JMSCR Volume||2||Issue||5||Pages 1176-1186||May 2014 2014

www.jmscr.igmpublication.org Impact Factor-1.1147
ISSN (e)-2347-176x

IGM Publication Journal Of Medical Science And Clinical Research
An Official Publication Of IGM Publication

Association of Insulin Resistance and Selected Adipokines in Cardiovascular Disease with and without Metabolic Syndrome

Authors

Savita Rathore[1], Anil Bidwai[2]

[1, 2] Department of Biochemistry, Index Medical College Hospital and Research Centre, Indore Madhya Pradesh, India

[1]Department of Biochemistry, Mahatma Gandhi Memorial Medical College, Indore Madhya Pradesh, India

Corresponding Author

Savita Rathore

Department of Biochemistry, Mahatma Gandhi Memorial Medical College, Indore, Madhya Pradesh, India

E-mail: *savi_gmcbpl@rediffmail.com*

Abstract

Adipose tissue produces several bioactive mediators' adipokines which influence insulin resistance, diabetes, body-weight homeostasis and inflammation. Plasma adiponectin and leptin are closely related to insulin sensitivity. In present study we measured adipokines, lipid profile, and insulin resistance in CVD patients with and without MetS. 100 patients with cardiovascular disease and 50 healthy persons as control subjects were included. Adiponectin, leptin, lipid profiles, glucose, and insulin levels were measured and insulin resistance was computed by the HOMA. Patients (n=100) and control subjects (n=50) were matched for age, BMI, WC and blood pressure category systolic and diastolic, all were (P<0.0001) highly significant. CVD

patients with and without MetS had highly elevated TC, LDL-C, VLDL-C cholesterol and TG and lower HDL-C level than the control subjects, all were P<0.0001. Glucose, insulin and insulin resistance were also significantly increased in both patient groups than control subjects. Leptin level was higher and adiponectin was significantly lower in patients with MetS and without Mets compared with control subjects, (P<0.0001). We summarized that the measurement of adiponectin and leptin may have a beneficial in the treatment and prevention of cardiovascular diseases in the future.

Key words: cardiovascular disease, metabolic syndrome, adiponectin, leptin, insulin resistance, lipid profile

INTRODUCTION

Adipocytes respond to metabolic and inflammatory stimuli by secreting a variety of molecules known as adipokines [1], including leptin, that are thought to modulate atherosclerosis and are candidate risk factors for CVD. Obesity is associated with a marked increase in circulating leptin concentrations. However, plasma leptin displays a strong association with cardiovascular risk factors, including insulin resistance, metabolic syndrome, and inflammatory markers, even after controlling for measures of body fat mass [2]. Leptin affects lipid metabolism, regulates food intake, modulates taste perception and the feeling of satisfaction after consumption, stimulates the sympathetic nervous system and regulates the metabolism of insulin, glucose and triglycerides [3]. Adiponectin has multidirectional biological action. It inhibits hepatic gluconeogenesis, reduces hepatic glucose output and decreases the level of free fatty acids as a result of their oxidation [4]. Moreover, it was shown that this adipokine has complex anti-atherogenic and anti-inflammatory activity. Unlike other adipokines, plasma concentration of adiponectin is inversely correlated with body mass [4]. In present study we measured the lipid profile, circulating leptin, adiponectin, and insulin resistance concentrations in CVD patients with and without MetS.

MATERIAL AND METHODS

A cohort population based study, total no (n=100) of patients with known history of cardiovascular disease, who attend OPD of MY Hospital, includes into the study. Exclusion criteria are: renal failure, liver disorder and presence of any acute inflammatory conditions that includes infections, trauma or fever. Persons who do not have any abnormal medical history, include as control subjects (n=50). After obtaining informed consent, a questionnaire that contained demographic characteristics including age, sex, cardiovascular risk factors and drug history was completed. Patients weight, height, waist are BP measured and recorded. After 12 hours fasting blood samples collected in the Clinical Biochemistry laboratory are used for in vitro biochemical analysis. The samples are collected by standard procedures under aseptic conditions. Standard procedures are followed for the

preservation and storage of samples before analysis. Adiponectin, leptin and insulin levels are measured using a commercially available ELISA Kit (Ray Biotech, Inc.). The lipid profiles and plasma glucose aere measured with enzymatic and using colorimetric methods. The degree of insulin resistance is estimated by the homeostasis model assessment (HOMA) index, which is computed using the formula: FBS (mmol/L) × serum insulin (μU/ mL)/22.5. Then, the correlation within each groups as well as the other variables are evaluated.

