RESEARCH ARTICLE
URL of this article: http://heanoti.com/index.php/hn/article/view/hn1419

The Role of Nutritional Status, Dietary Pattern and Free Radical in the Insulin Resistance Development

Meirna Dewita Sari*, Bambang Wirjatmadi*, Merryana Adriani*
*Department of Nutrition, Faculty of Public Health, Airlangga University, Indonesia
Email: meirnadewitasari@gmail.com

ABSTRACT

Insulin resistance is an early sign of diabetes type 2 that occurs in situations where target organ sensitivity decreases against insulin. Excess free radicals can be mutually insulin resistance. The purpose of this study was to determine how the effects of insulin can be a determining factor. This study used case control design conducted on 22 outpatients at Clinic University of Muhammadiyah Surabaya during June until July 2017. Data were collected using questionnaires, nutritional status, fasting blood glucose, fasting insulin, and MDA levels. Insulin resistance status using normal HOMA IR method of <2.77. Logistic Regression was used to determine the effect of free radical on the occurrence of insulin resistance. The results showed free radical against insulin resistance at α=0.05 with p=0.018. That result means boosting MDA levels also increases the risk of insulin resistance. In addition, the consumption of dietary sources of fat and excessive use of food additives, low fruit intake, BMI values are also factors affecting insulin resistance with p=0.027; p=0.036; and p=0.027. Given the elevated levels of MDA-free radicals can increase the risk of insulin resistance. Applying a healthy lifestyle by consuming fiber-rich foods can prevent the formation of excessive free radicals.

Keywords: Insulin resistance, Free radical, HOMA IR

INTRODUCTION

Background

Diabetes mellitus is included in non-communicable diseases that are still a public health problem and are very important to be handled immediately. Non-communicable diseases are the biggest cause of death in Indonesia, where an estimated 63% of deaths in Indonesia are caused by non-communicable diseases. Although still dominated by cardiovascular disease, but diabetes ranks third after cancer as the cause of death from non-communicable diseases in Indonesia. Epidemiological transitions and nutritional play an important role in changing these trends tersebut (Soewondo, Ferrario, & Tahapary, 2013).

Prevalence of diabetics mellitus tend to increase from year to year. International Diabetes Federation (IDF) estimates the number of diabetics in the world about 382 million people in 2013 (Kemenkes RI, 2014). Globally, by 2014 about 422 million adults live with diabetes. The number is quite a drastic increase to 108 million adults in 1980s (WHO, 2016). The number is estimated to increase and reach 592 million people in 2035 (Kemenkes RI, 2014).

Diabetes mellitus is a metabolic disorder disease characterized by elevated blood sugar levels and occurs due to destruction of pancreatic β cells so it cannot produce enough insulin or the body cannot use insulin that has produced effectively or both of them. The condition in which the body cannot use insulin effectively is known as insulin resistance (Kemenkes RI, 2014; Tiwari, Pandey, Abidi, & Rizvi, 2013; WHO, 2016). The case of diabetes mellitus is generally dominated by diabetes mellitus type 2 which is motivated by insulin resistance. Insulin resistance is an early of diabetes mellitus. At the beginning, insulin resistance has not shown clinical signs of diabetes in the form of elevated blood sugar levels. This is because β cells are still able to compensate by producing more insulin, then resulting hyperinsulinemia, glucose level still normal or slight increase (Evans, Goldfine, Maddux, & Grodsky, 2003; Merentek, 2006). The condition can be an early detection of the occurrence of diabetes mellitus even 10 years before diabetes mellitus is characterized by hyperglycemia actually occurs (Valko et al., 2007).

Insulin resistance can be caused by various factors including genetic factors and environmental factors that include high caloric food intake, low physical activity, obesity, pregnancy, and the increase of an abnormal

certain hormones (Erejuwa, 2012; Evans et al., 2003). In addition, oxidative stress is also known to play a role in the pathogenesis of insulin resistance that triggers the occurrence of diabetes mellitus (Erejuwa, 2012). But it is unclear what are the main factors affecting the occurrence of insulin resistance and how its role in influencing it.

Purpose

This study aimed to determine the factors that affect insulin resistance. It also aimed to identify the characteristics of respondents, conditions of insulin resistance status, and consumption patterns of respondents.

