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Role of Serum Leptin and its association with Malondialdehyde in Type 2 Diabetic Patients

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Abstract
Background: *Leptin is a hormone produced by adipose tissue that regulates a number of physiological processes and behaviors including appetite, body weight, neuroendocrine functions and glycaemia. These effects are mediated via actions on leptin receptors (LepRs) expressed by neurons in the central nervous system (CNS).*
Aim*: The aim of this study was to evaluate the significance of leptin levels in T2DM patients compared with controls and to determine the association of leptin with malondialdehyde (MDA)*
Materials and methods: *Thirty T2DM patients with the age group of 35 to 50 years were selected for this study and 30 healthy age matched subjects were selected as a control group. Serum leptin was assessed by ELISA, malondialdehyde (MDA) was assessed by Thiobarbituric Acid Reactive Substances (TBARS) method and routine investigations were done by ERBA EM-360 fully automated analyzer.*
Results: *The mean level of serum leptin was significantly increased in T2DM patients compared with controls. Serum leptin levels were positively correlated with FPG, PPG, HbA1c, Homeostasis model assessment of insulin resistance (HOMA-IR), malondialdehyde, Cholesterol, TGL, LDL, BMI and negatively correlated with HDL.*
Conclusion: *Leptin might be useful diagnostic marker for the severity of inflammation, chronicity of the disease and vascular complications.*
Keywords: *Leptin, Type 2 diabetes mellitus (T2DM), Malondialdehyde (MDA)*

I. Introduction

Diabetes mellitus (DM) is a significant cause of global morbidity and mortality worldwide [1]. As type 2 diabetes mellitus (T2DM) is commonly associated with obesity, it has been proposed that adipocytokines, which are produced by the adipose tissue, can directly influence the pro-atherogenic and pro-inflammatory environment of the vascular walls and other bioactivities [2]. It is thought that adipocytokines contribute to the increased risk of vascular complications in patients with type 2 diabetes by modulating vascular function and affecting inflammatory processes [3].

Leptin is a 16-kDa hormone identified and cloned in 1994, is synthesized and secreted from white adipose cells [4]. Leptin has a variety of important central and peripheral actions to regulate energy balance, fertility and bone metabolism that are mediated by specific cell surface leptin receptors expressed by neurons in the central nervous system (CNS) [5-8]. Leptin transmits signal by binding to its receptor Ob-R, which belongs to the class I cytokine receptor family. The long form, namely Ob-Rb is essential in mediating most of the biological effects of leptin and is highly expressed in the hypothalamus. It is also found to be expressed in several cell types relevant to Cardiovascular Diseases (CVD) (eg, macrophage, endothelial cell, and smooth muscle cells) thus providing evidence for the role of leptin in signaling for atherogenic events [9]. Leptin also plays an important role in inflammatory processes involving T cells and has been reported to modulate T-helper cell activity in the cellular immune response [10,11]. Hence, leptin has a dual role in inflammation: it activates monocytes and macrophages, potentiates production of the proinflammatory cytokines TNF-a, IL-6 and IL-9, and directs T-cell differentiation to Th1 phenotype [12-15]. Malondialdehyde (MDA) is one of the end products of oxidative reactions, can be detected in several biological fluids and tissues and is therefore used as a biomarker of lipid peroxidation and oxidative stress [16, 17]. So, the objective of the present study was to evaluate the significance of leptin levels in T2DM patients compared with controls and to determine the association of leptin with malondialdehyde.

II. Materials and methods

A total of 30 type 2 diabetic patients of both sexes aged between 35-50 years on oral hypoglycemic drugs, attending diabetic out-patient department of NIMRA Institute of Medical Sciences, Jupudi, Andhra Pradesh, India, were selected for our study. We excluded the patients based on the following criteria: Patients on

insulin, Smokers, Alcoholics, Tobacco chewers, abnormal urinary sediment, urinary tract infection, history of other renal disease and active or chronic persistent infection or inflammatory disorders, neoplastic disorders, uncontrolled thyroid disorders, severe liver dysfunction, history of acute myocardial infarction, stroke, and occlusive peripheral vascular disease. Thirty healthy individual age, sex matched subjects were selected as control. The informed consent was obtained from all the study subjects and the study was approved by the Institutional Human Ethics Committee (IHEC). Experiments were done in accordance with Helsinki declaration of 1975.