STATISTICAL ANALYSIS

Data analysis was performed using the XLSTAT 2014 program with a value of $p<0.05$ considered significant. One-way analysis of variance (ANOVA) for repeated measures within a group was measured. Comparison of two groups was done by the Student's paired t-test. Results are expressed as MEAN SD or as proportion (%). Pearson correlation coefficient was used to assess the correlation between serum adiponectin and leptin levels with other variables. Finally, linear regression analysis was applied for determination of factors predictive of serum adiponectin levels.

RESULTS

Patients (n=100) and control subjects (n=50) were matched for age (mean SD=54.62± 9.28 vs. 42.22 ± 10.59 years), BMI (mean SD=26.728 ± 2.43vs. 21.062 ± 3.416 kg/m2), WC (mean SD=93.01 ±6.313 vs. 81.17 ±6.76 cm) and blood pressure category systolic and diastolic (mean SD= 136.72 ±20.70 vs. 118.34 ±2.92, 86.11± 7.98 vs.79.60± 1.72 mmHg), all $P<0.0001$, respectively.

To identify cases of metabolic syndrome the ATP III (Adult Treatment Panel III) criteria were used i.e. the presence of three or more of the following characteristics: hyperglycemia, hypertension, low plasma HDL-C level, high plasma triglyceride level and central adiposity.

This allowed classification of the total (n=100) patients with CVD into two groups. CVD with MetS (n=56) patients (56%) and CVD without MetS (n=44) patients (44%). We found major differences between CVD with and without MetS subjects.

CVD patients with and without MetS had highly elevated mean TC, LDL, VLDL cholesterol and TG levels and lower HDL-C levels than the control subjects, all were ($P<0.0001$) significantly (Table 1) so. Mean glucose, insulin and insulin resistances were also significantly increased in both patient groups than in control subjects. Mean adiponectin was significantly lower while leptin and leptin/adiponectin ratio higher in both patient groups compared to control subjects.

Table 2 summarizes the linear correlation of serum adiponectin and leptin concentration between various metabolic parameters by fasting status. Serum adiponectin showed the inverse relationship with all variables except HDL ($r=0.038$, $P=0.711$) cholesterol and serum leptin found the linear correlation with some of variables and no correlation was found with age ($r=0.082$, $P=0.427$), triglyceride ($r=0.157$, $P=0.119$), VLDL

cholesterol (r=0.157, P=0.119) and HDL cholesterol (r= -0.110, P=0.277) while inverse correlation was found with adiponectin (r= -0.548).

Figure 1 & 2 summarizes the linear regression plots of total CVD (n=100) patients BMI and HOMA IR with serum leptin, leptin/adiponectin ratio and adiponectin. Serum adiponectin showed the inverse regression plot with BMI and insulin resistance while serum leptin and leptin/adiponectin ratio showed the linear regression plots.

Table 1: Anthropometric Measurements, Fasting Biochemical Variables and Clinical Characteristics of the Study Groups

