

HEATING THERAPY LOWERS BLOOD GLUCOSE LEVEL IN MICE (*Mus musculus*)

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ABSTRAK

*Diabetes Mellitus adalah penyakit metabolik yang telah menjadi salah satu penyakit yang mendesak di Indonesia. Bahkan dalam epidemiologi, diperkirakan bahwa pada tahun 2030 prevalensi Diabetes Mellitus (DM) di Indonesia mencapai 21,3 juta orang. Tujuan dari penelitian ini adalah untuk mengetahui efektivitas pemanasan terapi dalam menurunkan kadar glukosa darah. Penelitian ini dilakukan selama satu hari. Dengan jumlah sampel mencit (*Mus musculus*) jantan berat adalah 20grams. Mencit dibagi menjadi empat kelompok perlakuan, dan masing-masing kelompok terdiri dari 7 mices. Kelompok K1 tikus diperlakukan suhu 20°C (suhu kamar AC); Kelompok K2 tikus diperlakukan 28°C suhu (suhu kamar); Kelompok K3 tikus diperlakukan 37°C; dan kelompok K4 tikus yang diobati 40°C suhu. The mices telah berpuasa selama 16 jam, kemudian gula darah puasa diukur. Kemudian tikus diberi glukosa murni 0,2 gram setiap mices dan 30 menit kemudian tikus itu kembali diukur gula darah. Setelah itu, tikus dimasukkan ke dalam kotak terapi pemanasan dan diobati selama 30 menit. Dan kadar gula darah setelah tikus diukur ulang. Hasil: Dari hasil penelitian, ditemukan bahwa terapi pemanasan memberikan pengaruh yang signifikan dalam mengurangi kadar glukosa darah. Dalam penelitian ini, diperoleh bahwa pada suhu 37°C (kelompok K3) berbeda secara signifikan dengan pengobatan pada suhu 20°C (kelompok K1) dan 28°C (kelompok K2). Tetapi tidak berbeda nyata dengan perlakuan pada suhu 40°C (kelompok 4). Namun, hasil analisis, percobaan ini memberikan pemanasan pada kesimpulan bahwa terapi yang efektif dilakukan pada suhu 37°C-40°C. Tetapi suhu yang efektif adalah 37°C. Hal ini memberikan kesimpulan bahwa pengurangan efektif dalam kadar glukosa yang diperoleh pada suhu tidak melebihi suhu tubuh normal manusia. Hal ini diduga bahwa suhu tubuh mencapai 40°C hampir mencapai kelelahan dan suhu hipertermia dan durasi ketahanan akan menurun. Kesimpulan: Menurut hasil penelitian dapat disimpulkan bahwa terapi pemanasan di 37°C-40°C dapat mengurangi gluose darah mencit (*Mus musculus*). Perlu dilakukan penelitian lebih lanjut tentang durasi efektif dan mengenai efek samping dari terapi pemanasan. (FMI 2012;48:84-89)*

Kata kunci: terapi panas, kadar gula dar/ah, diabetes mellitus, penyakit metabolik

ABSTRACT

*Diabetes Mellitus is a metabolic disease that has become one of the urgent diseases in Indonesia. Even in epidemiology, it is estimated that by 2030 the prevalence of Diabetes Mellitus (DM) in Indonesia reached 21.3 million people. The objective of this study was to know the heating effectiveness of therapy in lowering blood glucose levels. The research was conducted for one day. With the number of samples of mice (*Mus musculus*) males weight was 20grams. Mice were divided into four treatment groups, and each group consist of 7 mice. K1 groups of mice were treated temperature of 20°C (room temperature AC); K2 groups of mice were treated 28°C temperature (room temperature); K3 groups of mice were treated 37°C; and K4 groups of mice treated 40°C temperature. The mice has been fasted for 16 hours, then fasting blood sugar is measured. Then the mice was given pure glucose 0,2 gram each mice and 30 minutes later mice were re-measured blood sugar. After that, mice incorporated into the heating therapy box and were treated for 30 minutes. And blood sugar levels after mice were re-measured. Results: From the research, found that the heating therapy provides a significant effect on reducing blood glucose levels. In this experiment, be obtained that at 37°C (group K3) differ significantly with treatment at a temperature of 20°C (group K1) and 28°C (group K2), but not significantly different from treatment at a temperature of 40°C (group 4). However, the results of the analysis, these experiments provide heating to the conclusion that effective therapy performed at 37°C-40°C. But the effective temperature is 37°C. This gives the conclusion that the effective reduction in glucose levels obtained at a temperature not exceeding the normal human body temperature. It is suspected that the body temperature reaches 40°C has almost reached exhaustion and hyperthermia temperature and duration of endurance will decrease. Conclusions: According the results of research it can be concluded that the heating therapy in 37°C-40°C can reduce blood gluose of mice (*Mus musculus*). It needs to do further research on effective duration and about the side effects of heating therapy. (FMI 2012;48:84-89)*