METHODS

Participant

This research was a case control study with 22 subjects of adults and the aged between 30-65 years who have visited Clinic University of Muhammadiyah Surabaya. The study was conducted in June-July 2017. The subjects consisted of 11 cases with the criteria having HOMA-IR of ≥2.77 and / or fasting blood glucose of >100 mg / dl, fasting blood insulin of >18 μU / mL obtained based on screening of the latest laboratory examination results and a history of the disease of respondents. The samples taken were patients who were diagnosed with diabetes according to the results of the blood sugar examination when more than normal. While subjects in control cases were 11 people with the criteria having HOMA-IR of <2.77 and/or fasting blood glucose of ≤100 mg/dl, fasting blood insulin level of ≤18 μU / mL obtained using simple random sampling technique from patient data have normal blood sugar levels. Characteristics of respondents obtained through interviews using a questionnaire, including age, gender, education level, income level, and family disease history.

Anthropometric and Clinical Measurements

Measurements of weight and height were measured against the applicable standards. Measurement of weight was using the bathroom scale with kg unit, while the height measured was using microtoise with cm units. Body mass index was calculated from body weight (kg) divided by height (m) squared.

Blood samples collected were blood samples after fasting at least 8 hours. The blood sample taken was 7 cc. Fasting plasma glucose levels were measured using hexokinase method. While fasting serum insulin levels were measured using Electro Chemiluminiscence Immuno Assay (ECLIA) method. The taking of plasma glucose and insulin serum was done at Parahita Laboratory Surabaya. HOMA-IR was used to assess insulin resistance (fasting insulin serum (μU/ml) x fasting plasma glucose (mg/dl)/405) (Tang, Li, Song, & Xu, 2015). The cutoff point used in determining insulin resistance status in this study was 2.77, so that respondents were predicted to have insulin resistance (positive insulin resistance group) if the HOMA-IR value was more than 2.77 (Kusumo (2005), Marfianti (2009).

Free radical levels were measured based on MDA levels examined from 1 cc of blood serum using tiobarbituric acid (TBA) test by colorimetric method. Readings of spectrophotometric results with wave length 532 nm (Moselhy, Reid, Yousef, & Boyle, 2013). MDA examination conducted at Nutrition Laboratory of Public Health Faculty of Airlangga University.

Dietary Intake

Dietary pattern was measured using semi-quantitative-FFQ. Questions included food consumption habits of carbohydrate sources, animal protein sources, vegetable protein sources, vegetables, fruits and food sources of fat and food additives. In addition to the frequency of consumption, also obtained the amount of food consumed.

Ethical Considerations

The Health Research Ethics Committee of the Faculty of Public Health Airlangga University approved the study protocol. All patients provided informed consent.

Statistical Analysis

Bivariate analyzes were used to examine differences in the independent variables between case and control groups. Parameters with normal distribution (tested with Kolmogorov Smirnov test) were compared with Student's t-test (age, income level, BMI, fasting blood insulin, HOMA-IR, total food intake, and MDA levels)

and expressed as mean ± SD . Parameters with skewed distribution were analyzed with Mann-Whitney test. Parameters with nominal scale data were analysed with chi-square (gender, level of education) and fisher exact test (family disease history). Bootstrap logistic regression based on 500 samples was used to obtain maximum results in searching for factors of independent variables that influence the occurrence of insulin resistance.

RESULTS

Participants were grouped by insulin resistance status. Participants with HOMA-IR values of ≥2.77 were grouped in case groups, whereas participants with HOMA-IR values of <2.77 were included in the control group. Both groups had different BMI, fasting glucose, fasting insulin, and significant HOMA-IR p-value of < α (0.05). Although it has not significantly different respondents, case groups tend to be older, have higher incomes and have a history of diabetes in their families. Consumption of food sources of carbohydrates, animal and vegetable protein, and food sources of fat was also higher in the group of case groups. It also appeared at higher MDA levels.