Biochemical analysis:

Fasting blood samples were obtained from the subjects and centrifuged at 2000×g for 10 min. Samples were analyzed for glucose, lipid profile (Total Cholesterol, HDL, LDL, triglycerides) using by ERBA EM-360 fully automated analyzer. Serum leptin and Insulin was assessed by Enzyme Linked Immuno Sorbent Assay (ELISA).Serum malondialdehyde (MDA) was estimated by Thiobarbituric Acid Reactive Substances (TBARS) method [18] and the 2 hour post prandial venous blood sample collected for plasma glucose (PPG) analysis. Homeostasis model assessment for insulin resistance evaluation (HOMA-IR) was calculated using the equation: fasting plasma insulin × glucose/22.5 [19].

Statistical analysis: Statistical analyses were carried out with SPSS 20.0. Values were expressed as mean ± standard deviation by t-test, p value < 0.05 was considered statistically significant. The Pearson correlation test was used for correlation analysis.

III. Results

Table 1: Comparison of baseline characteristics between controls and T2 DM patients

Parameters	Control group (n=30)	Study group (n=30)	p- value
Age	40.08±5.5	40.5±4.6	0.672
Males (%)	80.3	84.6	NA
Females (%)	19.7	14.4	NA
Body mass index (BMI)	24.2±1.3	26.8±2.9	0.001
Waist/Hip ratio	0.91±0.04	0.92±0.06	0.105
Systolic BP (mm Hg)	112.1±5.2	121.8±7.2	0.05
Diastolic (mm Hg)	78.5±3.2	83.2±5.0	0.03

Data are expressed as mean ±SD, p value <0.05 was considered statistically significant.

Table 2: Comparison of biochemical parameters between control and T2DM patients

Parameters	Control group (n=30)	Study group (n=30)	p- value
Fasting plasma glucose (mg/dl)	85.7±9.5	148.8±10.6	0.001
post prandial plasma glucose (mg/dl)	118.5±17.8	230.2±22.4	0.001
HbA1C (%)	5.8±0.4	8.2±2.3	0.001
Serum cholesterol (mg/dl)	174.5±18.1	203.1±27.6	0.001
Serum Triglycerides (mg/dl)	92.5±25.3	149.3±39.7	0.004
HDL cholesterol (mg/dl)	44.5±5.4	38.2±4.1	0.009
LDL cholesterol (mg/dl)	114.1±19.5	137.3±21.4	0.004
Creatinine(mg/dl)	0.69±0.14	0.7±0.19	0.08
Insulin (µIU/ml)	6.5±0.7	10.8±2.6	0.001
HOMA-IR	1.32±0.17	3.17±0.99	0.001
Malondialdehyde (µ mol/L)	1.9±0.52	3.9±0.51	0.001
Serum Leptin (ng/ml)	6.8±0.9	14.2±3.6	0.001

Data are expressed as mean ±SD, p value <0.05 was considered statistically significant.

Table 3: Correlation between Serum Leptin &measured parameters

Parameters	Correlation Coefficient(r)	p- value
FPG	0.531**	0.001
PPG	0.321*	0.042
HbA1c	0.489**	0.002
HOMA-IR	0.533**	0.001
Cholesterol	0.535**	0.002
TGL	0.639**	0.001
HDL	-0.598**	0.001
LDL	0.369**	0.004
Malondialdehyde	0.669**	0.001
BMI	0.294*	0.023
W/H ratio	0.138	0.292

**Correlation is significant at the 0.01 level (2-tailed).

*Correlation is significant at the 0.05 level (2-tailed).