	CVD with MetS (n=56) Subjects	CVD without MetS (n=44) Subjects	Control (n=50) Subjects
Age (years)	56.80±8.64	51.84±9.43§	42.22±10.59
BMI (Kg/M^2)	27.62±2.22	25.58±2.22*	21.06±3.41
WC (cm)	95.11±5.77	90.34±6.01*	81.17±6.76
BPS (mm/Hg)	142.76±21.03	129.02±17.68*	118.34±2.92
BPD (mm/Hg)	88.10±7.96	83.56±7.34§	79.60±1.72
Total Cholesterol (mmol/L)	5.50±0.82	4.97±0.88§	3.74±0.46
HDL Cholesterol (mmol/L)	0.77±0.11	0.80±0.12**	1.26±0.17
LDL Cholesterol (mmol/L)	3.71±0.83	3.24±0.90§	1.86±0.45
VLDL Cholesterol (mmol/L)	1.09±0.17	0.92±0.21§	0.60±0.06
Triglycerides (mmol/L)	2.22±0.38	2.02±0.47§	1.32±0.15
Glucose (mmol/L)	9.42±1.57	6.92±0.72*	5.06±0.52
Insulin (µU/ml)	13.66±0.84	11.08±0.76*	8.92±0.62
HOMA IR	5.73±0.91	3.41±0.43*	2.01±0.30
Adiponectin (µg/ml)	5.95±1.25	8.64±0.99*	12.66±0.39
Leptin (ng/ml)	8.25±0.72	6.36±0.37*	5.09±0.53
LAR	1.44±0.27	0.74±0.10*	0.40±0.04

CVD=Cardiovascular Disease, MetS= Metabolic Syndrome, BMI=Body Mass Index, WC=Waist Circumferences, BPS=Systolic Blood Pressure, BPD=Diastolic Blood Pressure, TC=Total Cholesterol, TG=Triglyceride, HDL-C=High Density Lipoprotein Cholesterol, LDL-C=Low Density Lipoprotein Cholesterol, VLDL-C=Very Low Density Lipoprotein Cholesterol, IR=Insulin Resistance, LAR= Leptin Adiponectin Ratio. Comparison between CVD with MetS and without MetS P<0.0001*, P<0.01§, P>0.05**

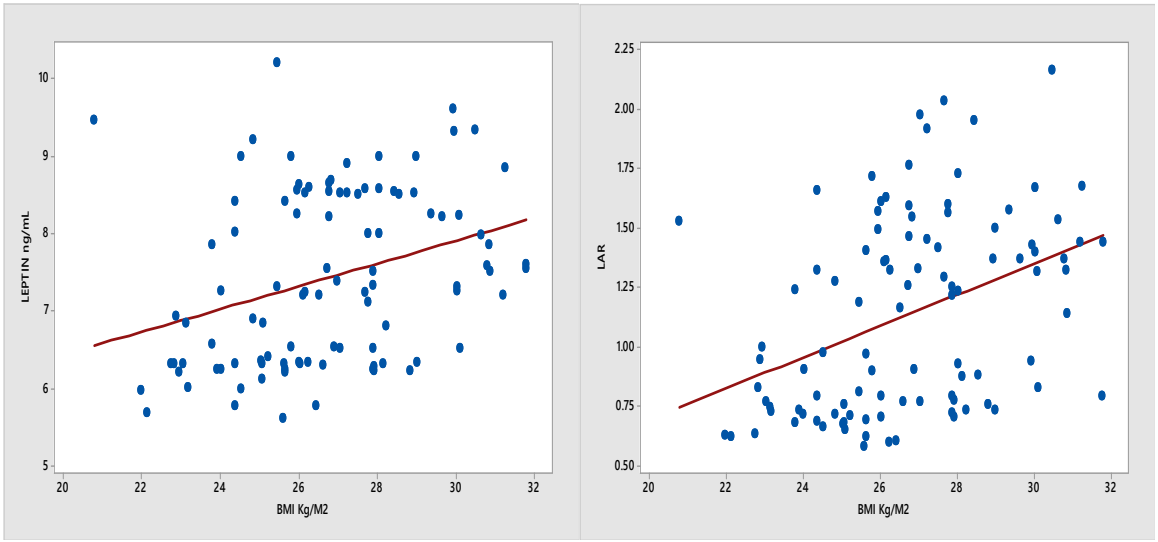
Table 2: Pearson Correlation between Adiponectin, Leptin and Metabolic Parameters By Fasting Status in Total Study Group (n=100)

