*Iranian Journal of Diabetes and Lipid Disorders; 2011; Vol 10, pp 1- 7*

# Correlation between oxidized-LDL and interleukin-6 in type 2 diabetic patients

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## Abstract

**Background:** Studies have suggested that oxidative stress is a common pathway of different leading mechanisms to diabetes complications. Oxidative stress play a crucial role in atherogenesis and cause oxidation of low density lipoprotein. Evidence has been shown that oxidized LDL in diabetic patients is higher than nondiabetic individuals. Regarding to known role of oxidative stress in developing of micro and macrovascular complications of diabetes and recent evidences about importance of IL-6 in initiating of inflammatory processes in atherosclerotic plaques formation and reports that shown the effects of Ox-LDL upon IL-6 release, in this study evaluation of serum levels of IL-6 and correlation of these two agents in diabetic patients in comparison with healthy persons was performed.

**Methods:** This stratified cross-sectional study was conducted in diabetic clinic of Imam khomeini Hospital, Tehran University of Medical Sciences during 2009-2010, recruiting 40 type2 diabetic (T2DM) patients as cases and 40 healthy subjects as controls. FBS, lipid profile, HbA1c, oxidized-LDL and IL-6 levels were measured for both patients and controls after 12 hours fasting state.

**Results:** The mean of Ox-LDL/LDL ratio in T2DM group (0.65±0.14) were significantly higher than control group (0.5±0.15) ($p<0.001$). The mean level of IL-6 in T2DM group were 2.6±1.8 pg/ml that was higher than control group (1.9± 0.8pg/ml) ($p=0.05$). BMI, systolic and diastolic blood pressures revealed significant correlation with IL- 6 level in diabetic group. There was no correlation between diabetes duration and IL-6 level.

**Conclusion:** We concluded that diabetes, as an independent factor, is responsible for increased IL-6 in T2DM.

**Keywords:** Interleukin-6, Diabetes mellitus, Oxidized LDL

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## Introduction

Diabetes is a chronic metabolic disorder that its prevalence is increasing and becomes a major worldwide health problem. Microvascular and macrovascular complications of diabetes decrease quality of life and life expectancy, and also imposes heavy economic and human burden on the community. Extensive studies have been accomplished to understanding the pathogenesis and preventive ways of diabetes complications. Studies have suggested that oxidative stress is common pathway of variant mechanism for diabetic complication's pathogenesis (1-2) . Oxidative stress play a crucial role in atherogenesis and cause oxidation of low density lipoprotein. Evidence has been shown that oxidized LDL in diabetic patients is higher than nondiabetic individuals (3-5). Study of Galland et al. showed increased Ox-LDL/LDL ratio in T2DM patients (6). In diabetic patients with macrovascular disease, this ratio is higher than patients without macrovascular disease (7) .

There are evidences that insulin resistance and type 2 diabetes are related to a chronic low grade inflammatory state. Cross sectional studies have reported that proinflammatory cytokines and acute phase markers are elevated in subjects with T2DM (8-12). IL-6 is a major cytokine that acts on the liver to stimulate the production of CRP (8). There is an increased risk for coronary artery disease in T2DM. It has been shown that subclinical (early) atherosclerosis increases with increasing degrees of glucose intolerance. Several mechanisms have been proposed to explain the accelerated and premature atherosclerotic changes seen with T2DM. This mechanisms includes inflammation (13-14), increased platelet activation (15-16) and oxidative stress (17). Studies have been shown the ability of oxidized-LDL to induce IL-6 secretion from monocytes (18), hepatocytes (19) and mesangial cells (20) in cell culture. In addition, IL-6 have found in atherosclerotic plaques. Given known role of oxidative stress in developing of micro and macrovascular complications of diabetes (21) and recent evidences regarding the role of IL-6 in initiating of inflammatory processes in atherosclerotic plaques formation and studies that shown the effects of ox-LDL on IL-6 release, in this study evaluation of serum level of IL-6 and correlation of these two agents in diabetic patients comparison with healthy persons is accomplished.

## Methods

This stratified cross-sectional study was conducted in diabetic clinic of Imam khomeini Hospital, Tehran University of Medical Sciences during 2009-2010, recruiting 40 type2 diabetic (T2DM) patients as cases and 40 healthy subjects as controls. Cases and controls were matched regarding age and gender and BMI. Smokers, those with creatinine levels equal or more than 1/5mg/dl, taking NSAIDS, Statins, Vitamins, Glucocorticoids; presence of heart failure class III and IV, infections and inflammatory disease and pregnant women were excluded from the study. The research was carried out according to the principles of the Declaration of Helsinki. The local ethics review committee of Tehran University of Medical Sciences approved the study protocol. All participants gave written informed consent before sampling. FBS, lipid profile, HbA1c, Oxidized-LDL and IL-6 levels were measured for both patients and controls after 12 hours fasting state. Serum oxidized-LDL and Il-6 were detected by ELISA assays.

