose-dependently increased lipolytic activity in cocultures, and this activity was neutralized by anti–tumor necrosis factor α neutralizing antibodies. These results suggest that periodontal infection, especially bacterial endotoxinemia, is associated with enhanced lipolysis and subsequent up-regulation of circulating TG in Japanese with midrange BMI.

It has been suggested that chronic low-grade infectious disease such as *Chlamydia* infection and/or periodontal disease is associated with enhanced risk for atherosclerosis and subsequent myocardial infarction [1,2]. It is well known that these diseases are associated with elevated levels of circulating C-reactive proteins as measured by highly sensitive assay (hsCRP) [3,4]. High-sensitivity CRP elevation has been documented as a sensitive marker for predicting future development of myocardial infarction [5,6]. Whereas several lines of literature have shown a direct

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potential involvement with CRP molecules in accelerating atherosclerotic changes, it remains to be clarified whether elevated CRP is a sensitive marker for future development of myocardial infarction or CRP molecule itself plays critical roles in promoting atherosclerosis [7]. Furthermore, a mechanistic association between low-grade infectious disease and enhanced risk for atherosclerosis is largely unknown. We recently performed metabolic syndrome medical examination in a suburban area of Gifu Prefecture (Ogaki City). At the same time, we also performed dental checkup under the support of the Ogaki City administration. We were able to take oral panoramic radiograph for all participants to check the status of periodontal tissue destruction. The first purpose of this study was to compare cumulative periodontal bone destruction as measured by oral panoramic radiograph with medical parameters to see potential association between periodontal disease and parameters associated with atherosclerosis. The second aim was to reveal the potential underlying mechanisms by in vitro approach if any association was observed.

1. Subjects and methods

One hundred sixty-two Japanese subjects residing in Ogaki City, suburban area of Gifu Prefecture, aged from 40 to 74 years (56 men and 106 women; mean age, 66.43 ± 6.25 years old) were enrolled in the study. Participants are attendants of a metabolic syndrome medical examination conducted by the health administration of the city. In the year 2009, the city administration also included dental examination to assess the potential association between periodontitis and medical parameters. Therefore, the city offered the cost for taking oral panoramic radiograph for all participants. Age, sex, waste circumstances, and body mass index (BMI) were recorded for all participants; and systolic and diastolic blood pressure was measured. Body mass index was calculated based on the weight (in kilograms) and height (in meters) measured by the nurses. Glycated hemoglobin, high-density lipoprotein cholesterol, low-density lipoprotein cholesterol, triglyceride (TG), creatinine, and parameters indicating liver function such as aspartate aminotransferase, alanine aminotransferase, γ-glutamyltranspeptidase were measured from blood sample. Baseline data of these participants are demonstrated in Table 1.

To assess the cumulative periodontal bone loss, we calculated simplified Schei bone score [8] suggested by the Japan Diabetes Complication and its Prevention (JDCP) prospective study. Briefly, we traced panoramic radiograph and calculated the ratio of alveolar bone resorption rate by measuring the bone loss in cemento-enamel junction to root end on both mesial and distal sides in each tooth. The data on each tooth were expressed by 4 simplified scores (score 1 [no or mild bone loss]: bone resorption rate as 0%-25% of entire root length; score 2 [medium bone loss]: 25%-50%; score 3 [severe bone loss]: 50%-75%; score 4 [extremely severe bone loss]: 75%-100%, respectively), and bone score was expressed by mean resorption score per tooth (JDCP score).