Table 1.The Difference of Characteristics, Insulin Resistance Status, Dietary Pattern, and MDA Level Between Positive Insulin Resistance Group and NegativeInsulin Resistance Group

Variable	Case group	Control Group	P-value
Age (years)	45.36±9.24	39.27±8.63	0.126
Sex (male/female)	6 (54.5%) male 5 (45.5%) female	6 (54.5%) male 5 (45.5%) female	0.671
Education level	2 (18.1%) Senior high school 4 (36.4%) Diploma/S1 5 (45.5%) S2/S3	2 (18.1%) Senior high school 4 (36.4%) Diploma/S1 5 (45.5%) S2/S3	0.281
Income level (Rp)	Rp 3.827.272.73	Rp 3.268.181.82	0,.445
Family history (yes/no)	5 (45.5%) Yes 6 (54.5%) No	3 (27.3%) Yes 8 (72.7%) No	0.201
BMI (kg/m^2)	28.69±3.70	24.31±3.22	0.008
Fasting glucose (mg/dl)	140. 82±67.07	87.45±9.34	0.014
Fasting insulin (µU/ml)	13.57±4.39	8.41±2.83	0.004
HOMA-IR	4.21±1.5	1,78±0,47	0.000
Carbohydrate-source food	519.10 ± 162.58	329.76 ± 123.82	0.008
Animal protein-source food	115.4 ± 76.24	83.77 ± 40.11	0.412
Plant protein-source food	207. 04 ± 167.93	204.91 ± 167.03	0.974
Vegetables	44.9 ± 29.25	164.68 ± 63.72	0.001
Fruits	38.10 ± 18.81	83.34 ± 38.21	0.014
Fat -source food	148.06 ± 49.72	94.15 ± 36.36	0.009
MDA level (µU/ml)	2.64±0.39	2.41±0.22	0.116

Incidence of insulin resistance is known to be influenced by several factors, including consumption patterns, body mass index (BMI), and MDA-free radical levels. Based on the results of processing and data analysis using Logistic Regression Test with bootstrap method (re-sampling method) of 500 samples can be obtained equation model to predict the incidence of insulin resistance.

Dependent variables analyzed to determine the factors that influence the occurrence of insulin resistance are insulin resistance status which is divided into two categories: positive insulin resistance and insulin negative resistance. While the independent variables analyzed include consumption patterns (total consumption of food sources of carbohydrates, animal protein sources, vegetable protein sources, vegetables, fruits, and sources of fat and food additives), body mass index (IMT), and MDA-free radical levels.

Model on logistic regression test was obtained by fit model with significance level of p <α (0.05). Based on 'Variable in the Equation and Bootstrap Variable in the Equation' obtained that significant independent variables and influenced the occurrence of insulin resistance was the amount of fruit consumption (OR=2.929, p=0.036), the amount of food consumption of fat source and food aditives (OR=0.544, p=0.027), body mass index (p=0.027), and free radical (p=0.018), and constant (p=0.009). While the variable amount of consumption of food sources of carbohydrates, animal and plant protein, and vegetables.

DISCUSSION

Insulin resistance is a condition in which the required insulin concentration is greater than normal to maintain normal blood sugar levels (Saini, 2010). This happens because the body cells, especially the liver, muscle, and fat/adipose tissue has decreased in sensitivity and even insulin resistance (IDF, 2015). Growing age increases the risk of insulin resistance when accompanied by weight gain and lack of physical activity. It can happen because the weight exceeds normal will cause the accumulation of fat tissue that affects the decrease in insulin sensitivity due to decreased mitochondrial function, especially skeletal muscle.

Men are at higher risk for insulin resistance even though the fat composition in men is lower than women. This is due to the lipogenic ability of female adipocytes, especially adipocytes in the perigonadal depots, which are higher so it is able to decrease visceral fat accumulation. The increase in lipogenic capacity is also known because of the stimulation of insulin signal tissue so that it can increase tissue sensitivity to insulin.

People with high socioeconomic tend to experience prediabetes. People in postgraduate education stages are 1,68 times more to experience insulin resistance (Yang, Hall, Piccolo, Maserejian, & Mckinlay, 2015). Jobs will affect the income received by a person. The income can affect the purchasing power of family and revenue. Yang *et al.* (2015) research was suggesting that people with higher income stages are 1.88 times more to experince insulin resistance and are in prediabetes. The high prevalence of insulin resistance in upper middle socioeconomic groups in developing countries occurs because wealth/prosperity is associated with sedentary lifestyles and consuming of excessive fat and calorie foods.

