

ASSOCIATION OF OBESITY AND LEPTIN WITH INSULIN RESISTANCE IN TYPE 2 DIABETES MELLITUS IN INDIAN POPULATION

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Abstract : Obesity and diabetes mellitus are two modern epidemics. But their interrelationship is debated. Here we explored the probable association among obesity, leptin and insulin resistance in type 2 diabetes mellitus. 60 recent onset (<5 years) diabetics and age-sex matched 33 non diabetic controls were assessed for physical and chemical parameters like Body Mass Index, abdominal circumference, waist/hip ratio, fasting blood glucose, insulin and leptin. Degree of insulin resistance was calculated by HOMA-IR method (Homeostatic Model Assessment). All the physical parameters showed positive correlation with leptin and the HOMA-IR score, strength of association being highest between insulin resistance and abdominal circumference. Leptin and insulin resistance showed no correlation. Findings were lower in controls. Study concluded that, obesity mainly central type might be responsible for insulin resistance in type 2 diabetes mellitus where as leptin, a potential marker for obesity, may not. This perhaps points towards the multifactorial causation of insulin resistance in type 2 diabetes mellitus.

Key words : leptin insulin resistance HOMA-IR obesity

INTRODUCTION

Type 2 diabetes is increasing rapidly to the extent of epidemic worldwide. It may lead to number of macro and micro vascular complications if glycemic control is not adequate. It has been seen that insulin

resistance is often an ancillary metabolic derangement in type 2 diabetes characterized by postprandial hyperglycemia and compensatory hyperinsulinemia. Overt diabetes results when beta cells of pancreas no longer can afford excess insulin secretion in compensation of insulin resistance (1). But

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exact underlying mechanism that may lead to insulin resistance is still unclear. Obesity is the leading cause by some school of thoughts, (2, 1) where as there are another group of studies who failed to establish any direct association between insulin resistance and obesity (3, 4). Traditionally, fat tissue was considered to be solely an energy storage depot having only a passive function in the body. However, recent studies have shown that fat tissue exerts important endocrine functions, which are mediated by a complex network of various soluble factors, derived from adipocytes (fat cells), called adipocytokines. These are a group of novel and highly active molecules, which are abundantly secreted by adipocytes, and act at both the local and systemic level. Since their discovery in the early 90s, around 20 members of the adipocytokine family have been identified so far. Adiponectin and leptin are the most abundant adipocytokines produced by adipocytes, and the best-studied molecules in this class till date. Their role has been studied in relation to various pathological conditions like deranged energy homeostasis, abnormal leukocyte migration, polycystic ovary etc (5). But adequate evidence is still lacking regarding the influence of above molecules in the pathogenesis of insulin resistance. Moreover, Many of the works done previously were either with healthy non diabetic population (6) or regarding some other co-morbidities like atherosclerosis, coronary artery diseases etc (7) or done in a different geographical background (8). Findings of the studies were also found to be controversial in some cases. When some of the Studies showed positive correlation between leptin and insulin resistance (6), other found reduced level of leptin in type 2 diabetes (8).

With this background we did our study in type 2 diabetics who are really uncompensated insulin resistant Indian population and compared the findings with that of non-diabetic controls. Thereby we tried to explore the association of obesity and leptin with actual insulin resistance in our study. We also tried to assess the strength of association between different physical parameters and degree of insulin resistance and thus to point out the type of obesity contributing more to insulin resistance. Therefore, our study indirectly explored the possibility of improving the degree of insulin resistance and thereby adequacy of glycemic control by tackling obesity and its specific type.

MATERIALS AND METHODS

This was a hospital based cross-sectional study.

Inclusion criteria

60 recent onset (<5 years of disease duration) type-2 diabetics of 30- 60 years of age and of both sexes, attending the General Medicine and Diabetic OPD of Calcutta National Medical College and Hospital and 33 age sex matched non diabetic healthy control were included in the study. Informed consent was duly taken from each subject under study, and the entire procedure was done as per the Institutional ethical permission.