Table 1
Clinical characteristics of the participants

Age (y)	66.43 ± 6.25 (40-74)
Male-female	56:106
TG (mg/dL)	111.26 ± 75.41 (29-718)
LDL cholesterol (mg/dL)	129.22 ± 37.18 (17-384)
HDL cholesterol (mg/dL)	62.46 ± 15.50 (29-130)
BMI (kg/m^2)	23.43 ± 4.08 (16.3-56)
Waste circumstances (cm)	84.18 ± 8.77 (60-111)
HbA_{1c} (%)	5.34 ± 0.85 (2-10.9)
Systolic blood pressure (mm Hg)	132.60 ± 16.36 (86-195)
Diastolic blood pressure (mm Hg)	74.99 ± 10.98 (45-118)
Creatinine (mg/dL)	0.74 ± 0.20 (0.4-1.7)
AST (IU/L)	24.80 ± 12.42 (13-136)
ALT (IU/L)	21.42 ± 14.88 (6-132)
γ-GTP (IU/L)	38.07 ± 46.32 (7-315)

LDL indicates low-density lipoprotein; HDL, high-density lipoprotein; HbA_{1c}, hemoglobin A_{1c}; AST, aspartate aminotransferase; ALT, alanine aminotransferase; γ-GTP; γ-glutamyltranspeptidase.

All statistical analyses were performed by using SPSS (Chicago, IL) software, and intercorrelation between each parameter was evaluated by Spearman correlation coefficient.

1.1. In vitro study

To evaluate lipolytic activity by bacterial endotoxin, we used coculture system between mouse 3T3-L1 adipocytes and mouse macrophage cell line RAW 264. Cocultures were performed by using the Transwell system as described in our previous study [9]. Lipolysis was evaluated by measuring glycerol levels released into culture supernatants following 24 hours after indicated concentration of *Escherichia coli* lipopolysaccharide (LPS) (Sigma, St Louis, MO) stimulation. Serum glycerol levels were measured by lipolysis assay kit provided by R & D Systems (LIP-1-NCL1, Minneapolis, MN), and the assay protocol followed the instruction suggested by the manufacturers. To see the possible involvement of tumor necrosis factor α (TNF-α) in enhanced lipolysis in cocultures, we used anti–TNF-α neutralizing antibody (goat polyclonal anti-mouse TNF-α neutralizing antibody, R & D Systems) and performed the same experiments in the presence of neutralizing antibodies or the same concentration of nonspecific control goat immunoglobulin G (IgG) (R & D). Significance of the difference of glycerol release among each culture condition was determined by Student *t* test.

2. Results

We compared the associations between alveolar bone score with other medical parameters. First, bone score significantly associated with donor age ($r = 0.169$, $P < .032$, Spearman correlation coefficient). Bone score also associated with TG levels ($r = 0.299$, $P = .000$) (Fig. 1A). This association became more evident when we narrowed the

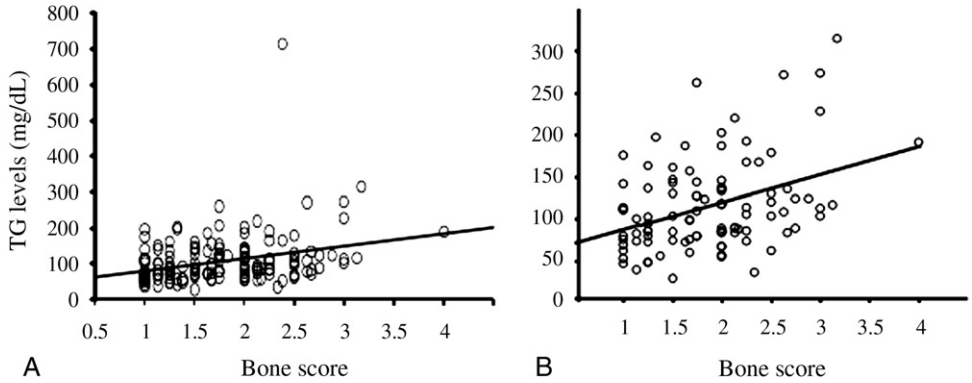

Fig. 1. Association between serum TG level and bone score in all subjects (A) and in subjects with midrange BMI (21.5-27.0). A positive correlation between bone score and TG levels (A) ($r = 0.299$, $P = .000$; Spearman correlation coefficient) was observed. This association became more evident when we narrowed the range of BMI to midrange (B) (21.5-27.0 kg/m^2) ($r = 0.332$, $P < .001$).