### *Statistical Analysis*

Data were analyzed using SPSS software (version 13.0; SPSS Inc; Chicago, USA). Continuous variables are expressed as Means±standard deviation (SD). Comparisons between patients and controls were performed by Student's T-test for quantitative variables. We used Pearson correlation for quantitative variables. Regression analysis used to calculate IL-6 level . $P \leq 0.05$ was considered as statistically significant.

## Results

A total of 40 diabetic patients and 40 age, sex and BMI matched healthy adult volunteers were recruited in this cross sectional study. Demographic and biochemical characteristics of patients and controls are expressed in Table 1. There was no significant difference between groups with respect to age, sex, BMI, systolic and diastolic blood pressures, total cholesterol,

triglyceride and OX-LDL levels. HDL-C and LDL-C levels revealed significant difference between groups.

The mean levels of Ox-LDL/LDL ratio in T2DM group (0.65±0.14) were significantly higher than control group (0.5±0.15)($p<0.001$)(Figure 1). The mean level of IL-6 in T2DM group were 2.6±1.8 pg/ml that was higher than control group (1.9±0.8pg/ml)(P=0.05)(Figure 2). According Pearson correlation, as presented in Table 2, IL-6 significantly ($p\leq0.05$) correlated with BMI (r=0.32), systolic blood pressure (r=0.32) and diastolic blood pressure (r=0.35) in diabetic patients. In healthy controls, IL-6 was correlated with BMI (r=0.34), OX-LDL (r=0.35) and Ox-LDL/LDL(r=0.47) (Table 3). In diabetic patients, IL-6 was not correlated with OX-LDL and Ox-LDL/LDL. Regression analysis used for determining IL-6 level. By using BMI, LDL cholesterol and OX-LDL, in control group, this equation was statistically correlated with $R^2= 0.26$, r=0.51; P=0.015. It was not significant in diabetic patients.

**Table 1. Characteristics of patients and control participants**

| Variable | Diabetic patients (n=40) | Healthy controls (n=40) |
|---|---|---|
| **Age (year)** | 49±10 | 49±9 |
| **BMI (kg/m2)** | 27.7±4.3 | 28±3.6 |
| **Systolic Blood Pressure (mmHg)** | 118.4±14.6 | 115.25±10 |
| **Diastolic Blood pressure (mmHg)** | 75±10.19 | 76.87±6.47 |
| **Diabetes duration (years)** | 6±5.7 | - |
| **FBS (mg/dl)*** | 156.2±64.1 | 89.7±10.6 |
| **HbA1c (%)*** | 9.19±2.6 | 5.24±0.6 |
| **Triglyceride (mg/dl)** | 162.6±60.5 | 158.2±79.4 |
| **Total cholesterol (mg/dl)** | 198.3±36.4 | 214.3±44.2 |
| **HDL-c (mg/dl)*** | 38.5±10.5 | 48.6±12.8 |
| **LDL-c (mg/dl)*** | 105.1±20 | 135.2±38 |
| **IL-6 (pg/ml)**** | 2.6±1.8 | 1.9±0.8 |
| **OX-LDL (u/l)** | 68.4±18.9 | 66±20.7 |
| **OX-LDL/LDL*** | 0.65±0.14 | 0.5±0.15 |

*: P<0.001, **: p=0.05
Cross sectional study
T test analysis
Variables are expressed as Mean ± SD

**Table 2. Correlation coefficients among IL-6 and other variables in diabetic group**

| Variable | R |
|---|---|
| **Age** | 0.02 |
| **BMI *** | 0.32 |
| **Systolic Blood Pressure *** | 0.32 |
| **Diastolic Blood pressure *** | 0.35 |
| **Diabetes duration** | 0.001 |
| **FBS** | _0.01 |
| **HbA1c** | _0.003 |
| **Triglyceride** | 0.07 |
| **Total cholesterol** | _0.23 |
| **HDL-c** | _0.11 |
| **LDL-c** | _0.21 |
| **OX-LDL** | _0.02 |
| **OX-LDL/LDL** | 0.19 |

Number of diabetic patients: 40
Cross sectional study
*p ≤ 0.05, Pearson correlation

cholesterol and OX-LDL, it is possible to determine IL-6 level in healthy adults. It is not true in diabetic patients. In our study two groups were matched regarding BMI but diabetic patients had higher IL-6 levels than controls. This finding is consistent with recent performed study (10). Morohoshi showed in-vitro production of glucose dependent Interleukin-6 by human peripheral blood monocytes (29). This data verified that increased IL-6 in T2DM is independent of insulin resistance and obesity. In conclusion, it seems that diabetes can be considered as an independent contributor to development of inflammatory cytokines such as IL-6.

Nonetheless, our study does not support the hypothesis that Oxidized-LDL can influence IL-6 level in diabetics. Our results suggest that diabetes is an independent factor for releasing of inflammatory cytokines.

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