	Correlation Coefficient R with Adiponectin	Correlation Coefficient R with Leptin
Age (years)	-0.260**	-
BMI (Kg/M^2)	-0.370*	0.321**
WC (cm)	-0.329†	0.310**
BPS (mm/Hg)	-0.292**	0.283**
BPD (mm/Hg)	-0.311**	0.193†
Total Cholesterol (mmol/L)	-0.229†	0.259**
LDL Cholesterol (mmol/L)	-0.180†	0.228†
VLDL Cholesterol (mmol/L)	-0.235†	-
Triglycerides (mmol/L)	-0.235†	-
Glucose (mmol/L)	-0.506*	0.648*
Insulin (µU/ml)	-0.666*	0.703*
HOMA IR	-0.624*	0.741*
Leptin (ng/ml)	-0.548*	-
Adiponectin (µg/ml)	-	-0.548*
LAR	-0.922*	0.781*

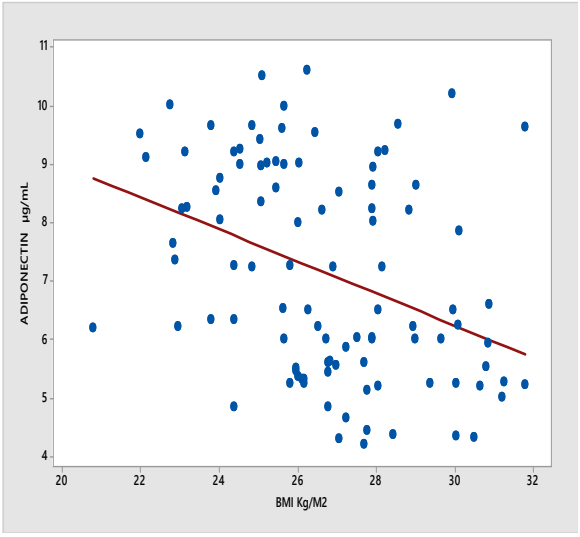
BMI=Body Mass Index, WC=Waist Circumferences, BPS=Systolic Blood Pressure, BPD=Diastolic Blood Pressure, TC=Total Cholesterol, TG=Triglyceride, LDL-C=Low Density Lipoprotein Cholesterol, VLDL-

C=Very Low Density Lipoprotein Cholesterol, IR=Insulin Resistance, LAR= Leptin Adiponectin Ratio, P<0.0001*, P<0.001**, P<0.05†

Figure 1: Linear Regression Plot of Total CVD (N=100) Patients BMI with Leptin, LAR and Adiponectin



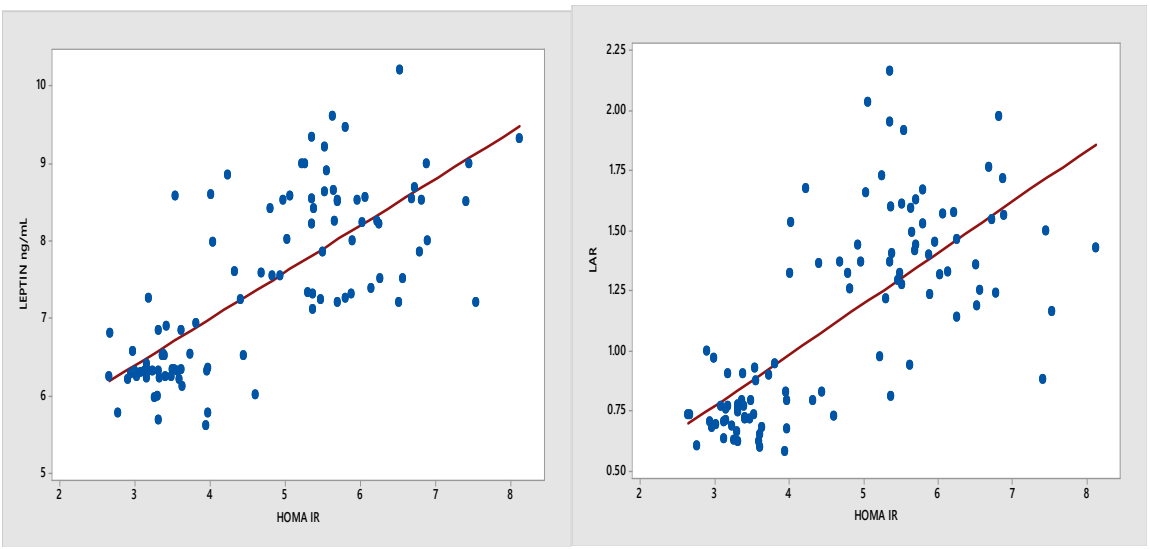
(a) (b)