Exclusion criteria

Subjects with any other chronic illness like tuberculosis, malignancy, hepatitis due to any cause, hypertension, renal diseases,

hormonal derangements other than diabetes like Cushing's syndrome, hypothyroidism etc, pregnancy, alcoholism, and any other acute or chronic illness related or unrelated to diabetes were excluded from the study. Patient on insulin therapy or on oral hypoglycemics other than short acting 2nd generation sulfonylurea were also excluded.

Study design

All the selected subjects were assessed for physical parameters (e.g. height, weight, BMI, abdominal circumference, waist/hip ratio). 5 ml of venous blood was collected from the ante cubital vein of each subject after 12 hours of fasting and separated the same in a fluoride- oxalate bulb (2ml) and a clot vial (3ml) for blood glucose and serum insulin, leptin respectively. From the findings degree of insulin resistance was calculated by HOMA-IR method (Homeostatic Model Assessment-Insulin Resistance). HOMA-IR scores were derived by multiplying fasting blood glucose (in millimoles/liter) and fasting serum insulin (in microunits/milliliter) divided by 22.5. Blood glucose was measured by Glucose Oxidase method by using Eco Gluco Kit from Crest Biosystems, Santacruz, India. Serum insulin was measured by using human ELISA kit from MAPS, Monobind Inc, Lake Forest, USA. Levels of leptin were estimated by ELISA method by using human ELISA kit from Ray Biotech, USA.

Statistical analysis

Data was analyzed in Microsoft Excel and SPSS software. Value of individual parameter was expressed as mean and one standard deviation. Association among the parameters was tested by calculating co-efficient of

correlation (r) and significance of difference of the means within the groups was tested by unpaired Student's t- test. Every where $P < 0.05$ was considered to be significant.

RESULTS

Total 60 type 2 diabetics (<5 years disease duration) and 33 age sex matched non-diabetic controls were studied. Average age, male: female ratio and mean values of different physical and bio-chemical parameters among them are summarized in Tables I & II respectively.

TABLE I: Physical parameters of study & control group.

<i>Parameters value</i>	<i>Study group (n=60)</i>	<i>Control group (n=33)</i>	<i>P value</i>
Age (years)	47.47±9	45±10.2	0.2304
Male: Female	27 : 33	14 : 19	
Total body weight (kg)	64.16±.19	55.15±0.73	0.00*
BMI (Weight in kg/ Height in meter ²)	27.34±.51	25.88±0.88	0.04*
Abdominal circumference (cm)	93.53±0.76	89.28±4.57	0.01*
Waist/Hip ratio	0.93±0.05	0.88±0.03	0.00*

Data expressed are mean±SD. *P<0.05 significant.

TABLE II: Bio-chemical parameters of study & control group.

<i>Parameters value</i>	<i>Study group (n=60)</i>	<i>Control group (n= 33)</i>	<i>P value</i>
Fasting blood glucose (mg/dl)	168.97±77.21	83.33±7.9	0.00*
HOMA-IR Score	2.14±1.78	1.25±0.48	0.005*
Leptin (ng/ml)	15.61±13.68	10.27±3.83	0.03*

Correlation values

Significant positive correlations were found between leptin and total body weight ($r=0.43$, $P<0.05$), BMI ($r=0.73$, $P<0.05$), abdominal circumference ($r=0.55$, $P<0.05$), but correlation between leptin and HOMA-IR score was not significant. Physical parameters showed significant positive correlations with HOMA-IR score with coefficients of correlation as follows: total body weight ($r=0.35$, $P<0.05$), BMI ($r=0.34$, $P<0.05$) abdominal circumference ($r=0.44$, $P<0.05$) and waist/hip ratio ($r=0.43$, $P<0.05$). Among the physical parameters strength of association was found highest between abdominal circumference and HOMA-IR score. All the physical parameters for obesity and chemical parameters like HOMA-IR score and levels of leptin were found to be significantly lower in non-diabetic controls than in diabetics.