range of BMI to midrange (21.5-27.0 kg/m^2) (n = 99; 43 men and 56 women aged 44-74 years; mean age, 67.41 ± 4.93 years; $r = 0.332$; $P < .001$) (Fig. 1B). Triglyceride levels were associated with waste circumstances and BMI when the analysis was done for all participants ($r = 0.244$, $P < .02$ for waste circumstances; $r = 0.176$, $P < .025$ for BMI) (Fig. 2A, B). However, in subjects with midrange BMI, no significant association was observed ($r = 0.069$, $P = .384$ for waste circumstances; $r = -0.120$, $P = .238$ for BMI) (Fig. 2C, D). Although there was a trend between TG level and age, the association was not statistically significant ($r = 0.154$, $P = .050$). Furthermore, in midrange BMI group, the association between TG levels and age completely disappeared ($r = 0.145$, $P = .153$). We then saw the association between alveolar bone score and waste circumstances as well as BMI. However, no associations were found in these parameters ($r =$

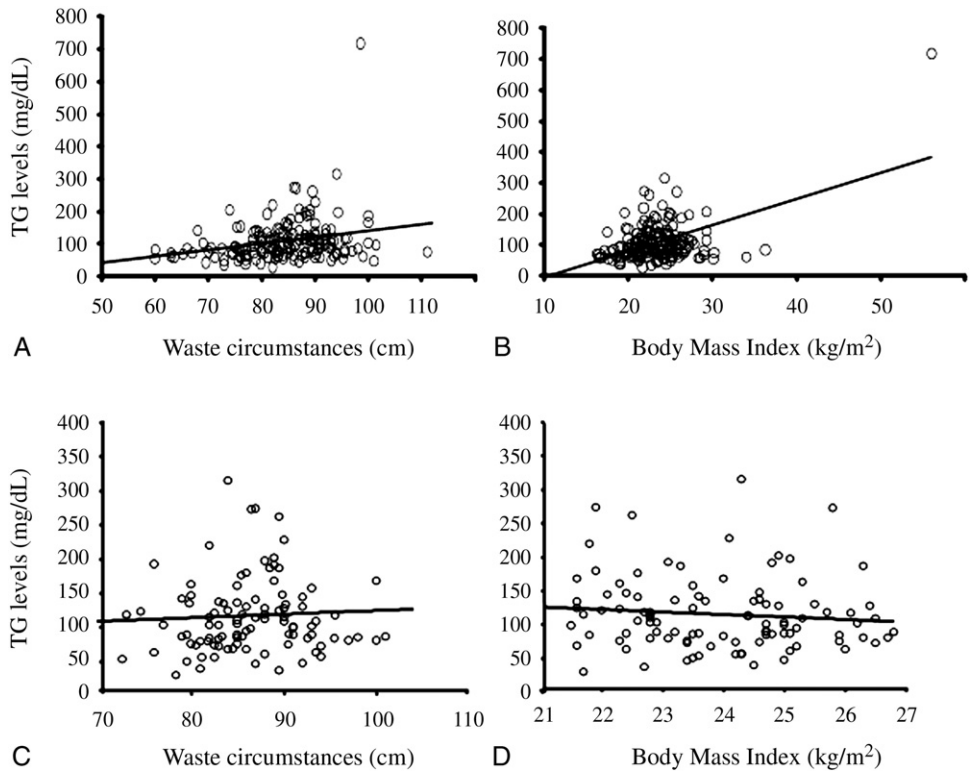

Fig. 2. Association between serum TG level and waste circumstances (A) and BMI (B) in all subjects and the association between TG level and waste circumstances (C) and BMI (D) in subjects with midrange BMI. The TG levels were associated with waste circumstances and BMI when the analysis was done for all participants ($r = 0.244$, $P < .02$ for waste circumstances [A]; $r = 0.176$, $P < .025$ for BMI [B]). However, in subjects with midrange BMI, no significant association was observed ($r = 0.069$, $P = .384$ for waste circumstances [C]; $r = -0.120$, $P = .238$ for BMI [D]).