Keywords: heating therapy, blood glucose level, diabetes mellitus, metabolic diseases

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INTRODUCTION

Diabetes Mellitus is a metabolic disease that has become one of the urgent diseases in Indonesia. Epidemiological fact, it is estimated that by 2030 the prevalence of diabetes mellitus (DM) in Indonesia reached 21.3 million people Riskesdas Results Health Research in 2007, found that the proportion of causes of death due to diabetes mellitus in the age group 45-54 years in urban areas occupy rank-2 is 14.7% and in rural areas, Diabetes Mellitus is ranked 6th, namely 5.8% (Ministry of Health, Republic of Indonesia, 2010). Based on two studies conducted a decade apart, indicating the potential for diabetes prevalence rate increased rapidly, in a short time the urban population. This study shows a threefold increase in the numbers of patients with Diabetes Mellitus in two decades (Cockram 2000). Diabetes is classified into several types, but the type most often found in the community is Diabetes Mellitus Diabetes Mellitus Type 1 and Type 2. Type 1 diabetes is caused by autoimmune destruction of beta cells. Most individuals with Diabetes Mellitus Type 1 terbukti have autoimmune disease (Braunwald et al 2001). Type II diabetes, insulin-dependent atapun Diabetes Mellitus (NIDDM), due to the effects of the target tissue experience reduction of insulin metabolic sensitivity. Decreased sensitivity to insulin in the long run this will lead to insulin resistance (Guyton & Hall, 2000).

Today, Type 2 diabetes affects more than diabetes type 1 with a ratio of 4: 1. Given the severity of long-term complications of diabetes, health epidemic consequences will become more powerful and become a threat to the health care system, particularly in developing countries due to the estimated number of patients increased more than twofold (Cockram 2000). Therapy for Diabetes Mellitus is the use of Oral Anti-Diabetic Agents and Insulin. Both will lead to dependency and increasing doses continuously for by increasing the levels of ligand, receptor sensitivity will decrease. Use of Insulin injection will increase the level of insulin in the circulation. In fact, an increase in circulating insulin can lead to insulin receptor down-regulation and create risk of insulin resistance (Molina 2006). Therefore we need an adjuvant therapy in lowering blood glucose levels.

Heating therapy is a process of organism exposure to high temperatures. Heating therapy aimed to increase the temperature of an organism that affects the levels of free radicals (ROS and NO) in the body of the organism. On heating therapy trial ever conducted, mice were placed in ambient temperatures for 20 minutes in the water bath so that the rectal temperature reaches 41°C - 41.5°C (Gupte et al 2009). Other studies using a

temperature of 41.5°C for 10-15 minutes using an electric heating blanket (Chung et al 2008). Heating therapy will trigger generates stress proteins called heat shock proteins. The use of heating Therapy can increase ROS and NO are present in skeletal muscle. This can lead to the synthesis of Hsp 72 and Hsp 32. Hsp 72 and Hsp Synthesis 32 will trigger the synthesis Adipoectin (cytokines from fat cells) that play a role in inhibiting insulin resistance (Gupte et al 2009). Hsp 72 inhibits the activation of JNK (June N-terminal kinase) receptors so that insulin resistance does not occur and insulin-induced glucose uptake can run. In addition, Hsp 32 also will inhibit JNK activation receptors and concomitant increase in Hsp 32, there will be an increase in GLUT-4 glucose transporter that serves as the muscles. The increase of GLUT-4 will increase glucose uptake in muscle. 80-90% is done by setting the levels of glucose glucose uptake by skeletal muscle. Therefore, heating therapy in skeletal muscle is expected to have an influence in the management of Diabetes Mellitus (Molina 2006). But now, research on the heating therapy is still very limited and its application in everyday life as well as the medical field is still very minimal. Therefore, the object of this therapy is skeletal muscle. In addition, the relative skeletal muscle located on the surface making it easier to increase the temperature compared to the overall increase in body temperature (Gupte et al 2009).