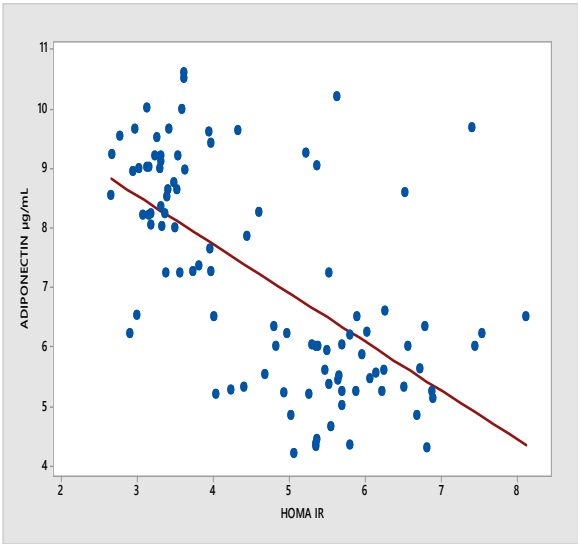
(c)

Figure 1 Linear regression plots of patients with CVD (n=100) displaying significant positive correlations between BMI and (a) leptin (r=0.321; p<0.001), (b) leptin adiponectin ratio (r=0.391; p<0.0001), and (c) adiponectin (r= -0.370; P<0.0001) inverse relationship.

Figure 2: Linear Regression Plot of Total CVD (N=100) Patients HOMA IR with Leptin, LAR and Adiponectin



(a) (b)



(c)

Figure 2 Linear regression plots of patients with CVD (n=100) displaying significant positive correlations between HOMA IR and (a) leptin (r=0.741; p<0.0001), (b) leptin adiponectin ratio (r=0.714; p<0.0001), and (c) adiponectin (r= -0.624; P<0.0001) inverse relationship.

DISCUSSION

Disturbed metabolism of fat tissue is the starting point in the discussion on the role of adipokines in the pathogenesis of cardiovascular disease. Differences in sophisticated biochemical structure of the fat tissue between healthy subjects and patients suffering from CVD are seen when levels of circulating leptin, adiponectin, insulin sensitivity and lipid profile are investigated. In the present study including well-characterized 100 stable CVD participants and 50 controls, we observed that adiponectin level was lower in CVD participants compared with controls in unadjusted analysis. Contrary to a previous study of Kumada et al, [5] reported that their result was no more significant after extensive adjustment for confounding factors but was in agreement with other previous study [6]. Our results showed a highly (table 2) significant negative correlation for adiponectin and positive for leptin with various metabolic parameter. Meanwhile, leptin was positively and adiponectin was negatively correlated with BMI (figure 1). This agreement with previous studies in Japanese individual had shown that the adiponectin concentration was negatively correlated with body mass index, while contrary to positive associations between adiponectin and metabolic parameters (total cholesterol, HDL cholesterol) were observed. Accordingly, it was lower in obese subjects than in lean subjects [7]. Similar conversely, serum adiponectin was inversely correlated with body mass index, waist circumference, serum insulin, fasting glucose, serum triglycerides, and HOMA-IR. The relationships of adiponectin with various metabolic factors reflect the classical association between low adiponectin and metabolic syndrome [7]. Inoue et al. reported that adiponectin and leptin levels tend to correlated with BMI, TG and HDL in an opposite manner [8].