0.134, $P = .090$ between waste circumstances and bone score; $r = 0.087$, $P = .272$ between bone score and BMI) (Fig. 3A, B). Similarly, no associations were found in these parameters in midrange BMI group as well ($r = 0.026$, $P = .795$ between waste circumstances and bone score; $r = -0.020$, $P = .846$ between bone score and BMI) (Fig. 3C, D). In addition, original association between bone score and age also disappeared in midrange BMI group ($r = 0.152$, $P = .133$). In midrange BMI group, therefore, the association between TG levels and bone score was only left as statistically significant. Thus, it appeared that periodontal disease associated with serum TG levels independent of the degree of obesity and age. Besides TG, none of the other serum parameters statistically associated with bone score as far as examined.

We then performed an in vitro study to see the effects of bacterial endotoxin on lipolytic activity. The results are shown is Fig. 4. As demonstrated, low-dose LPS dose-dependently enhanced lipolysis in cocultures of adipocytes and macrophages 24 hours following LPS stimulation (Fig. 4A). Because it has been suggested that TNF-α was a potent inducer of lipolysis in adipocytes, we speculated that macrophage-derived TNF-α could be a direct effector molecule for LPS-induced enhanced lipolysis. Therefore, we performed inhibition assay. As expected, neutralizing TNF-α action by specific antibodies suppressed enhanced lipolysis in cocultures compared with the cells treated with control IgG with LPS stimulation (Fig. 4B).

3. Discussion

In this study, we first confirmed an association between cumulative periodontal bone destruction and serum TG levels in Japanese subjects, especially with midrange BMI (21.5-27.0). As periodontal disease indices, we measured bone score by oral panoramic radiograph. The reasons why we chose panoramic radiograph are as follows: first, it is simpler and is a relatively sensitive method to assess the cumulative periodontal tissue destruction; and second, the recent Editors' Consensus position paper by the *American Journal of Cardiology* and the *Journal of Periodontology* suggested that one of the reasons for the large discrepancies seen in the previous results of the association between periodontitis and atherosclerosis might be due to differing measures and definitions of periodontitis, with some studies based only on clinical measures (ie, pocket depth, bleeding on probing, tooth attachment levels) and others, in which the relation appeared stronger, based on nonclinical measures such as systemic antibody response or radiographic evidence of alveolar bone loss [10]. Of course, there are also several limitations in assessing periodontal bone loss by oral panoramic radiograph as suggested by others before [11-13]. Original Shei measurement was described with intraoral periapical radiograph, but not with panoramic radiograph. However, 2 previous articles suggested that panoramic assessment may substitute for full-

