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Original Research

Assessment of Biomarkers of Coronary Heart Disease in Patients with Periodontitis

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Abstract

Introduction: Gingival inflammation is common in patients on anticonvulsant drugs and in bacterial and viral infections of the oral cavity. The prevalence of coronary heart disease (CHD) is reported to be higher in patients with periodontitis. The elevation in inflammatory markers and oxidative stress are the major causes predisposing an individual to CHD. No attempts were made earlier to evaluate the role of dyslipidemia and oxidative stress in patients with periodontal inflammation. Paraoxonase 1 (PON1) prevents the formation of lipid hydroperoxides and oxidized phospholipids by hydrolyzing them once they are formed, and its level is rarely studied in the disease of the oral cavity. Hence in the present study, an attempt is being made to evaluate the impact of drug-induced and bacterial gingivitis on lipid profile and oxidative stress which predisposes an individual to CHD.

Materials and Methods: Lipid profile, malondialdehyde, glutathione, and PON1 were studied in serum of the 32 patients with drug-induced gingival enlargement, 12 patients with gingival inflammation due to *Porphyromonas gingivalis* infection and seven with *P. gingivalis* infection and nifedipine-induced gingival inflammation. The results were compared with sixteen age and sex matched controls using the one-way ANOVA test, and Pearson correlation.

Results: Total cholesterol, triglycerides and low-density lipoprotein cholesterol were found to be elevated whereas high-density lipoprotein cholesterol and the PON1 was significantly decreased in the patients compared to controls. The gingival index was correlating well with all the parameters of lipid profile and PON1.

Conclusion: The patients of periodontal disease are at a high risk of developing CHD compared to subjects with healthy periodontal tissues. Reducing the inflammation by replacing the drug, eliminating the bacterial infection, and by providing antioxidant-rich diet are suggested as methods for reducing the risk of CHD in patients with periodontitis.

Key Words: Gingivitis, gingival index, glutathione, malondialdehyde, nifedipine, paraoxonase 1, periodontitis, *Porphyromonas gingivalis*

Introduction

The gingival inflammation is common in patients on anticonvulsant drugs and in bacterial and viral infections of the oral cavity. The drugs causing gingival inflammation mainly belongs to three major categories namely calcium channel blockers such as nifedipine, anti-convulsants drugs, and immunosuppressants.[1-3]

Among numerous bacterial and viral oral infections, infection caused by *P. gingivalis* is the major cause of periodontal inflammation.[4] Evidence relating the pathogenic role of these bacteria is available in the literature however the molecular and physiological mechanism remains unexplained.[5]

The prevalence of coronary heart disease (CHD) is reported to be higher in patients with periodontitis.[6] Reports on the role of the inflammatory markers, which predisposes an individual to CHD, are scanty. We have earlier reported the role of inflammatory markers, C-reactive protein, and fibrinogen, in the causation of the inflammation in periodontitis either due to drugs or infection.[7] The combined effect of the bacterial infection and nifedipine was also stressed.

The dyslipidemia is reported to be one of the major risk factors of CHD.[8] The elevation in inflammatory markers and oxidative stress along with inflammation are the major causes predisposing an individual to CHD.[9] No attempts were made earlier to evaluate the role of dyslipidemia and oxidative stress in patients with periodontal inflammation.

High-density lipoprotein (HDL) including its major apolipoprotein A-1 along with at least four enzymes, including paraoxonase 1 (PON1) and lecithin: Cholesterol acyltransferase contributes to the antioxidant effect.[10] Studies demonstrated that PON1 prevents the formation of lipid hydroperoxides and oxidized phospholipids by hydrolyzing them once they are formed.[11] Similar studies were rare in disease of the oral cavity.

Hence in the present study, an attempt is being made to evaluate the impact of drug-induced and bacterial gingivitis

on lipid profile and oxidative stress which predisposes an individual to CHD.

Materials and Methods

The 32 patients with gingival enlargement due to nifedipine and twelve patients with gingival inflammation due to *P. gingivalis* infection and seven with *P. gingivalis* infection and nifedipine-induced gingival inflammation were identified by the physicians and referred to the dental clinic formed the test group. The diagnosis was confirmed by the dentist based on clinical assessment by comprehensive periodontal examination. The detailed examination consists of a visual examination, radiographs, and probing of the gingiva to measure the extent of gingival damage and by laboratory investigation. *P. gingivalis* was identified by anerobic culture technique. The gingival status was assessed based on tools of gingival index (GI) of Loe and Silness.

