Association of Biomarkers of Inflammation and Endothelial Dysfunction in Obese Type 2 Saudi Diabetic Patients

O. Al-Jiffri, Fadwa M. Al-Sharif and Essam H. Al-Jiffri

Department of Medical Laboratory Technology, Faculty of Applied Medical Sciences, King Abdulaziz University, Saudi Arabia

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Abstract: The vascular complications of diabetes mellitus impose a huge burden on the management of this disease. Inflammation is one of the major factors in the formation of endothelial dysfunction. Endothelial dysfunction is a major contributor to the complications of diabetes mellitus. The aim of the present study was to investigate the possible relationship between inflammation and endothelial dysfunction in obese type 2 Saudi diabetic patients. Forty obese type 2 diabetic patients (24 males and 21 females) with body mass index (BMI) ranged from 31 to 35 Kg/m², non smokers, free from respiratory, kidney; liver, metabolic and neurological disorders were recorded in this study. Their age ranged from 35 to 55 years were included in this study as the first group (A). While a forty non-diabetic subjects (23 males and 22 females) not suffering of any disease, their age ranged from 30 to 45 years were included in this study as the second group (B) and considered as a control group. Findings of this study proved that elevated levels of BMI and HBA1c were associated with higher levels of CRP, TNF-α, IL-6 and E-selectin levels in diabetic patients. There was a strong association between elevated biomarkers of inflammation and endothelial dysfunction type 2 diabetes among Saudi people.

Key words: Endothelial Dysfunction Inflammatory Cytokine • Obesity and Non-Insulin Dependent Diabetes

INTRODUCTION

Diabetes mellitus (DM) is a growing metabolic disease that continues to be a leading health problem worldwide. The World Health Organization (WHO) has estimated that there are currently 346 million people affected by diabetes worldwide and anticipates that diabetes-related deaths would double by 2030 [1]. These figures highlight the importance of continued research and the need for novel methods to both prevent and treat this pandemic [2].

Obesity and diabetes are becoming pandemic and pose a major risk for a number of comorbidities including cardiovascular diseases. Adipose tissue is an active endocrine tissue, which secretes cytokines that appear to contribute to inflammation, atherosclerosis and may be involved in the etiology of type 2 diabetes, possibly constituting the missing link between obesity and insulin resistance (IR) [3].

Chronic low-grade inflammation, characterized by abnormal production of adipokines and inflammatory mediators, has been implicated in the pathogenesis of obesity-related chronic diseases including what may be called the obesity-type 2 diabetes mellitus (T2DM)-cardiovascular disease (CVD) triad [4]. Diabetes confers a high risk for CVD mortality that is modified little by the addition of the other MetS risk factors [5].

Cardiovascular disease (CVD), the number one cause of mortality in the USA, is almost twice as common in obese diabetic individuals [6]. Morbidity and mortality in type 2 diabetes is mainly associated with atherosclerotic cardiovascular disease and late complications as a result of dysfunction of plasma biomarkers of inflammation including tumor necrosis factor-α (TNF-α) and interleukin-6 (IL-6). [7]. Tumor necrosis factor α(TNF-α) induced insulin resistance by interacting with insulin receptor signaling and through activation of lipolysis and inhibition of lipoprotein lipase. TNF-α was proposed to
have preferentially paracrine effects and to be a regulator of insulin resistance at the tissue level [8-10]. C-reactive protein (CRP) was known to be produced primarily by the liver in response to inflammatory cytokines including interleukin (IL)-6 and TNF-α [9].

Interleukin (IL)-6 and tumor necrosis factor α (TNF-α) are two major pro-inflammatory cytokines, are secreted in significant amounts from adipose tissue and consequently obese women (healthy and diabetic) have higher cytokine levels than healthy lean women. Furthermore, increased levels of IL-6 and TNF-α are associated with deterioration of glycemic control, increased IR and dyslipidemia, contributing to the dysfunctional metabolic status of obese and type 2 diabetic individuals [11].