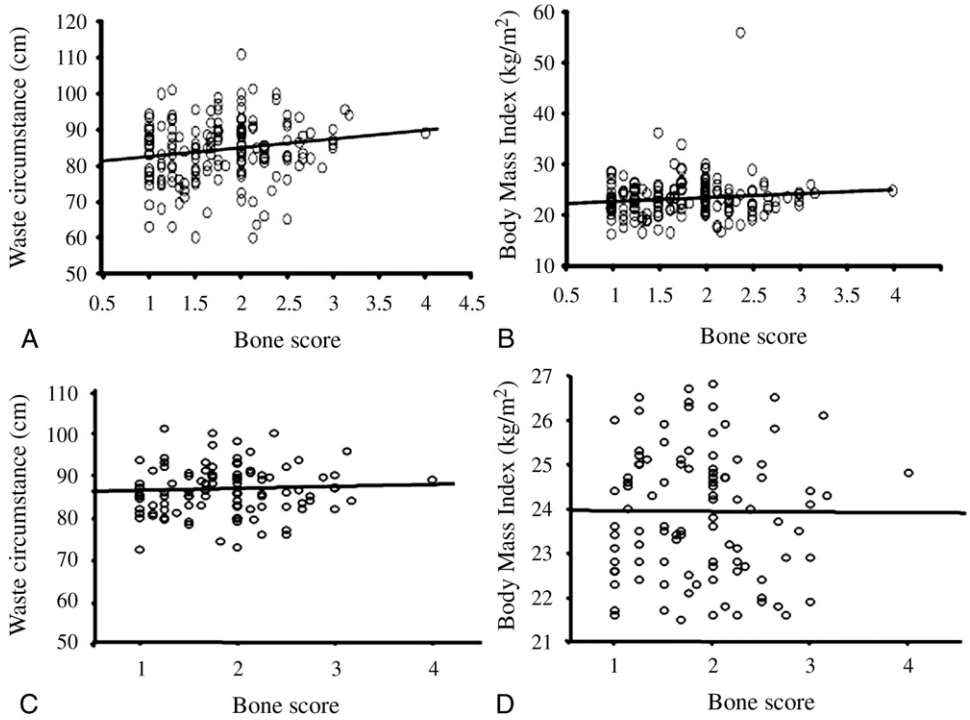

Fig. 3. Association between periodontal bone score and waste circumstances (A) and BMI (B) in all subjects and the association between bone score and waste circumstances (C) and BMI (D) in subjects with midrange BMI. Although a trend between the degree of obesity and periodontal bone score was observed, no statistically significant associations were found between waste circumstances and bone score (A) ($r = 0.134$, $P = .090$) or between BMI and bone score (B) ($r = 0.087$, $P = .272$). Moreover, when we narrowed the population to those with midrange BMI, absolutely no associations were found in these parameters as well ($r = 0.026$, $P = .795$ between waste circumstances and bone score [C]; $r = -0.020$, $P = .846$ between bone score and BMI [D]).

