ORIGINAL ARTICLE

Correlation of Serum Visfatin and Interleukin-6 in Obese and Non-Obese Male patients of Coronary Artery Disease (CAD)

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ABSTRACT

Background: Adipose tissue is an active endocrine secretory organ that produces different series of biologically active factors, the adipocytokines namely visfatin and interleukin-6 (IL-6). IL-6 is proinflammatory mediator in atherosclerosis which is released by macrophages. Visfatin is also a cytokine which function as growth factor for vascular smooth muscle cells, a hallmark of atherosclerosis and anti apoptotic effects on localized neutrophils.

Aim: To investigate the correlation of these adipocytokines in CAD to find a link between serum visfatin and IL-6 in atherosclerosis leading to CAD.

Methods: It was cross sectional analytical study conducted in Punjab Institute of Cardiology, Lahore from July 2013 to December 2013. In this study forty male subjects with angiographically proven stable symptomatic CAD further divided into 20 non obese of same age and waist circumference with CAD and 20 obese male patients with CAD. Serum visfatin and interleukin-6 levels were assayed by enzyme-linked immunosorbent assay (ELISA). The collected data was entered and analyzed using SPSS version 20. Data normality was explored by using the Shapiro Wilk's test.

Spearman rho correlation was used to correlate serum visfatin and interleukin-6 for non normally distributed data. A *p*-value of ≤0.05 was considered statistically significant.

Results: Significant positive correlation between serum visfatin and serum interleukin-6 was found in CAD in obese and non obese male patients (n=40) (rho=0.680, *p*=0.000). Significant correlation persist when partial correlation was applied after controlling waist circumference and body mass index.

Conclusion: Strong positive correlation between serum visfatin and IL-6 in male patients with CAD suggest that circulating serum visfatin may be related with proinflammatory effects in CAD.

Keywords: Serum visfatin, interleukin-6, coronary artery disease

INTRODUCTION

The adipose tissue is an active endocrine secretory organ that produces a heterogeneous series of bioactive factors called adipocytokines namely adiponectin, leptin, resistin, visfatin, free fatty acids, tumor necrosis factor, interleukin-1 & interleukin- 6 having both local & systemic effects[1'2].

They are involved in regulation of food intake, body weight, insulin sensitivity, immunity, inflammation and vascular homeostasis[3].

The abdominal adipocity and abnormal adipokine production exert greater influence on Coronary artery disease (CAD). CAD is the most significant cause of cardiovascular mortality all over world and causes >4.5 million deaths in the developing world[4]. The rising concept is that inflammation initiates the disease and plays a role in progression as well. Release of different cytokines by local inflammatory cells leads to activation of endothelial cells and changes their natural anticoagulant property.

Plasma levels of visfatin are increased in CAD and in acute ST elevation myocardial infarction[5]. IL-6 plays an important role in initiating coronary artery disease by recruiting monocytes & macrophages to the vessel wall[6]. Visfatin is an adipokine produced by lymphocytes and macrophages[7]. It causes atherosclerosis by inducing leukocyte adhesion to vascular endothelial cells by induction of cell adhesion molecules(CAM) and it acts a growth factor for vascular smooth muscle cells proliferation and inflammation by anti apoptotic effect on the localized neutrophils[1].

It causes release of interleukin- 6 by macrophages[8]. Interleukin-6 is a cytokine have proinflamatory effect on B cells and T cells. It is produced from T lymphocytes, monocytes, fibroblasts, endothelial cells & normal hematopoietic cells[9]. Plasma concentration of IL-6 is increased in patients of acute myocardial infarction[6].

The purpose of this study is to find the correlation of visfatin & IL-6, and visfatin as an inflammatory biomarker in the development of CAD in non obese & obese patients.

METHODOLOGY

This cross sectional and comparative study was done from July 2013 to December 2013.The subjects were selected from registered cases of Punjab Institute of

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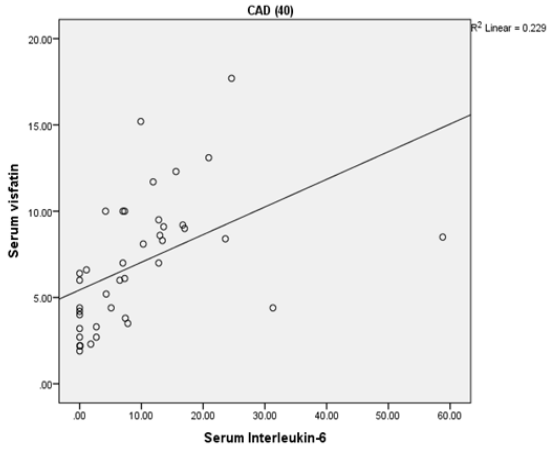
Cardiology (PIC). The study was approved by the respective Ethical Review Board of the institution. The participants included 20 non obese with coronary artery disease body mass index (BMI) less than 25 kg/m² waist circumference less than 90 cm(according to Asian pacific criteria) group 1 and 20 obese males with coronary artery disease, BMI more than 25 kg/m² and waist circumference more than 90 cm(all cases angiographically confirmed) group 2. All the participants were in the age range of 35-55 years. Diabetics and nonsmokers were excluded. All participants were explained about the nature of the study and an informed consent was taken from each of them. Blood pressure was recorded and blood samples were obtained from the subjects. Serum was aliquoted and kept at -20°C.