With the properties owned by heating therapy, the researchers initiated the use of heating therapy as adjuvant therapy that is easy, inexpensive and effective for patients with Diabetes Mellitus Type 2. However, the way the use of heating therapy not yet fully known. Therefore, in this study, researchers will compare the levels of effective temperatures for heating therapy in Diabetes Mellitus. The purpose of this study was to determine the effectiveness of the heating therapy in lower glucose levels.

MATERIALS AND METHODS

This research was Experimental Laboratories using a completely randomized design to determine the possibility of a causal relationship by providing treatment in the experimental group compared with the control group. The research design used is Pre Post-test Control Group Design (Zainuddin, 1999). The population in this study was mice (*Mus musculus*). Mice were obtained from the Laboratory of Pharmacology of the Faculty of Medicine, University of Airlangga. Selected adult mice, male sex, about 3 months old, weigh about 20 grams, and in good health, namely physical and psychological well complete morphology.

The sample size in this study on the samples of each group was 7 animals. Techniques in the study group allocation by simple random sampling. Of the samples that meet the criteria, ie mice (*Mus musculus*) adult, male sex, about 3 months old, weigh about 20 grams, and in good health, namely physical and psychological well complete morphology. Mice were obtained from the Laboratory of Pharmacology of the Faculty of Medicine, University of Airlangga. Selected adult mice, male sex, about 3 months old, weigh about 20 grams, and in good health, namely physical and psychological well complete morphology. Furthermore, these animals will be divided into four major groups, namely K1.K2.K3 and K4 are respectively receiving therapy with a heating temperature of 27°C, 16°C, 37°C and 40°C

Whole mice were fasted for 16 hours before treatment (Pederson et al 2005). It is intended to assess the blood glucose levels of the mice. After fasting, the blood sugar levels of mice was measured with a blood glucose check tool (optoelectronic evaluation of test strips) using the blood of mice (Ocktarini 2010). The whole mice were fed with high glucose feed as much as has been determined, ie 1 mL solution of glucose that has been made. Then the mice were allowed to rest for 1 hour. After one hour, the mice returned blood drawn from the tail end to determine the blood glucose levels of mice during the peak of the cycle of glucose in the brother was completely (Prieto et al, 2004). Thereafter, mice were given appropriate treatment of each group for 20 minutes.

K1 Group is the negative control group. Mice were placed in a sealed wooden box with little air holes. In the space of the city is given as a reference thermometer in the box room temperature. In this group was not given heating or cooling. The temperature inside the box adapted to ambient temperatures of around 27°C. In the K2 group, mice placed in a wooden box sealed with little air holes. In the space of the city is given as a reference thermometer in the box room temperature. In the group given cooling until the temperature reaches 16°C box. Cooling can be manipulated to give a box of ice cubes around the room until the thermometer shows a temperature of approximately 16°C. In the group of K3 mice placed in a wooden box sealed with little air holes. In the room the box is given as a reference thermometer temperature of the room in the box. In this group, the temperature adjusted to body temperature in mice. Body temperature of the mice was about 37°C. Heating bias manipulated by providing specific irradiation with light power until thermometer showed the room approximately 37°C.

In the group of mice K4 placed in sealed wooden box with little air holes. In the room the box is given as a reference thermometer temperature of the room in the box. The group was given with the heating temperature of 40°C. Heating can be manipulated to provide certain irradiation with light power until thermometer showed the room approximately 40°C. After treated for 20 minutes, immediately after treatment is completed, each rat was measured its rectal temperatures. After the mice were re-measured blood glucose. Furthermore, the experimental results were analyzed to see the effects of heating therapy to decrease blood glucose. From the experimental results, first, we performed data analysis and dissemination of data centralization. Second, the data were tested for normality. If the data are normal, it was followed by a test of meaningfulness or significance quantitatively by the method of ANOVA test and LSD test. If the data is not normal, qualitative testing is descriptively.

RESULTS

Research on the effect of heating therapy (therapies heating) to decrease blood glucose levels in mice (*Mus musculus*) have been carried out in the Laboratory of Experimental Animal, Biochemistry, Faculty of Medicine, University of Airlangga. Of the 28 samples were divided into four groups by the number of each of the seven mice. This study was conducted for one day on November 23, 2011. The results of the study, the data obtained delta mean blood glucose levels of mice after 30 minutes was given glucose and after treated as in Table 1.