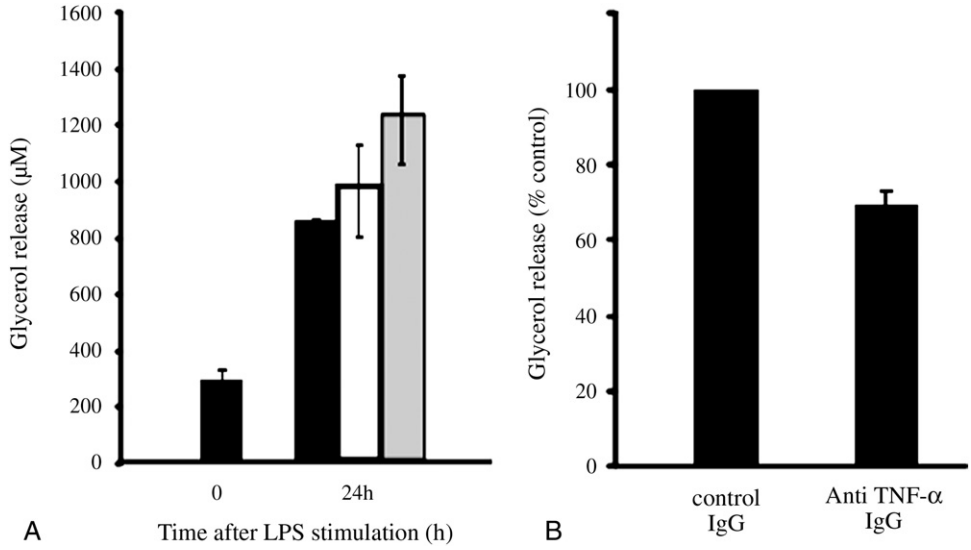

Fig. 4. The effects of bacterial endotoxin on lipolysis in cocultures of adipocytes and macrophages (A) and its inhibition by anti–TNF-α neutralizing antibodies (B). Lipolysis was evaluated by the amount of released glycerol after LPS stimulation as described in the "Subjects and methods" section. Lipopolysaccharide dose-dependently up-regulated lipolytic activity in cocultures (A). Black box: LPS (−); open box: LPS (1 ng/mL); gray box: LPS (10 ng/mL). Enhanced lipolytic activity was inhibited by the addition of anti–TNF-α neutralizing antibodies in to cultures (B). Data are expressed as percentage inhibition of glycerol release treated by control IgG.

mouth periapical radiographic evaluation, although the sensitivity might be slightly decreased [11,12]. In this study, we used simplified the JDCP method, which is much easier to calculate. Therefore, we speculated that the agreement rate would further increase by the use of simplified method rather than conventional Schei score calculated from oral panoramic radiograph. Another study pointed out that panoramic radiograph might be unsuitable around the mandibular anterior teeth because of superimposition of radio-opaque structures, reduced image detail compared with intraoral views, and uneven magnification [13]. To avoid this problem, we selected simplified JDCP methods, as we expected to overcome the problems of less sharp image around the anterior region. Although even the simplified method may not be enough to completely overcome this problem, we wished to include anterior regions for the analyses because it is well known that this area is very susceptible to periodontal tissue destruction and there is no other currently available simple way to assess the degree of bone loss in a large population.

Of particular note is that, although TG level was associated with waste circumstances as well as BMI, no association was observed between periodontal bone score and waste circumstances, or BMI in our Japanese subjects. This trend was more evident in subjects with midrange BMI. This is a surprising finding because it is believed that obesity itself was strongly associated with developing periodontal disease [14,15]. The reason why we could not find any association between periodontal bone score and the degree of overweight is not known at present. One reason is the different clinical characteristics studied. Our subjects were Japanese, and most of them had midrange BMI. Our team has previously shown that although the degree of overweight affects insulin resistance, the factors underlying insulin resistance differ in Japanese subjects with midrange BMI [16]. In midrange BMI groups, serum TG level but not BMI was mostly associated with insulin resistance. It may be argued that visceral fat area per se affects periodontal disease in these subjects because serum TG was reported to be associated with visceral fat area in humans [16]. However, we did not measure visceral fat area in the present study. An association of serum TG with visceral fat area was reported in Japanese subjects with BMI less than 26.5 kg/m^2 [17]. We previously demonstrated that serum TG was associated with visceral fat area but not with subcutaneous fat area in Japanese type 2 diabetes mellitus patients with midrange BMI ($21.5 < \text{BMI} < 27.0$) [16]. The prevalence of periodontal disease is known to be higher in type 2 diabetes mellitus patients. Furthermore, TG is synthesized in the liver following lipolysis in visceral fat [16]. Free fatty acids, by-products of lipolysis, have been suggested to enter the liver via portal vein to be used to synthesize TG. In fact, in our current study, most of the subjects were in midrange BMI. The number of the subjects with BMI greater than 30 kg/m^2 was only 5, and it corresponded to 3% of all participants. This ratio was also the same frequency of extremely obese subjects (BMI >30 kg/m^2) seen in entire Japan. Therefore, we speculate that the effects of periodontal disease on high TG levels will be diminished in a high-BMI population, as TG synthesis becomes more dependent on the degree of obesity than low-grade infectious disease such as periodontal disease as BMI increases. In fact, the association between TG and bone score was stronger in subjects with midrange BMI.

