

## Comparison of Perilipin Protein Levels in Obese with Metabolic Syndrome and Obesity Non Metabolic Syndrome

Rusdiana<sup>1\*</sup>, Maya savira<sup>2</sup>, Widjaja Sry<sup>1</sup>

<sup>1</sup>Departement of Biochemistr ,Faculty of Medical Universitas Sumatera Utara, North Sumatera Medan, Indonesia

<sup>2</sup>Departement of Physiology, Faculty of Medical Universitas Sumatera Utara, North Sumatera Medan, Indonesia

**Abstract :** Perilipin is a highly phosphorylated adipocyte protein that is localized on the surface of the lipid droplet, that has a role in controlling access to the lipid lipolytic enzymes and played a role in setting the stroge and mobilization of trigliseride in the adipocytes. Because of the potential importance of adipocyte lipolysis to obesity and increasing perilipin protein in obesity this study aimed to analyze the comparison of protein perilipin levels in obese with metabolic syndrome and obese non metabolic syndrome. The sample population is obese adults, then we examined the weight, height, waist size, blood pressure, laboratory tests such as blood sugar levels and lipid profile of sample population to separate obese with metabolic syndrome and obese non metabolic syndrome. After we determined each group we measured perilipin protein levels in blood in obese with metabolic syndrome and obese non metabolic syndrome by Enzyme Link Immunosorbent Assay (ELISA) method. With statistical analysis using T test found that there was significant difference of perilipin protein levels between obese with metabolic syndrome and obese without metabolic syndrome ( $p < 0.005$ ). Perilipin protein levels was higher in obese with metabolic syndrome than obese non metabolic syndrome.

**Keywords :** Obesity, metabolic syndrome, perilipin, trigliseride.

### Introduction

The incidence of obesity increase rapidly as a result of inactive lifestyle. The energy thst used for daily activity decrease parallels along with advances in technology. Based on WHO data there are 1.6 billion adults with overweight and 400 nmillion among them are obese<sup>1</sup>. Based on research data Riset Kesehatan Dasar in 2007<sup>2</sup>, obesity prevalence in Indonesia generally in the population aged  $\geq 15$  years old is 10.3% (men 13.9% and women 23.8%)<sup>3</sup>. An individual determined as obese based on Body Mass Index (BMI) it is a simple index of weight–height relationship calculated as weight in (kg) divided by height in (m) squared. One is categorized as obese I when the BMI 25-29,9 and obese II when the BMI BMI  $>30$  (4). Metabolic syndrome is a condition that characterized by visceral obesity, increasing trygliceride levels and glucose and decreasing *High Density Lipoprotein* (HDL) and hypertension that can cause a greater risk incidence of type 2 DM and cardiovascular deseases.<sup>4,5</sup> Prevalences of metabolic syndrome varies greatly it is caused by uniformity criterias that used to determine, ethnic difference, sex and age. It can be confirmed that metabolic syndrome likely to increase parallels with obesity or central obesity prevalences.<sup>6,7,8</sup>

Obesity is caused of accumulation of fat in adipose tissue. The adipocyte holds the major source stored energy in the body in the form of triacylglycerol (TAG)., it is covered by lipid droplet<sup>9,10,11</sup>. The perilipins are highly phosphorylated adipocyte proteins that are localized at the surface of the lipid droplet.<sup>9,10,11</sup> With activation by protein kinase A, perilipins translocate away from the lipid droplet and allow hormone-sensitive lipase to hydrolyze the adipocyte triglycerides to release nonesterified fatty acids (NEFA) and glycerol from fat tissue can be regulated by a cAMP-mediated process.<sup>12</sup> The perilipin proteins are polyphosphorylated by protein kinase A and phosphorylation is necessary for translocation of HSL to the lipid droplet and enhanced lipolysis<sup>13</sup>. Overexpression of perilipin inhibited adipocytes lipolysis<sup>15,16</sup>. This study aimed to analyze the comparison of protein perilipin levels in obese with metabolic syndrome and obese non metabolic syndrome, in other study about perilipin protein was evidenced that was elevated at obesity.<sup>17,18</sup>