Table 1. Average delta blood glucose levels of mice after 30 minutes was given glucose and after a given treatment

| Groups | Average delta blood glucose levels of mice after 30 minutes was given glucose and after a given treatment |
|--------|---|
| K1 | 16.4286 |
| K2 | 12.0000 |
| K3 | 89.5714 |
| K4 | 39.3929 |

After homogeneity test, ANOVA test performed on the data delta blood sugar levels of mice 30 minutes after being given glucose and after treatment at a specific temperature. The results showed that there were significant differences on the provision of heating therapy to decrease blood glucose levels minutes (*Mus musculus*). The results are consistent with the initial hypothesis, because the significance value (p) in the ANOVA test is less than 0.05. So further statistical test

followed by Duncan. From the results of the statistical analysis of the results obtained following LSD

Table 2. The mean delta blood sugar levels of mice after 30 minutes were given glucose and after treatment.

| Groups | Average delta blood glucose levels of mice after 30 minutes was given glucose and after a given treatment | SD |
|--------|---|----------|
| K1 | 16.0000 | 41.78118 |
| K2 | 8.2857 | 18.91837 |
| K3 | 89.5714 | 39.26770 |
| K4 | 39.5714 | 68.89329 |

From the results of further analysis of LSD, showed that the treatment at 37°C (K3 group) significantly different from treatment at a temperature of 20°C (group K1) and 28°C (group K2). However, it was not significantly different from the treatment at 40°C (group 4). From the results of the statistical analysis, it was found that the therapy was effective temperature heating at 37°C it is supported by a decrease in glucose is highest in the group treated at 37°C (K3).

DISCUSSION

This study aims to determine the effect of therapy heating to decrease glucose levels in mice (*Mus musculus*). Mice used were male sex mice, aged 2-3 months, and in healthy physical condition. Each group consisted of six mice and the mice primary backup for a total of seven individuals. This study was conducted for one day on November 23, 2011 in Animal Testing Laboratory Department of Biochemistry Try Airlangga University School of Medicine. Heating therapy treatment conducted with different temperature on each group of mice.

Measurement of blood sugar levels in mice conducted Try Animal Testing Laboratory Department of Biochemistry, Faculty of Medicine, University of Airlangga. Blood glucose was measured three times, the first blood glucose measured after mice were fasted for 16 hours. The second 30 minutes after the mice were fed. Third 30 minutes after treatment with temperature. Fasting blood glucose levels are used to test the homogeneity minutes. While the difference of blood glucose levels of mice after 30 minutes were fed with glucose and after treatment (delta) is used to determine how large a decrease in blood sugar levels of mice. Results of data from the delta measurement of blood sugar levels of mice after 30 minutes were fed with

glucose and after treatment were analyzed by ujiAnova (Analysis of Variance) with an error rate? = 5%, and advanced test LSD.

From the research, it was found that the heating therapy gives significant effect on the decrease in blood glucose levels. In this experiment, it was found that at 37°C (K3 group) significantly different from treatment at a temperature of 20°C (group K1) and 28°C (group K2). However it was not significantly different from the treatment at 40°C (group 4). While the heating therapy trials ever conducted, rats were placed in a hot ambient temperature until the rectal temperature reaches 41°C - 41.5°C for 20 minutes using a water bath (Gupte et al 2009). Other studies using a temperature of 41.5°C for 10-15 minutes using an electric heating blanket (Chung et al 2008). However, from the results of the analysis, these experiments lead to the conclusion that the effective heating therapy performed at 37°C-40°C. However, the effective temperature for this study was 37°C. This gives the conclusion that the effective reduction in glucose levels obtained at a temperature that does not exceed the normal temperature of the human body. It is suspected that the body temperature of 40°C reached exhaustion and nearly temperature hyperthermia and duration of endurance will decrease.

Heating therapy is a process of organism exposure to high temperatures. Heating therapy aimed to increase the temperature of an organism that affects the levels of free radicals (ROS and NO) in the body of the organism. This is what plays a role in the decrease in blood glucose levels. Additionally, the provision of heating therapy in diabetes mellitus lowered glucose levels and improved insulin action at the receptor. One is the insulin signal transduction pathway through the molecule PKB (protein kinase B) and the work of these molecules increase with the heating therapy (Gupte et al 2009).

Heating therapy will trigger generate stress proteins called heat shock proteins. The use of heating Therapy can increase ROS and NO are present in skeletal muscle. This can lead to the synthesis of Hsp 72 and Hsp 32. Whereas, in diabetes mellitus, Hsp 72 and Hsp 32 levels are decreased (Kurucz et al 2002). In fact, this protein has a protective role in the cell, thus helping cells cope with environmental imbalances such as in diabetic conditions. Heating therapy will trigger the generation of stress proteins called heat shock proteins. The following will discuss the effect of heating to elevated levels of Hsp therapy.