Table 4: **Correlation between MDA& measured parameters**

Parameters	Correlation Coefficient(r)	p- value
FPG	0.541**	0.001
PPG	0.311*	0.004
HbA1c	0.498**	0.001
Cholesterol	0.535**	0.002
TGL	0.639**	0.001
HDL	-0.538**	0.001
LDL	0.369**	0.004

**Correlation is significant at the 0.01 level (2-tailed).
*Correlation is significant at the 0.05 level (2-tailed).

IV. Discussion

Adiposity and altered adipokine secretion seem to predispose to the development of type 2 diabetes [20]. In the present study, we observed that serum leptin and MDA levels were significantly increased in T2DM subjects compared with controls. It also shows that leptin levels were positively correlated with FPG, PPG and HbA1c, HOMA-IR which means indicating its relevance of low grade systemic inflammation. Moreover, it is shown that not only chronic hyperglycemia but also lipid abnormalities and elevated BMI accelerate development of inflammatory process in T2DM patients. Increased leptin levels leads to activation of several proinflammatory cytokines and its lack of activity leads to insulin resistance [21-23]. Leptin potentiates secretion of tumor necrosis factor and interleukins 2 and 6 increases generation and accumulation of reactive oxygen species, and enhances expression of monocyte chemoattractant protein-1 [24,25]. Leptin stimulates production of proinflammatory cytokines and enhances production of Th1-type cytokines. In endothelial cells, leptin stimulates transforming growth factor-β synthesis [26, 27].

Leptin may increase oxidative stress through multiple mechanisms. In bovine aortic endothelial cells, leptin increases formation of reactive oxygen species in a process coupled with increased fatty acid oxidation and activation of protein kinase A [25]. And also our study shows that serum MDA levels shows strong positive correlation between FPG, PPG, HbA1c, total cholesterol, LDL,TGL and negative correlation with HDL. Co-occurrence of obesity and T2DM could lead to increased MDA, in which leptin may be involved, at least in part. So, in this regard, the current data support for the hypothesis that, chronicity of T2DM may leads to inflammatory condition. Oxidative stress can be described as a condition resulting from an uncontrolled increase in free oxygen radicals or an insufficiency in the antioxidant system under certain pathological states. Free oxygen radicals have important toxic effects; chiefly the hydroxyl radical and to a lesser extent the superoxide anion lead to peroxidation of membrane lipids thereby causing production of MDA [28]. Therefore, alternation of leptin levels may be a part of the etiology of inflammatory process and vascular complications of T2DM.

In conclusion serum leptin might be potentially useful diagnostic marker for the severity of inflammation, chronicity of the disease and vascular complications.

References

[1]. Soares AL, Fernandes AP, Cardoso JE, Sousa MO, Lasmar MC, Novelli BA, Lages GF, Dusse LM, Vieira LM, Lwaleed BA, Carvalho MG. Plasma total homocysteine levels and methylenetetrahydrofolate reductase gene polymorphism in patients with type 2 diabetes mellitus. Pathophysiol Haemost Thromb. 2008; 36(5):275-81.
[2]. Meier U, Gressner AM. Endocrine regulation of energy metabolism. Review of pathobiochemical and clinical chemical aspects of leptin, ghrelin, adiponectin, and resistin. Clin Chem 2004; 50: 1511-1525.
[3]. Matsuda M, Shimomura I, Sato M, et al Role of adiponectin in preventing vascular stenosis. J Biol Chem 2002; 277: 27487-27491.
[4]. Zhang Y, Proenca R, Maffei M, Barone M, Leopold L, Friedman JM. Positional cloning of the mouse obese gene and its human homologue. Nature. 1994; 372: 425-432.
[5]. Margetic S, Gazzola C, Pegg GG, Hill RA. Leptin: a review of its peripheral actions and interactions. Int J Obes Relat Metab Disord. 2002; 26: 1407-1433.
[6]. Seufert J. Leptin effects on pancreatic beta-cell gene expression and function. Diabetes. 2004; 53 (suppl 1): S152–S158.
[7]. Friedman JM. Leptin at 14 y of age: an ongoing story. Am J Clin Nutr. 2009;89:973S-979S.
[8]. Flier JS, Maratos-Flier E. Lasker lauds leptin. Cell. 2010;143:9-12.
[9]. Dubey L, Hesong Z. Role of leptin in atherogenesis. Exp Clin Cardiol. 2006;11:269-75.
[10]. Juge-Aubry CE, Meier CA. Immuno modulatory actions of leptin. Mol Cell Endocrinol .2002; 194:1-7.
[11]. Matarese G, Moschos S, Mantzoros C. Leptin in immunology. J Immunol 2005; 173 :3137-42.
[12]. Matarese G, La Cava A, Sana V et al. Balancing susceptibility to infection and autoimmunity: a role for leptin? Trends Immunol. 2002; 23:182-6.
[13]. La Cava A, Alviggi C, Matarese G. Unravelling the multiple roles of leptin in inflammation and autoimmunity. J Mol Med .2003; 82:4–11.
[14]. Meyers JA, McTiernan A, Ulrich CM. Leptin and immune function: integrating the evidence. Nutr Res 2005; 25:791-803.
[15]. Li J, Li F, Zhao A. Inflammation and leptin. Drug Discov Today 2006; 3:387-93.
[16]. Milne GL, Yin H, Brooks JD, et al. Quantification of F2-isoprostanes in biological fluids and tissues as a measure of oxidant stress. Methods Enzymol .2007; 433:113-126.