## 1. Method

This study involved 40 obesity subjects, all of whom were in good health. All subjects gave informed consent. Subjects divide two groups, one group is obesity with metabolic syndrome and other group is obesity without metabolic syndrome. Each group consisted of 20 subjects. To determine whether the subject of the metabolic syndrome so examination of weight, height, waist size, blood pressure, laboratory tests such as blood sugar levels and lipid profile. Sample So, two these groups examined the levels of protein perilipin by ELISA method

**Table 1. Baseline characteristic of the 40 samples**

	<b>Obesitas with metabolic syndrome</b>	<b>Obesitas non metabolic syndrome</b>
Age	43.9±11.3	34.55±10.8
BMI	33.86±5.0	31.75±4.0
Waist size	107±10	104±15
FBG	101.85± 50.8	88.49±7.2
HDL	63.2±23.85	46.05 ±6.99
Trig	193.15±88.59	91.9±32.81
Sistole	139.85 ± 16.3	123 ± 155
Diastole	87± 8.4	81.3 ±9.1
Perilipin	35.07±29.84	29.35±20.95

## Statistical Analysis

The data were statically analyzed, using the statistical analysis with T test, with T Test we found  $p < 0.005$ , it means that there was significant difference of perilipin protein levels between obese with metabolic syndrome and obese without metabolic syndrome.

**Table 2 One-Sample Test**

	<b>Test Value = 0</b>					
	<b>T</b>	<b>Df</b>	<b>Sig.(2-tailed)</b>	<b>Mean Difference</b>	<b>95% Confidence Interval of the Difference</b>	
					<b>Lower</b>	<b>Upper</b>
Perilipin SM	5.255	19	.000	35.06685	21.1013	49.0324
Perilipin Non SM	6.264	19	.000	29.34848	19.5417	39.1552

$P < 0.005$

## Results and Discussions

The characteristics of the subjects of this study are shown in Table 1. Subjects in this study were not 20 years old . Body Mass Index (BMI) in the sample used in both obesity with metabolic syndrome and

obesity non metabolic syndrome is  $>27$ , in this study the waist size of the samples at the obesity with metabolic syndrome found from 89- 119 cm and waist size of the obesity non metabolic syndrome found 97- 117cm, Fasting Blood Glucose (FBG) in the obesity non metabolic syndrome samples were normal but the obesity with metabolic syndrome range from low to the moderately elevated range. Profile lipid like HDL value of the samples of obesity with metabolic syndrome range 34-60 mg/dL and HDL value of the samples of obesity non metabolic syndrome range 46-162 mg/dL. Triglyceride value of the samples of obesity with metabolic syndrome range 91-452 mg/dL and obesity non metabolic syndrome was 46-162mg/dL. Dividing sample group in to obesity with metabolic syndrome and obesity non metabolic syndrome base on 3 criteria from 5 criteria, that are LP  $>102$  cm at male and  $>88$  at female, triglyceride levels  $\geq 150$ mg/dL, HDL  $<40$ mg/dL at male and  $<50$  mg/dL at female, Blood Pressure (BP)  $\geq 130/85$  Hg, so can be categorized as obesity with metabolic syndrome or obesity non metabolic syndrome. Perilipin protein in serum was measured by ELISA method. Allow samples to clot for 2 hours at room temperature or overnight at  $4^{\circ}\text{C}$  before centrifugation for 20 minutes at approximately 1000xg. Collect the supernatant and carry out the assay immediately. Microplate reader with wavelength 450nm. The result of measuring perilipin protein was found the lower value at obesity with metabolic syndrome was 5.52 ng/ml and the highest value at obesity with metabolic syndrome was 150.24 ng/ml and the value at obesity non metabolic syndrome was 4.76 ng/ml and the highest was 85.77 ng/ml. This study aimed to analyze the comparison of protein perilipin levels in obese with metabolic syndrome and obese non metabolic syndrome, so we used the statistical analysis with T test found that there was significant difference of perilipin protein levels between obese with metabolic syndrome and obese without metabolic syndrome ( $p<0.005$ ). This study protein perilipin value was highest at obesity with metabolic syndrome than obesity no metabolic syndrome. Normal value perilipin protein range 0.156-10 ng/ml. This study protein perilipin value was highest at obesity with metabolic syndrome than obesity no metabolic syndrome. Normal value perilipin protein range 0.156-10 ng/ml. The study was done by Philip and friends found increasing perilipin protein at obesity and increasing perilipin, so decrease lypolysis<sup>18</sup>. As we know that perilipin protein is the function of inhibit the lypolysis process. It is be evidenced that perilipin knockout mice so basal adipocyte lipolysis was increased, resulting in a lean mouse<sup>19,20,21,22</sup>. The other research that finding low concentration of perilipin is related with high basal and catecholamin that induction of lypolysis rate in the cell<sup>21</sup>. So many studies prove that perilipin protein have the function of inhibited the lypolysis process and it is be evidenced with increasing perilipin protein at obese<sup>24,25</sup> and that this study we found that concentration of perilipin protein was highest at obese with metabolic syndrome than obese non metabolic syndrome. However, further research needs to be investigated whether there is a correlation perilipin protein with the occurrence of metabolic syndrome