DISCUSSION

The prevalence of obesity and overweight is steadily increasing in most human populations and has reached epidemic proportions in many westernized societies. According to the World Health Organization's 2005 global estimates, about 1.6 billion adults are overweight and 400 millions are obese. Globally, obesity is a major contributor to the burden of disabilities and several chronic diseases including hypertension, type 2 diabetes and heart diseases (9).

Leptin is a multiple-function adipocytokine involved in the regulation of food intake, energy storage and saccharide and lipid metabolism. Impaired regulation of food intake in consequence of leptin resistance

is presented in connection with the aetiopathogenesis of obesity and insulin resistance, but its role in development of these diseases is still not clear (10). Our study revealed a positive correlation between leptin and adiposity. This has been supported by various previous studies (11). Being the product of the ob gene secreted from adipose tissue leptin signals the amount of energy stores to the brain and is implicated in the regulation of food intake and energy balance. Hyperleptinemia in obesity may be due to leptin resistance, which may arise from impaired leptin transport across the blood-brain barrier (BBB), defects in leptin receptor signaling, and blockades in downstream neuronal circuitries (12). Being supported by all these, our study emphasizes the potential of leptin to be used as a marker for obesity.

Present study also reveals a significant positive correlation among the physical parameters of obesity and degree of insulin resistance (HOMA-IR score). Further the strength of association was found to be highest between abdominal circumference and insulin resistance. This suggests a probable role of obesity mainly the central or visceral type in the pathogenesis of insulin resistance. Finding is corroborative with some previous studies that also found direct correlation between central obesity, insulin resistance and cardiovascular risk factors (13, 14). Regarding the adverse effects of obesity in particular the visceral obesity on glucose metabolism many probable mechanisms have been suggested which include, excessive lipid "supply" by a mechanism currently referred to as "lipotoxicity." When FFA (Free Fatty Acids) is elevated for a prolonged period, they have

a direct effect on insulin action in skeletal muscle tissue and liver, reducing the normal responses to insulin to promote glucose uptake and to suppress hepatic glucose output, respectively. In both of these tissues, FFA increase cellular levels of acyl-CoA derivatives, which lead to an increase in the activity of cellular signaling molecules, termed serine kinases that oppose the normal tyrosine phosphorylation cascade of the insulin receptor. The increased intracellular lipid accumulation that occurs in obese subjects as "ectopic fat", that is, triglyceride stored in the target organs themselves rather than in a benign adipose depot is an important source of intracellular acyl-CoA molecules that can affect normal insulin signal transduction. Other proteins secreted by adipose tissue, including the important inflammatory mediators like interleukin-6 (IL-6) and tumor necrosis factor-(TNF), may have adverse effects on energy metabolism and insulin sensitivity in liver and muscle and play key roles in the development of insulin resistance in obesity. Revolutionary concepts is that adipose tissue is not only a simple reservoir for energy stored as triglycerides but also serves as an active secretory organ, releasing many peptides, complement factors, and cytokines into the circulation called adipocytokines. Most important of this class so far are leptin, adiponectin, resistin etc. Complex interactions between these mediators are proposed to be involved in the causation of insulin resistance. However, any single molecule in this category (adipocytokines) may have insignificant association alone with the level of insulin resistance as we found in our study. Leptin though proved to be an obesity marker did not show any significant correlation with the degree of insulin resistance in type 2 diabetics. Corroborative

with our findings, some of the previous studies also reported that the leptin/adiponectin ratio is a more effective parameter of insulin resistance than adiponectin or leptin alone (15). Also there may be difference in the levels of endogenous leptin secretion between men and women and pre and postmenopausal women irrespective of their BMI (16). As our study included males and females almost in equal proportion and substantial number of females were in premenopausal age group, levels of leptin might get deviated amongst the subjects, irrespective of their HOMA- IR score. There are some other schools of thoughts that are not in opinion that insulin resistance is contributed by leptin. Many of them have shown that exogenous leptin may improve the state of glucose metabolism and insulin sensitivity especially in lypodystrophic subjects (17). Based on all such previous data and our present findings it seems that, though obesity is a definite precondition in insulin resistance, single adipocytokine like leptin may not be solely responsible for its causation.