[17]. Todorova I, Simeonova G, Kyuchukova D, Dinev D, Gadjeva V. Reference values of oxidative stress parameters (MDA, SOD, CAT) in dogs and cats. Comp Clin Path.2005; 13: 190-194.
[18]. Mahfouz MO, Hariprasad CH, Shaffie IA, Sadasivudu B. Serum Malondialdehyde levels in myocardial infarction and chronic renal failure. IRCS Med Sci 1986; 14: 1110- 1111.
[19]. Matthews D.R., Hosker J.P., Rudenski A.S. Homeostasis model assessment: Insulin resistance and beta cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia. 1985;13:412-419.
[20]. Combs TP, Berg AH, Obici S, Scherer PE, Rossetti L.Endogenous glucose production is inhibited by the adipose-derived protein Acrp30.J Clin Invest. 2001 Dec;108(12):1875-81.
[21]. Zhang Y, Proenca R, Maffei M, Barone M, Leopold L, Friedman JM. Positional cloning of the mouse obese gene and its human homologue. Nature. 1994 Dec 1;372(6505):425-32.
[22]. Santos-Alvarez J, Goberna R, Sánchez-Margalet V. Human leptin stimulates proliferation and activation of human circulating monocytes.Cell Immunol. 1999 May 25;194(1):6-11.
[23]. Enriori PJ, Evans AE, Sinnayah P, Cowley MA. Leptin resistance and obesity. Obesity (Silver Spring). 2006 Aug;14 Suppl 5:254S-258S.
[24]. Loffreda S, Yang SQ, Lin HZ, Karp CL, Brengman ML, Wang DJ, Klein AS, Bulkley GB, Bao C, Noble PW, Lane MD, Diehl AM. Leptin regulates proinflammatory immune responses. FASEB J. 1998; 12: 57-65.
[25]. Yamagishi SI, Edelstein D, Du XL, Kaneda Y, Guzmán M, Brownlee M. Leptin induces mitochondrial superoxide production and monocyte chemoattractant protein-1 expression in aortic endothelial cells by increasing fatty acid oxidation via protein kinase A. J Biol Chem. 2001; 276: 25096-25100.
[26]. Sanchez-Margalet V, Martin-Romero C, Santos-Alvarez J, Goberna R, Najib S, Gonzalez-Yanes C. Role of leptin as an immunomodulator of blood mononuclear cells: mechanisms of action. Clin Exp Immunol. 2003; 133: 11-19.
[27]. Wolf G, Hamann A, Han DC, Helmchen U, Thaiss F, Ziyadeh FN, Stahl RA. Leptin stimulates proliferation and TGF-beta expression in renal glomerular endothelial cells: potential role in glomerulosclerosis. Kidney Int. 1999; 56: 860-872.
[28]. Stocker R, Frei B. Endogenous antioxidant defences in human blood plasma. In: Sies H ed. Oxidative Stress: Oxidants and Antioxidants. London: Academic Press.1991; 213-242.