Several studies show the increase of inflammation markers in type 2 diabetes mellitus. This sub-clinical inflammation could cause changes in the function of the endothelium, which is a biologically active inner layer of the blood vessels. Endothelial cells secrete several substances that ensure the integrity of the blood vessel, prevent the accumulation of leukocytes and the occurrence of thrombosis on the surface of the endothelium and promote fibrinolysis to aid the dissolution of the micro-thrombi which may form [12].

Type II diabetes is characterized by three main metabolic disturbances (triggers): (1) hyperlipidemia, (2) early hyperinsulinemia and (3) hyperinsulinemia followed by pancreatic β-cell failure leading to hyperglycemia. Each of these metabolic disturbances acts as “triggers” eventually causing endothelial dysfunction through the influence of different “mediator” molecules [13].

Insulin regulates vascular function by stimulation of the expression of vascular cell adhesion molecule and E-selectin on endothelium. So, endothelial dysfunction is associated with insulin resistance [14].

Endothelial dysfunction is characterized by a shift of the actions of the endothelium toward reduced vasodilation, proinflammatory state and prothrombic properties [15]. Several of these substances, such as E-selectin, P-selectin, soluble intercellular cell adhesion molecule -1 (ICAM-1), soluble vascular cell adhesion molecule-1(VCAM-1), interleukin-6 and monocyte chemoattractant protein-1(MCP-1) have been studied in patients with type 2 diabetes mellitus and insulin resistance, showing that these substances are probably implicated in the pathophysiology of the disease [16].

The aim of the present study was to investigate the possible relationship between inflammation, endothelial dysfunction in obese type 2 Saudi diabetic patients.

**MATERIALS AND METHODS**

**Subjects:** Forty obese type 2 diabetic patients (24 males and 21 females) with body mass index (BMI) ranged from 31 to 35 Kg/m², non smokers, free from respiratory, kidney; liver, metabolic and neurological disorders were recorded in this study. Their age ranged from 35 to 55 years were included in this study as the first group (A). While a forty non-diabetic subjects (23 males and 22 females) not suffering of any disease, their age ranged from 35 to 55 years were included in this study as the second group (B) and considered as a control group. Informed consent was obtained from all participants.

This study was approved by the Scientific Research Ethical Committee, Faculty of Applied Medical Sciences at King Abdulaziz University. All participants were free to withdraw from the study at any time. If any adverse effects had occurred, the experiment will be terminated and the Human Subjects Review Board will be informed. However, no adverse effects occurred and so the data of all the participants were available for analysis.

**Evaluated Parameters:** Overnight fasting blood samples were obtained from each patient in clean tubes containing few mg of K2EDTA, centrifuged and plasma was separated and stored frozen at -20 ° used for estimation of C-reactive protein (CRP), plasma TNF-α, interleukin-6 (IL-6) and glycated hemoglobin (HBA1c) using colorimetric method were assessed enzymatically on a Beckman Synchrony CX5 Delta Clinical System (Beckman Coulter, Fullerton, CA, USA) using commercial reagents. Soluble E-selectin was measured by commercially available multiplex assays (Millipore, Massachusetts).

**Statistical Analysis:** The mean values of TNF-α, CRP, IL-6, HBA1c (%) and E-selectin obtained before and after two months in both groups were compared using paired "t" test. Independent "t" test was used for the comparison between the two groups (P<0.05). Pearson’s product moment correlation coefficients (r) were applied to examine the degree of correlation inflammation, endothelial dysfunction in obese type 2 Saudi diabetic patients.