Groups of people who have a family history of diabetes are at higher risk for experiencing insulin resistance that may occur due to genetic mutations (Stern & Mitchell, 1999). However, environmental factors also play a role in causing insulin resistance that triggers the development of diabetes mellitus.

Based on the results of multivariate analysis in this study obtained the results that the consumption of foods in high fat and low in fiber, especially fruit, BMI, and free radical levels into factors that affect the occurrence of insulin resistance. The results of this study indicated that a person's risk for insulin resistance increased to 1.83 times greater in each increase of the number dietary consumption of fat sources and food additives as much as 100 grams in one day.

The group of positive insulin resistance tended to be more and more often consume food sources of fat especially fried. Similar results in Japanese populations also showed that high-fat western diet (high intake of fried foods, fried foods and fried meat) was positively correlated with HOMA-IR as an indicator of insulin resistance (Arisawa et al., 2014). High calorie food intake leaded to interference in the body's metabolism. Studies conducted in groups with energy consumption exceeding adequacy in healthy individuals suggested the formation of mitochondrial ROS and cytosol, MDA, mitochondrial antioxidant enzymes, and increased levels of expression of SOCS-3 (suppresor of cytokine signaling-3) associated with mediated insulin signal transduction insulin resistance, hyperinsulinemia, to increased insulin resistance (Lim et al., 2011).

Consumption of fiber, especially fruit, can reduce the risk of insulin resistance. The results of this study indicated that fruit consumption has an effect in reducing the risk of insulin resistance. A person who consumes more can prevent insulin resistance 2.9 times more than those who consume less fruit. The adequacy of dietary fiber for children, adolescents and adults is 14 grams of dietary fiber per 1000 kcal of energy adequacy (Hardinsyah, Riyadi, & Napitupulu, 2012). Fruits contain water-soluble food fiber that plays a role in slowing slow glucose absorption thereby lowering blood sugar levels.

Vegetables and fruits are an important source of antioxidants and play a role in reducing the risk of cardiovascular disease including diabetes mellitus type 2. A recent study suggested that people who eat fruits, such as apples, grapes and blueberries at least twice a week can lower the risk of type 2 diabetes by 23 percent (Sekarsari, 2016). Bananas and potatoes contain potassium capable of maintaining tissue cells in the heart and kidneys so that it works better and can prevent the occurrence of insulin resistance that leads to diabetes. Vitamin C (ascorbic acid) contained in apples, oranges, tomatoes and strawberries is known to play effectively capture O2* (superoxide anions) and $_1O_2$ (singlet oxygen), but it can also break the radical reaction produced by per oxidation lipid. Vitamin C as an antioxidant serves to bind O_2 so it does not support oxidation reactions or as oxygen scavenger (Sayuti & Yenrina, 2015). Vitamin A is found in yellow fruits such as oranges and tomatoes. Melons and carrots contain carotenoids (vitamin A precursors). Carotenoids are composed of lycopene, beta carotene, zeaxanthin and cryptoxanthin. Carotenoids serve as a damper of oxygen singlets and free radical deactivators. Lycopene has twice the antioxidant activity stronger than beta carotene.

The weight gain leads to body fat build up that affects the structure changes and function of adipose tissue. Because adipose tissue is not only as a storage place for triglycerides but also produces some hormones called adipokinase. Adipokinase affects the regulation of the action of insulin not only on adipose tissue, but also skeletal muscle and liver which are the main organ target insulin. Derived hormones, which include cytokines (TNF-α and IL-6), leptin, resistin and adiponectin, further affect the occurrence of insulin sensitivity through different mechanisms (Merentek, 2006; Park, 2008).

Fatty acids can cause insulin resistance because they interfere glucose oxidation which is stimulated by insulin and muscle glycogen synthesis resulting from unstable stimulation of glucose / phosphorylation. This is due to a decrease in the ability of insulin to activate the insulin/phosphoinositol 3-kinase subtrate signaling pathway (Karakelides, Irving, Short, Brien, & Nair, 2010). Increased levels of MDA is an indication of oxidative stress. Increased lipid per oxidation will trigger free radical activity which can lead to insulin resistance. Free radical activity that occurs will trigger the activation of stress sensitive pathways that are in line with the occurrence of insulin resistance (Kalaivanam, Dharmalingam, & Marcus, 2006). Obesity and increased central obesity are known to be a major determinant of elevated plasma MDA levels and are directly related to the development of glucolipotoxic states (Moreto, Oliveira, Manda, & Burini, 2014).