Interestingly, in these subjects, TG did not correlate with waste circumstances or BMI. As described, previous studies including ours had suggested the mild elevation of hsCRP in association with periodontal infection or inflammation [4]. This association was also more evident in midrange BMI population or less obese US population [4,18]. Especially in Japanese type 2 diabetes mellitus subjects whose BMI was greater than 27 kg/m^2, the association between hsCRP and periodontal infection as assessed by systemic antibody responses appeared to be completely masked by the influences of obesity [4]. Statistically significant association was only seen in subjects whose BMI was less than 27 kg/m^2. C-reactive protein is believed to be synthesized in the liver in response to an inflammatory cytokine, interleukin-6 (IL-6) [19]. In this context, we previously reported that IL-6 production was markedly enhanced when adipocytes were cocultured with macrophages in the presence of endotoxin, and suggested that this might be an underlying important mechanism up-regulating CRP production in association with periodontal disease [9]. We also demonstrated that macrophage-derived TNF-α played an important role in enhanced IL-6 production, as neutralizing TNF-α activity by specific antibody suppressed enhanced IL-6 production [9]. We further suggested that periodontal inflammation suppressed insulin action in type 2 diabetes mellitus subjects possibly because of the enhanced TNF-α production in adipose tissues or in the liver [20]. In fact, in Japanese type 2 diabetes mellitus subjects, CRP has been suggested to well correlate with the degree of insulin resistance, suggesting that IL-6 and upstream TNF-α play important roles in enhanced CRP production [21]. Therefore, we hypothesized that such conditions act to provide an environment leading to enhanced TG synthesis as well based on the current clinical observations. For this reason, we used adipocyte-macrophage coculture system and stimulated the cocultures with bacterial endotoxin because macrophages that have infiltrated into adipose tissues have been suggested to play important roles in inducing inflammatory changes of adipose tissues [22,23] and because toll-like receptor 4, a receptor for bacterial endotoxin, has been suggested to play a crucial role in amplifying inflammatory responses in adipose tissues infiltrated with macrophages [24]. The result indicated that low-dose LPS up-regulated TG synthesis and that TNF-α derived from macrophages appeared to play an important role in enhanced TG synthesis in cocultures, as neutralizing TNF-α action by specific antibodies suppressed lipolysis. In fact, TNF-α has previously been suggested to enhance lipolysis in single culture of adipocytes [25]. All these phenomenon well account for the observed adverse effects of periodontitis on metabolic disorders such as enhanced CRP production, increased insulin resistance, and hypertriglyceridemia as demonstrated in our current study. In fact, periodontal disease has previously been suggested to be associated with increased lipid deposition on the surface of aorta in rabbit model [26].

In conclusion, we believe that increased TG in association with enhanced lipolysis by infection might be one of the mechanisms by which periodontal disease promotes atherosclerotic changes; and we suggest that visceral adipose tissues, even in nonobese subjects, play a crucial role in mediating this adverse effect.

