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SHORT COMMUNICATION

Oxidized-low density lipoprotein in gingival crevicular fluid of patients with chronic periodontitis: a possible link to atherogenesis

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Abstract

Objective. To investigate a possible link between periodontitis and atherogenesis by examining the levels of anti-oxidized low density lipoprotien (ox LDL) and low density lipoprotien (LDL) in gingival crevicular fluid (GCF) and serum of healthy subjects and chronic periodontitis patients. ***Methods.*** Sixty male subjects (35–55 years) were grouped into 30 healthy individuals and 30 subjects with chronic periodontitis. Serum and GCF samples were obtained from each subject and were assessed for anti-ox LDL and LDL levels. ***Results.*** A significant difference ($p < 0.001$) was found between the anti-ox-LDL levels in GCF of healthy vs chronic periodontitis groups. Also the ratio of GCF anti-ox LDL to GCF LDL was significantly higher ($p < 0.001$) in chronic periodontitis patients as compared to the healthy group. ***Conclusions.*** A significant rise in ox LDL level in otherwise systemically healthy chronic periodontitis patients may put these subjects at an increased risk of developing atherosclerosis.

Key Words: *LDL, oxidized LDL, periodontitis, serum*

Introduction

Periodontal and atheromatous diseases share many risk factors including age, gender, lower socio-economic status, stress, smoking and pathogenic pathways [1]. Evidence suggesting an association between periodontitis and atheromatous diseases is mounting [2].

OxLDL is a mixture of heterogeneously modified particles. The proposition that lipid oxidation plays a central role in atherogenesis is well documented and accepted [3]. Schenkein et al. [4] demonstrated that anti-ox-LDL antibodies are increased in GCF in patients with aggressive periodontitis and they may be responsible for mediating immune reactions of relevance to cardiovascular diseases. A recent pilot study by Sakiyama et al. [5] was done to establish the presence of ox-LDL in GCF of systemically and periodontally healthy patients. We hypothesized that the ox-LDL produced in the gingival tissues as a result of periodontal disease might affect the serum ox-LDL levels in otherwise systemically healthy subjects and might have a role in association between chronic periodontitis and atherogenesis. Therefore, the aim of our study was to investigate whether chronic periodontitis could affect levels of ox LDL in GCF and serum of otherwise healthy subjects.

Materials and methods

Sixty male subjects within the age group of 35–55 years having at least 24 teeth were included. The study protocol conformed to the Declaration of Helsinki and was approved by the institutional ethical committee. Written informed consent was obtained from each participating subject. None of the subjects had received antibiotics or undergone periodontal treatment within the past 6 months. Also, subjects having systemic diseases and smokers were excluded from the study. All the participants were categorized into two groups: Group 1, healthy control subjects ($n = 30$ mean age 45 ± 8.9 years) consisted of periodontally healthy and normotensive subjects and Group 2, systemically healthy chronic periodontitis subjects ($n = 30$ mean age 47 ± 11.2 years) consisted of those diagnosed with chronic generalized periodontitis and normotensive based on criteria proposed by the 1999 International Workshop for a Classification

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Table I. Mean and standard deviation of GCF ox LDL, GCF LDL (low density lipoprotein), serum ox-LDL and serum LDL.

Variable	Group 1 (n = 30)	Group 2 (n = 30)	p-value
GCF ox-LDL	20.2 ± 2.8	37.4 ± 4.3	< 0.001
GCF LDL	10.5 ± 1.1	13.1 ± 1.5	< 0.001
Serum ox-LDL	27.6 ± 2.4	46.3 ± 4.7	< 0.001
Serum LDL	125.6 ± 8.0	136.4 ± 5.4	< 0.001
GCF oxLDL to GCF LDL ratio	1.92 ± 0.26	2.88 ± 0.37	< 0.001

of Periodontal Diseases and Conditions [6] and JNC recommendation [7].

Five millilitre venous blood samples were obtained from each subject between 9:00 am and 11:00 am, which were processed for serum and then stored at 20°C until further processing. GCF samples were collected from a randomly selected tooth in the healthy controls and the deepest site in the dentition for the periodontitis group as described by Sakiyama et al. [5] and stored at 80°C until further analysis.

ELISA (Immulisa, IMMCO Diagnostics, USA; New Delhi, India) was used in the determination of serum and GCF concentrations of ox-LDL [5]. LDL determination in the serum and GCF samples were done using a commercial kit and was analysed by routine spectrophotometry. Since the volume of GCF collectable from each tooth varies, the ratio of anti-ox LDL/LDL was also calculated to evaluate the anti-ox LDL levels of GCF samples [5].

Results

Mean GCF and serum anti ox-LDL levels in the periodontitis group were observed to be significantly elevated ($p < 0.001$) as compared to the healthy control group. Serum and GCF levels of LDL were observed to be significantly lower ($p < 0.001$) in the healthy control group as compared to the periodontitis group. The ratio of GCF oxLDL to GCF LDL was found to be significantly higher ($p < 0.001$) in the periodontitis group as compared to the healthy control group (Table I).

There was a strong positive correlation between serum anti-ox LDL and GCF anti-ox LDL in the periodontitis group ($p < 0.001$) (Table II). Based on our observations, a schematic diagram showing the possible interactions of locally produced factors in diseased periodontal tissues and circulating ox-LDL and LDL have been depicted (Figure 1).