Thus the accumulation of exogenous free radical exposure and adipose tissue buildup may activate the inflammatory gene that causes disorders of adipokinase regulation. Increased leptin, resistin, TNF-α and IL-6, and decreased adeponectin activate the serine / threonin pathway thus disrupting the tyrosine normal pathway. Serine phosphorylation decreases PI-3 kinase signal activity which may decrease insulin sensitivity resulting in insulin resistance. These conditions lead to decrease glucose uptaken by cells and glucose levels elevated in the blood, which further develops into diabetes mellitus type 2.

CONCLUSION

Factors that affect the occurrence of insulin resistance are the consumption of fat food sources and food additives, fruit consumption, BMI, and MDA levels. Free radicals affect the occurrence of insulin resistance where the develpoment of MDA-free radical increases the risk of insulin resistance. Further research is needed on a larger scale and observation of the effect of food consumption levels on insulin resistance using quantitative methods of measuring nutrient intake, such as food recall or food record. Further studies are also needed to determine the factors that affect elevated levels of MDA as a trigger for insulin resistance.

REFERENCES

Arisawa, K., Uemura, H., Yamaguchi, M., Nakamoto, M., Hiyoshi, M., Sawachika, F., & Katsuura-kamano, S. (2014). Associations of Dietary Patterns with Metabolic Syndrome and Insulin Resistance : a cross-sectional study in a Japanese population. *The Journal of Medical Investigation*, *61*(4), 333–344. Retrieved from https://www.ncbi.nlm.nih.gov/pubmed/25264052

Erejuwa, O. O. (2012). Oxidative Stress in Diabetes Mellitus : Is There a Role for Hypoglycemic Drugs and / or Antioxidants? *Oxidative Stress and Disease*. Retrieved from cdn.intechopen.com/pdfs/35949.pdf

Evans, J. L., Goldfine, I. D., Maddux, B. A., & Grodsky, G. M. (2003). Perspectives in Diabetes: Are Oxidative Stress Activated Signaling Pathways Mediators of Insulin Resistance and Cell Dysfunction? *Diabetes*, *52*, 1–8. Retrieved from https://www.ncbi.nlm.nih.gov/pubmed/12502486

Hardinsyah, Riyadi, H., & Napitupulu, V. (2012). *Kecukupan energi, protein, lemak dan karbohidrat*. Bogor.

IDF. (2015). IDF Diabetes Atlas. International Diabetes Federation. Retrieved from www.diabetesatlas.org

Kalaivanam, K. N., Dharmalingam, M., & Marcus, S. R. (2006). Lipid peroxidation in type 2 diabetes mellitus. *Int J Diab Dev Ctries*, *26*(1), 30–32. Retrieved from http://www.ijddc.com

Karakelides, H., Irving, B. A., Short, K. R., Brien, P. O., & Nair, K. S. (2010). Age, Obesity, and Sex Effects on Insulin Sensitivity and Skeletal Muscle Mitochondrial Function. *Diabetes Care*, *59*(January), 89–97. http://doi.org/10.2337/db09-0591.

Kemenkes RI. (2014). *Situasi dan Analisis Diabetes*. Jakarta. Retrieved from www.depkes.go.id/download.php?file=download/pusdatin/infodatin/...diabetes.pdf%0A

Kusumo, A. S. L. D. (2005). *Hubungan antara Resistensi Insulin dengan Kadar Vitamin E Plasma pada Penderita Diabetes Mellitus Tipe 2*. Universitas Gadjah Mada. Retrieved from http://etd.repository.ugm.ac.id/index.php?mod=penelitian_detail&sub=PenelitianDetail&act=view&typ=html&buku_id=28403

Lim, S., Won, H., Kim, Y., Jang, M., Jyothi, K. R., Kim, Y., … Kim, S. S. (2011). Antioxidant enzymes induced by repeated intake of excess energy in the form of high-fat, high-carbohydrate meals are not sufficient to block oxidative stress in healthy lean individuals. *British Journal of Nutrition*, *106*, 1544–1551. http://doi.org/10.1017/S0007114511002091