In present study showed the leptin levels were increased in CVD patients with and without (table1) MetS than control group. The same finding was reported by Wallace et al., that the leptin levels were increased in patients with the metabolic syndrome and in those with cardiovascular disease [7]. Leptin stimulates lipoprotein lipase secretion in cultured human and murine macrophages. Leptin increases accumulation of cholesterol esters in foam cells, especially at high glucose concentrations. However, under normoglycemic conditions leptin may protect macrophages from cholesterol overload [9]. Several studies demonstrate an inverse relationship between leptin and high-density lipoprotein (HDL) cholesterol and/or apolipoprotein A-I in humans. Leptin promotes hepatic HDL clearance by up regulating scavenger receptor type B1 and decreases plasma HDL level in mice [10]. Thus, in the context of hyperglycemia, leptin may impair cholesterol removal from peripheral tissues by lowering HDL and unfavorably affect local cholesterol balance in diabetic patients.

Another remarkable finding was adiponectin significantly decreased in CVD patients with and

without (table1) MetS than control group. In previous studies reported the adiponectin levels were low in patients with metabolic syndrome and in those with type 2 diabetes mellitus, suggesting that hypoadiponectinaemia contributes to the pathogenesis of these conditions [11] & [12]. Adiponectin is a collagen-like protein produced specifically by adipose tissue and is abundantly present in the circulation. When the vascular endothelium is injured, adiponectin accumulates in the sub-intimal space of the arterial wall through its interaction with collagens in the vascular intima [13]. Adiponectin attenuates TNF-a –induced expression of adhesion molecules in endothelial cells [12], which is an initial step of atherosclerosis. This finding was also similar with the Kikuko Hotta et al. [14].

One of the interesting results was found insulin resistance relationship with the measured adipokines in the present study groups. Insulin resistance is a major risk factor for the development of atherosclerosis and it also affects plasma levels of adiponectin, which become gradually decreased with increasing insulin resistance [15]. Although adiponectin exerts potent insulin-sensitizing actions in experimental in vivo models, it remains to be clarified whether the inverse association between adiponectin and insulin sensitivity is a cause-effect relationship [15]. Insulin regulates the secretion of various proteins from adipose tissue. Elevated plasma insulin in the diabetic subjects in this study may have been responsible for the decreased plasma adiponectin concentrations. The plasma level of leptin, another molecule specifically secreted from adipocytes, was positively correlated with fasting plasma insulin [16], On the other hand, the plasma adiponectin concentration was negatively correlated with the fasting plasma insulin level. Leptin secretion by adipocytes is stimulated by insulin, and plasma leptin significantly correlates with plasma insulin. By contrast, under some conditions, leptin negatively regulates insulin signaling and glucose uptake [17].

Data from recent studies suggested that leptin/adiponectin ratio can serve as a clinical marker of atherosclerosis both in non-diabetic and diabetic subjects. In this study we obtained that leptin/adiponectin ratio is a dependent predictor in CVD subjects. It showed the linear correlation with BMI and HOMA IR. This is agreement by Finucane et al. [18], who showed in 2097 non-diabetic subjects, 890 men and 1207 women, that the leptin/adiponectin ratio is a useful measure of insulin resistance. Zaletel et al., and Oda et al., suggested that leptin/adiponectin ratio is a more effective parameter of insulin resistance than adiponectin, leptin and HOMA-IR in both non-diabetic and type 2 diabetic patients.

CONCLUSION

In conclusion, the utility of plasma leptin levels in predicting progression of cardiovascular events needs to be examined in additional and the measurement of serum adiponectin level might represent a novel diagnostic tool to stratify patients at risk for CVD and to identify those

patients who would benefit most from preventive strategies. Serum adiponectin level should be considered in the laboratory work-up of CVD patients. Our data provide further support for a link between adipocyte function and/or mass (rather than total BMI) and cardiovascular disease.

ACKNOWLEDGMENTS:

The author acknowledges the Research Center and the subjects who participated in the study.

CONFLICT OF INTERESTS: The author declares that there is no conflict of interests regarding the publication of this paper.