Table II. Correlation between serum and GCF ox LDL and LDL.

	Group 1		Group 2	
Correlation between	r-value	p-value	r-value	p-value
Serum ox LDL and GCF ox LDL	+0.26	0.16	+0.89	< 0.001
Serum LDL and GCF LDL	+0.36	0.05	+0.45	0.01

Discussion

Ox-LDL is thought of being an integral in the final pathway of the many risk factors leading to foam cell formation and raised ox-LDL levels in serum are considered to be risk indicators of various cardiovascular diseases [8]. Although a direct causal relationship between periodontitis and atherosclerotic CVD has not been established, the association between atherogenesis and periodontitis is well documented. Our results demonstrated that mean GCF anti ox-LDL levels were significantly higher in periodontitis as compared to the healthy control group. This can be explained by the fact that the environment in a periodontal pocket is highly oxidative and has many possible agents which are known oxidizers of LDL [9] (reactive oxygen species (ROS) [10], myeloperoxidase (MPO) [11], phospholipase A2 [12] and nitric oxide (NO) [13]). One or multiple of these pathways may be responsible for conversion of the circulating LDL to ox-LDL locally in the periodontal tissues.

Serum levels of anti-ox-LDL levels were significantly higher in the periodontitis group as compared to the healthy control group. Also, the ratio of anti-ox-LDL to LDL in GCF is highly increased in subjects with chronic periodontitis. This suggests that chronic periodontitis may have an effect on serum anti-ox-LDL levels. This increased ox-LDL may be attributed to the presence of periodontal inflammation. It is known that elevated serum levels of anti-ox-LDL are related to thrombosis, stroke, myocardial infarction and atherosclerosis [14]. A recent study by Tamaki et al. [15] reported that improved oral hygiene and non-surgical periodontal treatment were effective in decreasing serum ox LDL level. This indicates that, with control of periodontal inflammation, the systemic circulating oxidative stress can be reduced. A recent review by Itabe [16] stresses the role ox-LDL may play as a biomarker of oxidative stress in periodontitis. In summary, our results suggest that the serum and GCF levels of LDL and ox-LDL are increased in patients with chronic periodontitis as compared to healthy controls. Such a profile puts an otherwise healthy chronic periodontitis patient at a higher risk of atherogenesis-related

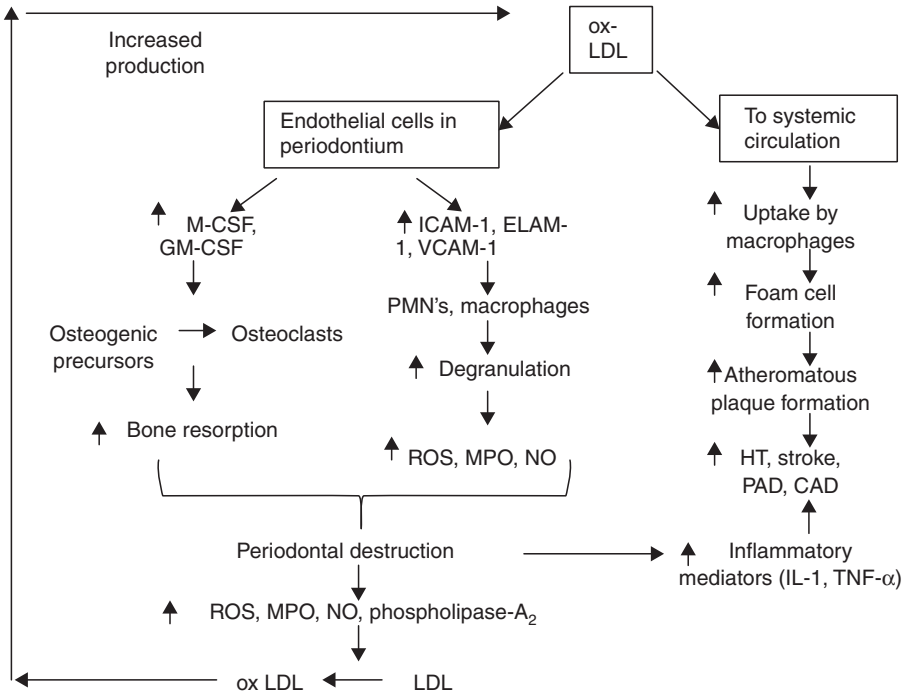

Figure 1. Possible mechanisms of production of ox-LDL in periodontal tissues and its effect on systemic atherosclerosis. ox-LDL, oxidized low density lipoprotein; M-CSF, Macrophage colony stimulating factor; GM-CSF, Granulocyte macrophage colony stimulating factor; ICAM-1, intercellular adhesion molecule-1; ELAM-1, Endothelium leukocyte adhesion molecule-1; VCAM-1, Vascular cell adhesion molecule-1; PMN, Polymorphonuclear neutrophils; ROS, Reactive oxygen species; MPO, Myeloperoxide; NO, Nitric oxide; HT, Hypertension; PAD, Peripheral artery disease; CAD, Coronary artery disease; IL-1, Interluekin–1; TNF-α, Tumour necrosis factor-α.

diseases. Although the exact mechanism of how systemic ox-LDL increases in chronic periodontitis patients is not known, our study provides evidence for correlation between chronic periodontitis and increased systemic levels of ox-LDL.

Declaration of interest: The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

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