

Effect of Periodontal Diseases on Plasma Level of LDL, HDL and Total Cholesterol in Rats

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Abstract

Introduction- Cardiovascular diseases (CVD) are known as the major life-threatening factors and the most common causes of mortality around the world, especially in developed countries. Many risk factors for CVD are well known, like dyslipidemia, diabetes mellitus, cigarette smoking, hypertension, positive family history, and aging. However, there is evidence recently showing a relation between periodontal diseases (PD) and increased risk of CVD. The basis of this study was to determine any relation between PD and serum levels of total cholesterol, LDL, and HDL so as to investigate whether periodontal disease can facilitate coronary atherosclerosis due to dyslipidemia.

Methods- In this experimental study, 20 healthy male rats weighing 200 – 250 grams were divided into case and control groups. In the case group (10 rats), we injected 0.6 ml of complete Freund's adjuvant in the mid-buccal area of both upper and lower jaws; and a sample of blood was taken from all 20 rats to measure the LDL, HDL, and cholesterol. After two weeks, the injection was repeated in the same areas with the same amount of drug; and at the end of the 4th week, blood sampling was repeated in both groups. The inflammation in the case group was confirmed with direct clinical observation and based on histological study at the end of the 4th week. Finally, the serum levels of LDL, HDL, and total cholesterol were compared between the two groups using the independent samples t-test.

Results- The statistical tests did not show any significant difference between the two groups. Also, we found no significant difference between the lab test values before and after the study procedure.

Conclusion- There was no certain relation between PD and cardiovascular diseases, except for their common risk factors. However, if any relation exists, it might be due to a mechanism other than the serum cholesterol level (*Iranian Heart Journal 2009; 10 (1):48-51*).

Key words: cardiovascular disease ■ periodontal disease ■ total cholesterol ■ LDL ■ HDL

The major pathology of coronary artery disease (CAD) is atherosclerotic plaque (AP) formation, which is related to the local collection of lipids. High amounts of serum LDL will lead to an increased accumulation of LDL in the AP and can result in coronary vascular accidents.

The importance of dyslipidemia and especially high levels of LDL in AP formation is clearly defined; and other factors such as cigarette smoking, arterial hypertension, diabetes mellitus, positive family history, and aging are also the main

risk factors of atherosclerosis.¹

The relation between inflammation and atherosclerosis and cardiovascular diseases (CVDs) has been proven^{2,3} and some evidence recently depicts a role for oral infections in atherosclerosis.⁴ It seems that a probable mechanism via inflammatory mediators and increase in WBC and platelet count accelerates the process of AP formation.^{5,6}

Periodontitis is a bacterial infection classified as a chronic local infection and caused by anaerobic gram negative microorganisms originating from dental plaque, which leads to inflammation due to entering lipo-polysaccharides and other microbial components into the gums.^{7,8} The pathogenesis of inflammation in gums is the increase of pre-inflammatory cytokines, resulting in the damage of the periodontal ligaments and alveolar bone.⁸ Many CAD risk factors are assumed as risk factors for periodontal diseases (PD) as well; in other words, there might be many common risk factors for PD and cardiovascular diseases.^{5,8} Hypercholesterolemia, especially increased LDL, is determined as a main risk factor for atherosclerosis; instead increased HDL cholesterol is related with a decreased risk of CAD.^{8,10}

A relation between periodontitis and hyperlipidemia has been shown in some animal laboratory studies,¹¹ and some human studies have shown that the periodontal state is worse in patients with hypercholesterolemia and CVDs⁸ and the amount of periodontal damage is related to the serum cholesterol level.¹²

There are still controversies about the relation between PD and CVDs in certain studies, and researchers have mentioned a need for more experimental results to clarify the exact relationship between these factors.¹³

The main aim of this study was to assess the effect of PD on serum levels of total cholesterol, LDL, HDL, and dyslipidemia in rats.

Methods

In this experimental case-control study, we entered 20 healthy male rats weighing between 200 – 250 grams after an initial clinical examination by a periodontist and a physiologist to confirm their periodontal health. All the rats were kept in similar environmental conditions (daylight, temperature) during the study period and had free access to a single kind of food.