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References

[1] Taylor-Robinson D, Thomas BJ. *Chlamydia pneumoniae* in atherosclerotic tissue. J Infect Dis 2000;181:S437-40.
[2] Moutsopoulos NM, Madianos PN. Low-grade inflammation in chronic infectious disease: paradigm of periodontal infections. Ann N Y Acad Sci 2006;1088:251-64.
[3] Johnson SC, Zhang H, Messina LM, et al. *Chlamydia pneumoniae* burden in carotid arteries is associated with upregulation of plaque interleukin-6 and elevated C-reactive protein in serum. Arterioscler Thromb Vasc Biol 2005;25:2648-53.
[4] Nishimura F, Taniguchi A, Iwamoto Y, et al. *Porphyromonas gingivalis* infection is associated with elevated C-reactive protein in nonobese Japanese type 2 diabetic subjects. Diabetes Care 2002;25:1888.
[5] Ridker PM, Cushman M, Stampfer MJ, et al. Inflammation, aspirin, and the risk of cardiovascular disease in apparently healthy men. N Engl J Med 1997;336:973-9.
[6] Arima H, Kubo M, Yonemoto K, et al. High-sensitivity C-reactive protein and coronary heart disease in a general population of Japanese: the Hisayama study. Arterioscler Thromb Vasc Biol 2008;28:1385-91.
[7] de Maat MP, Trion A. C-reactive protein as risk factor versus risk marker. Curr Opin Lipidol 2004;15:651-7.
[8] Adams RA, Nystrom GP. A periodontal severity index. J Periodontol 1986;57:176-9.
[9] Yamashita A, Soga Y, Iwamoto Y, et al. Macrophage-adipocyte interaction: marked IL-6 production by co-cultures stimulated with LPS. Obesity 2007;15:2549-52.
[10] Friedewald VE, Kornman KS, Beck JD, et al. The *American Journal of Cardiology* and *Journal of Periodontology* Editors' Consensus: periodontitis and atherosclerotic cardiovascular disease. Am J Cardiol 2009;104:59-68.
[11] Persson RE, Tzannetou S, Feloutzis AG, et al. Comparison between panoramic and intra-oral radiographs for the assessment of alveolar bone levels in a periodontal maintenance population. J Clin Periodontol 2003;30:833-9.
[12] Molander B, Ahlqwist M, Grondahl HG, Hollender L. Agreement between panoramic and intra-oral radiography in the assessment of marginal bone height. Dentomaxillofac Radiol 1991;20:155-60.
[13] Walker C, Thompson D, KcKenna G. Case study: limitations of panoramic radiography in the anterior mandibule. Dent Update 2009;36:620-3.
[14] Saito T, Shimazaki Y, Sakamoto M. Obesity and periodontitis. N Engl J Med 1998;339:482-3.
[15] Wood N, Johnson RB, Streckfus CF. Comparison of body composition and periodontal disease using nutritional assessment techniques: Third

National Health and Nutrition Examination Survey (NHANES III). J Clin Periodontol 2003;30:321-7.

[16] Taniguchi A, Fukushima M, Sakai M, et al. The role of the body mass index and triglyceride levels in identifying insulin-sensitive and insulin-resistant variants in Japanese non–insulin-dependent diabetic patients. Metabolism 2000;49:1001-5.

[17] Kobayashi H, Nakamura T, Miyaoka K, et al. Visceral fat accumulation contributes to insulin resistance, small-sized low-density lipoprotein, and progression of coronary artery disease in middle-aged non-obese Japanese men. Jpn Circ J 2001;65:193-9.

[18] Slade GD, Ghezzi EM, Heiss G, et al. Relationship between periodontal disease and C-reactive protein among adults in the Atherosclerosis Risk in Communities study. Arch Intern Med 2003; 163:1172-9.

[19] Gabay C, Kushner I. Acute-phase proteins and other systemic responses to inflammation. N Engl J Med 1999;340:448-54.

[20] Nishimura F, Iwamoto Y, Soga Y. Periodontal host response with diabetes. Periodontology 2007;2000:245-53.

[21] Taniguchi A, Nagasaka S, Fukushima M, et al. C-reactive protein and insulin resistance in non-obese Japanese type 2 diabetic patients. Metabolism 2002;51:1578-81.

[22] Xu H, Barnes GT, Yang Q, et al. Chronic inflammation in fat plays a crucial role in the development of obesity-related insulin resistance. J Clin Invest 2003;112:1821-30.

[23] Weisberg SP, McCann D, Desai M, et al. Obesity is associated with macrophage accumulation in adipose tissue. J Clin Invest 2003;112: 1796-808.

[24] Shi H, Kokoeva MV, Inouye K, et al. TLR4 links innate immunity and fatty acid–induced insulin resistance. J Clin Invest 2006;116: 3015-25.

[25] Kawakami M, Murase T, Ogawa H, et al. Human recombinant TNF suppresses lipoprotein lipase activity and stimulates lipolysis in 3T3-L1 cells. J Biochem 1987;101:331-8.

[26] Jain A, Batista Jr EL, Serhan C, et al. Role for periodontitis in the progression of lipid deposition in an animal model. Infect Immun 2003;71:6012-8.