**RESULTS**

The mean values of the clinical and laboratory parameters variables for participants in the two groups have a statistical significant difference (Table 1). Concerning the comparison between values of TNF-α,
Table 1: Mean value of clinical and laboratory parameters for participants in the two groups

<table>
<thead>
<tr>
<th></th>
<th>Diabetic group Mean +SD</th>
<th>Control group Mean +SD</th>
<th>T-value</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (year)</td>
<td>43.73±6.87</td>
<td>35.40±6.05</td>
<td>5.42</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>33.11±2.25</td>
<td>26.58±2.97</td>
<td>4.52</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>FBS (mg/dl)</td>
<td>180.32±14.45</td>
<td>87.63±6.75</td>
<td>9.87</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>PPS (mg/dl)</td>
<td>253.67±21.46</td>
<td>105.92±15.21</td>
<td>8.95</td>
<td>&lt;0.05</td>
</tr>
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<td>CRP (mg/dl)</td>
<td>17.86±3.17</td>
<td>9.45±2.12</td>
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<td>TNF- α (pg/mL)</td>
<td>5.98±1.65</td>
<td>4.02±1.43</td>
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<td>IL-6 (pg/mL)</td>
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<td>HBA1c (%)</td>
<td>7.96±1.24</td>
<td>6.05±0.87</td>
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<td>E-selectin (ng/ml)</td>
<td>14.89±6.02</td>
<td>8.71±3.51</td>
<td>5.84</td>
<td>&lt;0.05</td>
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BMI = Body mass index  FBS = Fasting blood sugar  PPS = Postprandial blood sugar

Table 2: Mean value and significance of TNF- α, IL-6, CRP, HBA1c (%) and E-selectin in diabetic group (A) and control group (B).

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DISCUSSION

There is a rising burden of type 2 diabetes which is mainly correlated to its vascular complications. This is reflected by a 4-fold increase in the incidence of coronary artery disease, a 10-fold increase in peripheral vascular disease and a 3- to 4-fold higher mortality rate with as much as 75% of diabetics ultimately dying from vascular disease [17, 18]. The primary findings of this study are that elevated levels of BMI, HBA1c, CRP, TNF- α, IL-6 were associated with higher levels of E-selectin levels in diabetic patients.

Our findings are consistent with other studies that have reported a significant association between elevated CRP and diabetes [19, 20]. However, Swellam et al. stated that elevated levels of CRP can be used for early diagnosis of type 2 diabetes mellitus and can predict diabetic complications [21]. Also Pradhan et al. concluded that elevated levels of CRP and IL-6 predict the development of type 2 diabetes mellitus [22]. Also, Liu et al. confirmed the role played by TNFα, IL-6 and CRP as a cause of Type 2 diabetes and further, have identified these molecules as early biological markers that may be used to more accurately predict future incidence of diabetes among apparently healthy individuals [23].

In our study E-selectin level is significantly higher in diabetic group than the control group. Our findings are consistent with Meigs et al. reported that endothelial dysfunction predicts type 2 diabetes in women [19]. Also, Thorand et al. supported the role of endothelial dysfunction in the etiology of type 2 diabetes [24]. However, levels of sE-selectin have been shown to be independently associated with diabetes [25, 26]. Also, the Women's Health Initiative Observational Study found E-selectin to be a predictor of diabetes; however, the U.S. women had higher median E-selectin levels, higher mean BMI as participants included in our study [27].

The mechanisms responsible for increased cardiovascular morbidity and mortality in individuals with diabetes are not fully explained by traditional risk factors and may be partially explained by inflammation and endothelial dysfunction. Visceral adiposity is associated with increased production of pro-inflammatory cytokines and hepatic CRP. ICAM-1 is also associated with visceral adiposity and may be upregulated by the increased production of pro-inflammatory cytokines [28]. Inflammation may promote development of diabetes by triggering beta cell dysfunction, apoptosis and impaired...
insulin signaling or development of hypertension by influencing platelet adhesion and aggregation, production of oxidants and induction of renal vasoconstriction [29].

CONCLUSION

The results from this study indicated a strong association between elevated biomarkers of inflammation and endothelial dysfunction type 2 diabetes among Saudi people. Strategies to reduce inflammation and endothelial dysfunction may be important therapeutic targets for people at risk of developing diabetes.

ACKNOWLEDGMENT

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REFERENCES