## Acknowledgments

The authors gratefully acknowledge that the present research is supported by Ministry of Research and Technology and Higher Education Republic Indonesia. The support is under the research grant TALENTA USU of Year 2016.

## References

1. World Health Organization. Obesity and overweight fact sheet.IOTF report. 2015. Available at <http://www.who.int/mediacentre/factsheets/fs311/en/>. Accessed 10 February 2015.
2. Riskesdas 2013. Riset kesehatan dasar
3. Departemen Kesehatan RI, 2009
4. World Health Organization. The Asia Pacific perspective.Redefining obesity and its treatment.World Health Organization. International Association for the study of Obesity and International Obesity Task Force. Melbourne: International Diabetes Institute; 2000.
5. Stern M, Williams K, Gonzalez-Villalpando C et al. Does the metabolic syndrome improve identification of individuals at risk of type 2 diabetes and/or cardiovascular disease. *Diabetes Care* 2004;27(11):2676-81.
6. Sargowo D, Andarini S. Pengaruh komposisi asupan makanan terhadap komponen sindrom metabolik. *J Kardiologi Indones*. 2011; 32:14-23

7. Carr DB, Utzschneider KM, Hull RL et al. Intra-abdominal fat is a major determinant of the National Cholesterol Education Program Adult Treatment Panel III criteria for the metabolic syndrome. *Diabetes* 2004;53(8):2087-94
8. Puspardini. *Obesitas Sentral, Sindroma Metabolik dan Diabetes Melitus tipe 2*. Universa Medicina. 2007; 28(4):195-204
9. Holm, C., Langin, D., Manganiello, V., Belfrage, P., and Degerman, E. (1997) Regulation of hormone-sensitive lipase activity in adipose tissue. *Methods Enzymol.* 286, 45 – 67.
10. Miyoshi , H. , J. W. Perfield 2nd , M. S. Obin , and A. S. Greenberg . 2008 . Adipose triglyceride lipase regulates basal lipolysis and lipid droplet size in adipocytes. *J. Cell. Biochem.* **105** :1430 – 1436 .
11. Schweiger , M. , R. Schreiber , G. Haemmerle , A. Lass , C. Fledelius , P. Jacobsen , H. Tornqvist , R. Zechner , and R. Zimmermann . 2006 . Adipose triglyceride lipase and hormone-sensitive lipase are the major enzymes in adipose tissue triacylglycerol catabolism. *J. Biol. Chem.* **281** :40236 – 40241 .
12. Greenberg AS, Egan JJ, Wek SA, Garty NB, Blanchette-Mackie EJ, Londos C 1991 Perilipin, a major hormonally regulated adipocyte specific phosphoprotein associated with the periphery of lipid storage droplets. *J Biol Chem* 266:11341–11346.
13. Egan, J. J., Greenberg, A. S., Chang, M. K., Wek, S. A., Moos, M. C., Jr., and Londos, C. (1992) Mechanism of hormone-stimulated lipolysis in adipocytes: translocation of hormone-sensitive lipase to the lipid storage droplet. *Proc. Natl. Acad. Sci. U.S.A.* 89, 8537 – 8541.
14. Belfrage, P., Fredrikson, G., Olsson, H., and Stralfors, P. (1987) Regulation of Adipose tissue lipolysis through reversible phosphorylation of hormone-sensitive lipase. *Adv. Cyclic Nucleotide Protein Phosphorylation Res.* 17, 351 – 359.