Serum visfatin was measured by, Human ELISA kit (Enzo Life sciences (ELS) AG.Switzerland) with an analyzer STAT FAX 303 Reader and interleukin-6 levels were analyzed by Enzyme Link Immunosorbent Assay (Elisa) kit (IBL international GMBH Germany) with analyzer STAT FAX 303 Reader. Laboratory work was performed at Research Laboratory, Lahore General Hospital. Data was entered and analyzed by using SPSS version 20.0. Non-parametric tests of significance were applied where data regarding serum visfatin and interleukin-6 levels deviated from normality. Spearman's rho correlation was applied and partial correlation was also applied. P-value ≤ 0.05 was considered statistically significant.

RESULTS

Significant positive correlation between serum visfatin and serum interleukin-6 was found in CAD in obese and non obese male patients (n=40) (rho= 0.680, *p*= 0.000). Significant correlation persist when partial correlation was applied after controlling waist circumference and body mass index.



Determination of Variables

Biochemical parameters	A (n=20)	B (n=20)
Serum interleukin-6 (pg/ml)	0.6000 (0.000- 4.9000)	12.800 (7.300-16.425)
Serum Visfatin (ng/ml)	3.90 (2.700 - 5.000)	9.05000 (8.150-10.00)

Scatter plot showing significant positive correlation between serum interleukin-6 and serum visfatin in coronary artery disease males (A and B, n=40) using spearman's rho correlation coefficient (p value= 0.000*)

Partial correlation:

Controlling variable, BMI Waist circmference	Interleukin-6	Visfatin

P value=0.049*

DISCUSSION

The results of the present study showed significant positive relation of levels of serum visfatin with IL-6 in CAD (n=40) patients. This positive correlation of visfatin and IL-6 has been reported in literature[10],[11]. Visfatin /NAMPT promotes the macrophages survival through the release of IL-6[12]. Circulating visfatin levels are positively related with serum levels of IL-6 and CRP in healthy Japanese American population[8] as well as in healthy Korean women[10]. Non significant positive correlation has been observed in experimental studies in which visfatin has been shown to stimulate the production of IL-6 in monocytes and in circulation in mice[8].

The interrelationship between Il-6 and serum visfatin has been observed in different other studies. IL-6 treatment of an amniotic epithelial cell line stimulated the visfatin /PBEF gene expression[13]. Since IL-6 is a reliable inflammatory marker[14],[15] a positive correlation with visfatin means serum visfatin can be considered an inflammatory marker. Expression of visfatin has been seen to be up-regulated in activated neutrophils and it has inhibitory effect on the apoptosis of neutrophils[16]. Monocytes and macrophages are additional sources of serum visfatin which shows its role in acute inflammatory diseases. Serum visfatin activates human leukocytes and induces cytokine production especially IL-6[8]. The raised levels of serum visfatin in different non metabolic diseases probably show an inflammatory role, like osteoarthritis, crohn's disease, ulcerative colitis, rheumatoid arthritis[17], inflammatory bowel disease and in septic conditions like chorioamnionitis[18]. In acute lung injury[19,20] visfatin was considered as biomarker of inflammation[21].

Serum visfatin had also strong association with endothelial cell adhesion molecules which make it biomarker for prediction of endothelial damage and

future risk of CAD in chronic kidney disease[17,22]. Vanhoutte et al., in 2009[23] reported serum visfatin as marker of endothelial dysfunction and it is first step in atherosclerotic process in CAD. So raised levels of serum visfatin play a significant role in atherosclerotic plaque damage.

When partial correlation was applied BMI and waist circumference was controlled even then significant correlation persist (p value= 0.049*). This showed probably serum visfatin and IL-6 were not only released from adipose tissue. There are other sources also. These results suggest that circulating visfatin may be related with some proinflammatory conditions even in non diabetic situation and independent of obesity.

CONCLUSION

Strong positive correlation between serum visfatin and IL-6 in male patients with CAD suggests that circulating serum visfatin may be related with proinflammatory effects in CAD independent of obesity.

Limitations of study: The sample size of the study was not large enough to validate these results in entire population. Only male subjects were included in the study because the levels of serum visfatin are variable in female as proved in studies. In order to include females sample size had to be extended.

Recommendation: Further clinical and prospective studies with large no of subjects are needed to determine the predictive value of serum visfatin as biomarker of inflammation.

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