First, we randomly divided the rats into two groups and took a blood sample from their periorbital vessels using hematocrit glass pipes. Then, the serum was separated with a centrifuge and after coding, the samples were sent to a lab. All the rats were identified with color marks on their bodies and codes of their blood samples. On the same day, we injected 0.06cc of complete Freund's adjuvant, a drug proven to cause chronic inflammation, into the midbuccal area of the upper and lower jaws of the rats in the case group. In the control group, we did not perform any injection. During the study, we induced general anesthesia in all the rats in a closed glass container with ether-inoculated cotton. Ethical considerations in working with animals were observed at all times.

After the procedures, the rats were moved to animal rooms in the laboratory; and after 14 days, we repeated the injection in the same regions with the same amount of drug. On the 28th day, blood sampling was done; and the serum levels of LDL, HDL, and total cholesterol were measured with an auto-analyzer.

To compare the mean of variables, the paired samples t-test and independent samples t-test at the 0.05 significance level were performed using SPSS[®] for Windows software.

The existence of inflammation in the case group was determined by direct clinical observation during the study period and confirmed in a microscopic examination of the histologic samples by a pathologist.

Results

In the case group (10 rats), the means of serum total cholesterol, LDL, and HDL levels were not significantly different from those in the control group in the first day of the study (Table I).

Table I. Comparison of the means of total cholesterol, LDL and HDL in rats of case and control groups.

Variable	Group	Before (1 st Day)		After (28 th Day)		P Value**
		Mean±SD	P Value*	Mean±SD	P Value*	
Cholesterol	Case	98.25±4.99	NS	78.11±10.2	NS	NS
	Control	98.40±4.31		71.62±8.78		
LDL	Case	49.80±4.20	NS	42.00±6.98	NS	NS
	Control	48.20±1.44		37.87±4.58		
HDL	Case	41.80±1.72	NS	33.33±3.31	NS	NS
	Control	43.00±1.24		32.00±4.44		

* Independent Samples t-test for equality of means in two groups

** Paired Samples t-test for equality of means before and after

One rat in the case group and two in the control group died during the study. There was no significant difference between the case and control groups at the end of the 28th day according to the independent samples t-test. The Kolmogrov-Smirnoff test was performed first and showed that the variables were normally distributed. Because of losing 3 rats, we controlled the loss bias with a sensitivity analysis method.

Discussion

The results of the current study are similar to those in previous studies in some aspects, although due to a lack of a specifically similar study we could not compare all the results together. Because of ethical limitations, many studies on humans are cross-sectional and the researchers cannot induce PD; therefore, assessing the association between CVDs and PD is not clearly applicable. Consequently, an assessment of the coexistence of PD with CVDs or some predisposing factors of CVDs has been considered in several studies (Losche et al. 2000; Emingil et al. 2000; Katz

et al. 2001; Katz et al. 2002; Ancabazile et al. 2002; Jeffrey et al. 2002; Scott et al. 2002 and Lopez et al. 2002¹⁴⁻¹⁸).

The coexistence of the two diseases has been noted apart from other risk factors;^{1,7,8,19-22} and in other studies, no evidence has emerged to define a relation between them.^{23,24} Also, in certain studies, the researchers have evaluated the relation of the two diseases without considering the common risk factors like cigarette smoking.^{23,24}

The results of the current study cannot precisely define a coexistence and relation between PD and plasma lipid levels. Also, there is no evidence in favor of a cause-and-effect relationship between the two phenomena. In fact, our results support studies which question the relation between PD and cardiovascular problems, apart from other risk factors.

Based on our present knowledge and the results, a lack of a proven cause-and-effect relationship between PD and plasma lipid levels can lead to two conclusions. Either there is no relation between PD and CVD and previous studies might have methodological errors or biased positive results; or the physiological mechanism in cardiovascular problems related with PD is not due to the effect of PD on plasma lipids (especially cholesterol, LDL, and HDL levels).

Conflict of Interest

No conflicts of interest have been claimed by the authors.

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