Marfianti, E. (2009). Perbedaan Kadar Resistin pada Obes dengan Resistensi Insulin dan Obes Tanpa Resistensi Insulin. *Jurnal Kedokteran Dan Kesehatan Indonesia*, *1*(1). Retrieved from http://journal.uii.ac.id/index.php/JKKI/article/view/546

Merentek, E. (2006). Resistensi Insulin Pada Diabetes Melitus Tipe 2, (150), 38–41. Retrieved from www.itokindo.org/?wpfb_dl=118

Moreto, F., Oliveira, E. P. De, Manda, R. M., & Burini, R. C. (2014). The Higher Plasma Malondialdehyde Concentrations Are Determined by Metabolic Syndrome-Related Glucolipotoxicity. *Oxidative Medicine*

and Cellular Longevity, *2014*. Retrieved from https://www.hindawi.com/journals/omcl/2014/505368/

Moselhy, H. F., Reid, R. G., Yousef, S., & Boyle, S. P. (2013). A Specific, Accurate and Sensitive Measure of Total Plasma Malondialdehyde by HPLC. *Journal of Lipid Research*. Retrieved from https://www.ncbi.nlm.nih.gov/pubmed/23264677

Park, K. Y. E. (2008). *The Mechanisms Underlying Free Fatty Acid Induced Hepatic Insulin Resistance*. University of Toronto. Retrieved from https://tspace.library.utoronto.ca/.../Park_K_Y_Edward_20080...

Saini, V. (2010). Molecular mechanisms of insulin resistance in type 2 diabetes mellitus. *World Journal of Diabetes*, *1*(3), 68–75. http://doi.org/10.4239/wjd.v1.i3.68

Sayuti, K., & Yenrina, R. (2015). *Antioksidan Alami dan Sintetik* (1st ed.). Padang: Andalas University Press.

Sekarsari, B. (2016). Aturan Makan Ini Terbukti Mencegah Diabetes. Retrieved October 30, 2016, from http://www.1health.id/id/article/category/sehat-a-z/aturan-makan-ini-terbukti-mencegah-diabetes.html

Soewondo, P., Ferrario, A., & Tahapary, D. L. (2013). Challenges in diabetes management in Indonesia : a literature review. *Globalization and Health*, *9*(63), 1–17. Retrieved from https://www.ncbi.nlm.nih.gov/pubmed/24299164

Stern, M. P., & Mitchell, B. D. (1999). Genetics of Insulin Resistance. In G. M. Reaven & A. Laws (Eds.), *Insulin Resistance: The Metabolic Syndrome X*. New Jersey: Humana Press Inc.

Tang, Q., Li, X., Song, P., & Xu, L. (2015). Optimal cut-off values for the homeostasis model assessment of insulin resistance (HOMA-IR) and pre-diabetes screening: Developments in research and prospects for the future. *Drug Discoveries & Therapeutics*, *9*(6). Retrieved from http://www.ddtjournal.com/files/DDT_2015Vol9No6_pp380_429.pdf

Tiwari, B. K., Pandey, K. B., Abidi, A. B., & Rizvi, S. I. (2013). Markers of Oxidative Stress during Diabetes Mellitus. *Journal of Biomarkers*, *2013*, 378790. http://doi.org/10.1155/2013/378790

Valko, M., Leibfritz, D., Moncol, J., Cronin, M. T. D., Mazur, M., & Telser, J. (2007). Free radicals and antioxidants in normal physiological functions and human disease. *The International Journal of Biochemistry & Cell Biology*, *39*, 44–84. http://doi.org/10.1016/j.biocel.2006.07.001

WHO. (2016). *Global Report On Diabetes*. Geneva. Retrieved from apps.who.int/iris/bitstream/10665/.../9789241565257_eng.pdf

Yang, M. H., Hall, S. A., Piccolo, R. S., Maserejian, N. N., & Mckinlay, J. B. (2015). Do Behavioral Risk Factors for Prediabetes and Insulin Resistance Differ across the Socioeconomic Gradient ? Results from a Community-Based Epidemiologic Survey. *International Journal of E*, *2015*(April 4).