Conclusion

Our study concluded that leptin is a novel marker for obesity, though its direct association was not established with insulin resistance. Physical parameters especially abdominal circumference was found to have significant positive correlation with insulin resistance. Therefore, it seems that, obesity mainly central type may lead to insulin resistance and thereby diabetes mellitus. Leptin, though an obesity marker, is not significantly correlated with insulin resistance alone, which may point towards the multifactor causation of insulin resistance in type 2 diabetes mellitus.

REFERENCES

1. Ceriello A, Mokdad AH, Bowman BA, et al. The continuing epidemics of obesity and diabetes in the United States. *JAMA* 2001; 286: 1195–1200.
2. Meyre D, Froguel P. ENPP1, the first example of common genetic link between childhood and adult obesity and type 2 diabetes. *Med Sci (Paris)* 2006; 22(3): 308–312.
3. Park J. Obesity. In: Park's textbook of preventive and social medicine, 18th edition. Premnagar, Banarasidas Bhanot Publications. 2005; 316–319.
4. Meigs JB, Larson MG, Fox CS, et al. Association of Oxidative Stress, Insulin Resistance, and Diabetes Risk Phenotypes. *Diabetes Care* 2007; 30: 2529–2535.
5. Lang K, Ratke J. Leptin and Adiponectin: new players in the field of tumor cell and leukocyte migration. *Cell Commun Signal* 2009; 7: 27.
6. Yadav A, Jyoti P, Jain SK, Bhattacharjee J. Association of TNF- α promoter gene G-308A polymorphism with metabolic syndrome, insulin resistance, serum TNF- α and leptin levels in Indian adult women. *Ind J Clin Biochem* 2011; 26(2): 193–199.
7. Gupta V, Gupta A, Jafar T, et al. Plasma leptin in insulin-resistant and insulin-nonresistant coronary artery disease and its association with cardio-metabolic risk factors among Asian Indians. *Cytokine* 2012; 57(1): 32–38.
8. Mahadik SR, Deo SS, Mehtalia SD. Is there a relationship between leptin and insulin sensitivity independent of obesity? A population-based study in the Indian Ocean nation of Mauritius. Mauritius NCD Study Group. *Metab Syndr Relat Disord* 2008; 6(2): 121–129.
9. Doumatey AP, Lashley KS, Huang H, et al. Relationships among Obesity, Inflammation, and Insulin Resistance in African Americans and West Africans. *Obesity* 2010; 18(3): 598–603.
10. Lichnovská R, Gwozdziwiczová S, Chlup R, et al. Serum leptin in the development of insulin resistance and other disorders in the metabolic syndrome. *Biomed Papers* 2005; 149(1): 119–126.
11. Satoh N, Naruse M, Usui T et al. Leptin-to-Adiponectin Ratio as a Potential Atherogenic Index in Obese Type 2 Diabetic Patients. *Diabetes Care* 2004; 27(10): 2488–2490.
12. Banks WA, Coon AB, Robinson SM, et al. Triglycerides induce leptin resistance at the blood-brain barrier. *Diabetes* 2004; 53(5): 1253–1260.
13. Klaus JR, Hurwitz BE, Llabre MM, et al. Central obesity and insulin resistance in the cardiometabolic syndrome: pathways to preclinical cardiovascular structure and function. *J Cardiometab Synd* 2009; 4(2): 63–71.
14. McKeigue PM, Shah B, Marmot MG. Relation of central obesity and insulin resistance with high diabetes prevalence and cardiovascular risk in South Asians. *Lancet* 1991; 337(8738): 382–388.
15. Seung-Hyun Ko. The Adiponectin/Leptin Ratio and Metabolic Syndrome in Healthy Korean Adult Males. *Korean Diabetes J* 2010; 34(4): 220–221.
16. Soodini G, Hamdy O. Adiponectin and Leptin in Relation to Insulin Sensitivity. *Met synd rel dis* 2004; 2(2): 114–123.
17. Oral EA, Burant C. Leptin and insulin resistance: good, bad, or still unclear? *AJP Endo* 2009; 296(2): E394–E395.