15. Holm, C., Osterlund, T., Laurell, H., and Contreras, J. A. (2000) Molecular mechanisms regulating hormone-sensitive lipase and lipolysis. *Annu. Rev. Nutr.* 20, 365 – 393.
16. Londos C, Gruia-Gray J, Brasaemle DL, Rondinone CM, Takeda T, Dwyer NK, Barber T, Kimmel AR, Blanchette-Mackie EJ 1996 Perilipin: possible roles in structure and metabolism of intracellular neutral lipids in adipocytes and steroidogenic cells. *Int J Obes Relat Metab Disord* 20(Suppl 3):97–101.
17. Miyoshi , H. , S. C. Souza , H. H. Zhang , K. J. Strissel , M. A. Christoffolete , J. Kovsan , A. Rudich , F. B. Kraemer , A. C. Bianco , M. S. Obin , et al . 2006 . Perilipin promotes hormone-sensitive lipase-mediated adipocyte lipolysis via phosphorylation-dependent and -independent mechanisms. *J. Biol. Chem.* 281 :15837 – 15844 .
18. PHILIP A. KERN, GINA DI GREGORIO, TONG LU, NEGAH RASSOULI, AND GOURI RANGANATHAN Central Arkansas Veterans Healthcare System and Department of Medicine, Division of Endocrinology, University of Arkansas for Medical Sciences, Little Rock, Arkansas 72205. *The Journal of Clinical Endocrinology & Metabolism* 89(3):1352–1358.
19. S.Mottagui-Tabar, M.Ryden, P.Lofgren, G.Faulds, J.Hoffstedt, A.J. Brookes, I.Anderson, P.Arner.Department of Medicine and Center for Genomics and Bioinformatics (SMT and AJB), Karolinska Institutet, Stockholm,Sweden. *Diabetologia* 2003) 46:789-797.
20. Souza SC, Muliro KV, Liscum L, Lien P, Yamamoto MT, Schaffer JE, Dallal GE, Wang X, Kraemer FB, Obin M, Greenberg AS 2002 Modulation of hormone-sensitive lipase and protein kinase A-mediated lipolysis by perilipin A in an adenoviral reconstituted system. *J Biol Chem* 277:8267–8272
21. Martinez-Botas J, Anderson JB, Tessier D, Lapillonne A, Chang BH, Quast leanness and reverses obesity in *Lepr(db/db)* mice. *Nat Genet* 26:474–479
22. Tansey JT, Sztalryd C, Gruia-Gray J, Roush DL, Zee JV, Gavrilova O, Reitman ML, Deng CX, Li C, Kimmel AR, Londos C 2001 Perilipin ablation results in a lean mouse with aberrant adipocyte lipolysis, enhanced leptin production, and resistance to diet-induced obesity. *Proc Natl Acad Sci USA* 98:6494–6499
23. Clifford GM, Londos C, Kraemer FB, Vernon RG, Yeaman SJ 2000 Translocation of hormone-sensitive lipase and perilipin upon lipolytic stimulation of rat adipocytes. *J Biol Chem* 275:5011–5015.
24. YH, Werber Y, Greenberg AS, Fried SK 2003 Perilipin expression in human adipose tissues: Effects of severe obesity, gender, and depot. *Obes Res* 11:930 –936.
25. Wang Y, Sullivan S, Trujillo M, Lee M-J, Schneider S, Brodin RE, Kang YH, Werber Y, Greenberg AS, Fried SK 2003 Perilipin expression in human adipose tissues: Effects of severe obesity, gender, and depot. *Obes Res* 11:930 –936

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