Five ml of venous blood was collected from all the subjects after getting their informed consent. Lipid profile, triglycerides total cholesterol, HDL cholesterol, and low-density lipoprotein (LDL) cholesterol were estimated in the fully automated analyzer (AU480, Beckman). PON1, oxidative stress, antioxidant status was assessed by measuring malondialdehyde (MDA) and glutathione (GSH) by spectrophotometric method.[12-14]

The values were compared with 16 age and sex matched controls. The comparison is done using the one-way ANOVA test, and Pearson correlation is done using the SPSS 17 software and the Minitab 15 software. A $P \leq 0.05$ was considered as statistically significant.

Result

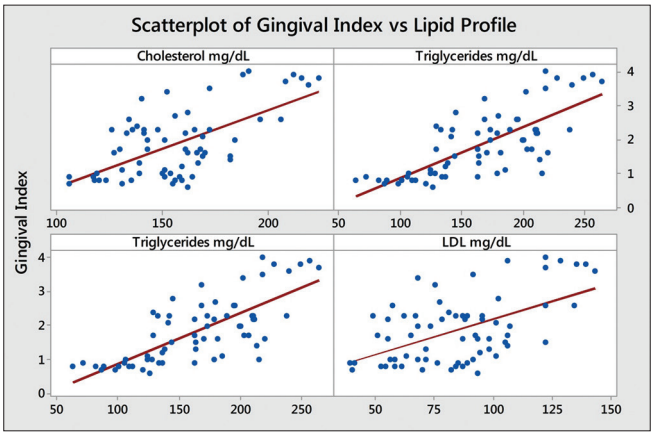
The results of the serum lipid profile and PON1 of the test and control subjects are given in Table 1. Except HDL cholesterol and PON1 all other parameters were found to be elevated in the test groups whereas HDL cholesterol and PON1 were significantly decreased in the test group. The statistical correlation of these values with GI is given in Table 2 and Graph 1. All these parameters were significant in the patient group compared to normal healthy subjects. One-way ANOVA of lipid profile and PON1 among the categories are given in Table 3. Here also the above parameters were significant in all the test group subjects.

Among all the parameters studied PON1 showed the maximum correlation ($r = -0.781$), with GI, followed closely by triglycerides ($r = 0.750$). The one-way ANOVA also showed a similar trend where the F value for PON1 and triglycerides were highly significant ($P < 0.001$). Even though the other parameters namely total cholesterol, LDL cholesterol and HDL cholesterol were significantly altered, the extent of alteration was lesser that of triglycerides and PON1.

The oxidative stress of the subjects was evaluated by measuring MDA, and antioxidant status was evaluated by measuring GSH. The results are given in Table 4 and Graph 2. There was a significant increase in MDA ($r = 0.510$) and decrease in GSH ($r = -0.500$) in patients compared to control subjects suggesting increased oxidative stress and decreased antioxidant status in patients with periodontitis.

Discussion

We observed elevated levels of LDL cholesterol, total cholesterol, and triglycerides in patients with periodontitis irrespective of the cause. This is well in agreement with the previous reports.[15,16] In the present study, PON1 and the HDL cholesterol was significantly lower than that of the control. Decreased HDL cholesterol in periodontal disease is



Graph 1: Matrix plot of gingival index versus lipid profile. HDL: High-density lipoprotein cholesterol, LDL: Low-density lipoprotein cholesterol. All lipid profile parameter and the paraoxonase 1 are statistically correlating well with the gingival index.

Table 1: Comparison of lipid profile and PON1 in control and test groups.

Parameters	Category			
	Control	Drug	Bacterial	Both
Cholesterol (mg/dL)	138.63±21.08	156.52±19.82	157.67±25.46	197.29±30.7
Triglycerides (mg/dL)	109±23.92	176.26±30.31	181.5±39.77	205.5±71.63
HDL (mg/dL)	47.94±5.34	35.16±5.3	37.67±5.25	35.57±11.86
LDL (mg/dL)	68.63±19.33	86.03±21.24	83.67±22.86	118.29±28.22
PON1 (nmol/ml/min)	75.21±6.62	57.58±3.19	54.92±5.67	45.49±12.73

All values are mean±standard deviation, HDL: High-density lipoprotein cholesterol, LDL: Low-density lipoprotein cholesterol, PON1: Paraoxonase 1

Table 2: Pearson correlation statistic of GI against the lipid profile and PON1.