REFERENCES

1) Rajala MW, Scherer PE. Mini-review: the adipocyte at the crossroads of energy homeostasis, inflammation, and atherosclerosis. Endocrinology, 2003;144: 3765–3773.
2) Van Dielen FM, Veer C, Schols AM, Soeters PB, Buurman WA, Greve JW. Increased leptin concentrations correlate with increased concentrations of inflammatory markers in morbidly obese individuals. Int J Obes Relat Metab Disord, 2001;25:1759–1766.
3) Trayhurn P, Bing C, Wood IS. Adipose tissue and adipokines – energy regulation from the human perspective. J Nutrition 2006; 136: 1935S-9S.
4) Abdullah AR, Haydar AH, Raigangar VL, et al. Analysis of the relationship of leptin, high-sensitivity C-reactive protein, adiponectin, insulin and uric acid to metabolic syndrome in lean, overweight and obese young females. Metabolic Syndrome and Related Disorders 2008; 7: 17-22.
5) umada M, Kihara S, Sumitsuji S, Kawamoto T, Matsumoto S, Ouchi N, et al. CAD Study Group. Coronary artery disease. Association of hypoadiponectinemia with coronary artery disease in men. Arterioscler Thromb Vasc Biol. 2003;23:85–89.
6) Luc G, Empana JP, Morange P, Juhan-Vague I, Arveiler D, Ferrieres J,et al. Adipocytokines and the risk of coronary heart disease in healthy middle aged men: the PRIME Study. Int J Obes (Lond). 2010;34:118–126.
7) Wallace, A.M., McMahon, A.D., Packard, C.J., Kelly, A., Shepherd, J., Gaw, A., et al. Plasma leptin and the risk of cardiovascular disease in the West of Scotland Coronary Prevention Study (WOSCOPS). Circulation, 2001;104: 3052–3056.
8) Soebijanto N, Waspadji S. Adiponectin levels and its role in insulin resistance among adult women with metabolic syndrome. Acta Med Indones, 2010; 42(4):187-91.

9) O'Rourke L, Gronning LM, Yeaman SJ, Shepherd PR. Glucose- dependent regulation of cholesterol ester metabolism in macrophages by insulin and leptin. J Biol Chem. 2002;277: 42557–42562.
10) Lundasen T, Liao W, Angelin B, Rudling M. Leptin induces the hepatic high density lipoprotein receptor scavenger receptor B type I (SR-BI) but not cholesterol 7-hydroxylase (Cyp7a1) in leptin deficient (ob/ob) mice. J Biol Chem. 2003;278: 43224–43228.
11) Kershaw, E.E. & Flier, J.S. Adipose tissue as an endocrine organ. Journal of Clinical Endocrinology and Metabolism, 2004;89: 2548–2556.
12) Ouchi N, Kihara S, Arita Y, Maeda K, Kuriyama H, Okamoto Y, et al. Novel modulator for endothelial adhesion molecules: adipocyte-derived plasma protein, adiponectin. Circulation. 1999; 100:2473–2476.
13) Okamoto Y, Arita Y, Nishida M, Muraguchi M, Ouchi N, Takahashi M, et al. An adipocyte-derived plasma protein, adiponectin, adheres to injured vascular walls. Horm Metab Res. 2000; 32:47–50.
14) Kikuko Hotta, Tohru Funahashi, Yukio Arita, Masahiko Takahashi, Morihiro Matsuda, et al. Plasma Concentrations of a Novel, Adipose-Specific Protein, Adiponectin, in Type 2 Diabetic Patients. Arterioscler Thromb Vasc Biol. 2000;20:1595-1599.
15) Kadowaki T, Yamauchi T. Adiponectin and adiponectin receptors. En docr Rev 2005;26:439–451.
16) Caro JF, Sinha MK, Kolaczynski JW, Zhang PL, Considine RV. Leptin: the tale of an obesity gene. Diabetes. 1996;45:1455–1462.
17) Hennige AM, Stefan N, Kapp K, Lehmann R, Weigert C, Beck A, Moeschel K, et al. Leptin down- regulates insulin action through phosphorylation of serine-318 in insulin receptor substrate 1. FASEB J. 2006;20: 1206–1208.
18) Finucane FM, Luan J, Warenham NJ, Sharp SJ, O`Rahilly S, Balkau B, et al. Correlation of the leptin/adiponectin ratio with measures of insulin resistance in non-diabetic individuals. Diabetologia, 2009; 52:2345-2349.