Parameters	GI	
	***r* value**	***P* value**
Cholesterol (mg/dL)	0.63	<0.001
Triglycerides (mg/dL)	0.75	<0.001
HDL (mg/dL)	−0.512	<0.001
LDL (mg/dL)	0.546	<0.001
PON1 (nmol/ml/min)	−0.781	<0.001

r value: Pearson correlation coefficient, HDL: High-density lipoprotein cholesterol, LDL: Low-density lipoprotein cholesterol, PON1: Paraoxonase 1

Table 3: One-way ANOVA of lipid profile and PON1 among the categories.

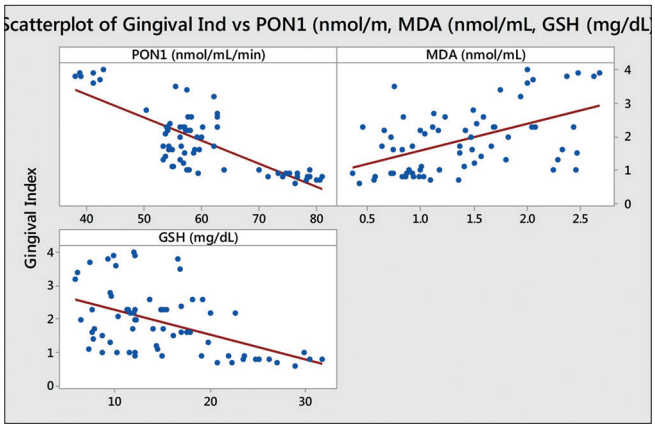
Parameters	*F*	Significance
Cholesterol (mg/dL)	2.903	0.001
Triglycerides (mg/dL)	5.037	<0.001
HDL (mg/dL)	3.129	0.001
LDL (mg/dL)	2.200	0.012
PON1 (nmol/ml/min)	13.741	<0.001

F: *F*-statistic of one-way ANOVA, HDL: High-density lipoprotein cholesterol, LDL: Low-density lipoprotein cholesterol, PON1: Paraoxonase 1, GI: Gingival index

Table 4. Pearson correlation statistic of GI against anti-oxidant status and oxidative stress.

Parameters	GI	
	***r* value**	***P* value**
MDA (nmol/mL)	0.510	<0.001
GSH (mg/dL)	−0.500	<0.001

r value: Pearson correlation coefficient, MDA: Malondialdehyde, GSH: Glutathione, GI: Gingival index



Graph 2: Matrix plot of gingival index versus paraoxonase 1 (PON1), malondialdehyde (MDA) and glutathione (GSH). HDL: High-density lipoprotein cholesterol, LDL: Low-density lipoprotein cholesterol, PON1, MDA and GSH are statistically correlating well with the gingival index.

reported earlier by O'Neill *et al.*[17] Increase in triglycerides, total cholesterol and LDL cholesterol are considered as high-risk markers of CHD.[16] Our results also indicate that patients with periodontitis may be at high risk of developing CHD. This fact is also supported by low levels os HDL cholesterol in the test groups compared to the controls.

We also observed decreased levels of PON1 in the patient groups irrespective of the cause of the periodontitis. PON1, is reported to be one of the most important enzymes which is having a potent antioxidant effect. The correlation of PON1 with CHD are recently reported.[18,19] However, studies correlating PON1 with periodontal disease is rare and scanty. There were recent reports that calcium channel blockers such as nifedipine and oxidative stress, reduces the PON1 activity, *in vitro*.[20] Söyüt *et al.*, 2014, reported the decreasing effect of the antimicrobial drugs in the PON1 activity.[21] The alteration was found to be more pronounced if the periodontal inflammation is caused by both nifedipine and bacterial infection. In the present study, the altered PON1 activity in the patients may be due to the drugs or may be due to the periodontal inflammation.

Increased oxidative stress and reduced antioxidant status as assessed by measuring MDA and GSH were earlier reported to be the major risk factors for CHD. Recently, it was reported that even minor alterations in systemic inflammation can impair the endothelial protective effects of HDL suggesting that the inflammatory process has a profound effect of the coronary artery disease.[17] However, similar studies in periodontal diseases were rarely reported.[22] Our study indicates that patients with the periodontal disease may be at a high risk of developing CHD. Improving the antioxidant status and reducing the gingival inflammation may be of help in reducing the risk of developing CHD.

Conclusion

The patients with periodontal disease are at a high risk of developing CHD compared to subjects with healthy periodontal tissues. Reducing the inflammation by replacing the drug, eliminating the bacterial infection, and by providing antioxidant-rich diet are suggested as methods for reducing the risk of CHD in patients with periodontitis.

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