Intramuscular levels of Hsp 72 in patients with type 2 diabetes decreased by 33% (Kurucz et al 2002). Reduction will significantly related to the development

of insulin resistance in the cells. Increased Hsp 72 induced by heating therapy can prevent hyperglycemia, hyperinsulinemia, glucose intolerance and insulin resistance (Chung et al 2008). Increased heat shock proteins may be one solution and can be triggered by heating therapy. As a result of the heating process of therapy, there are two pathways that play a role in the activation of HSF (Heat shock factor), thereby increasing the expression of Hsp 72. Levels of ROS such as superoxide anion (O₂⁻) and hydrogen peroxide (H₂O₂) will increase after the heating process of therapy (Zuo et al 2000). Superoxide anion will trigger the activation of HSF transcription factor gene encoding heat shock protein (Nishizawa et al 1999). ROS will activate HSF and improve transcription of heat shock proteins 72.

The second mechanism, heating therapy increases the levels of nitric oxide (NO). NO activates pathways Phospho Kinase C (PKC), which subsequently activates HSF increases ROS synthesis of Hsp 72. Hsp 72 that activates expression increased, this can be overcome insulin resistance occurrence. Heating therapy can improve the condition of hyperglycemia and impaired glucose tolerance (Chung et al 2008). Hsp 72 works in several ways. First, Hsp 72 inhibits JNK pathway and IK-β. In patients with insulin resistance, activation of JNK signal transduction pathway This pathway will inhibit the increased tyrosine phosphorylation of the IRS thereby inhibiting pathways ending in the release of GLUT-4 as a transporter to transport the glucose enter the cells (Nakatani et al 2004). Hsp 72 inhibits the activation of JNK that insulin resistance does not occur and insulin-induced glucose uptake can run (Gupte et al 2009, Chung et al, 2008). Second, Hsp 72 increases in circulating adiponectin. Adiponectin, a cytokine that is released by cells adipocytes or fat cells. Adiponectin can inhibit the phosphorylation of IK-β which plays a role in insulin resistance. Increased levels of Hsp 72 may increase the levels of adiponectin in circulation, so as to prevent the occurrence of insulin resistance (Gupte et al 2009). Third, Hsp 72 increases the activity of mitochondria. In patients with diabetes mellitus, lipid accumulation can occur in skeletal muscle myositis. The accumulation of these lipids can reduce the function of mitochondria is important as an energy-producing cells, by inhibiting oxidative phosphorylation and ATP-induced decrease in insulin production (Braunwald et al 2001). Increased Hsp 72 in skeletal muscle is closely related to an increase in mitochondrial function. Gupte Research in 2009 showed that skeletal muscle therapy heating using oxygen therapy coupled ATP more than the control. Improved mitochondrial function occurs due to inhibition of the Stress Kinase Activation done by Hsp72.

Heating therapy may increase the levels of NO in the cell. There was also a Reactive Oxygen Species such as O₂ (Zuo et al 2000). ROS and NO levels were increased in the cells, induces the formation of heme oxygenase-1. Induction of ROS and NO are done can be done in parallel or to form compounds peroxynitrite (ONOO⁻). Thus, therapeutic heating therapy can increase the production of HO-1 compounds. HO-1, hereinafter referred to as the protein Hsp 32 is produced when there is stress on the environment and has a molecular weight of 32,000 Da. HO-1 was first discovered in the spleen which functions oxidizes heme (Tenhunen et al 1972). It turns out that this molecule is not only synthesized in the spleen but also other organs and synthesis triggered by a disturbance in the cells.

Hsp 32 is increased in skeletal muscle lowers fasting blood glucose levels and post-prandial conditions of diabetes mellitus. Inhibition of JNK activation occurs, a factor which inhibits signal transduction of insulin, causing insulin resistance. Increased Hsp32 followed by increased expression of GLUT-4 which is a glucose transporter in muscle. Hsp32 also increases in circulating adiponectin (Ndisang & Jadhav, 2009). Adiponectin is an anti-inflammatory cytokine that is produced only by cells adipocytes. Adiponectin increases insulin sensitivity and inhibit the inflammatory process. In muscle, adiponectin increases glucose transport and increases fatty acid oxidation in relation to the activation of AMP kinase (Braunwald et al 2001). Therefore, Hsp 32 helps overcome insulin resistance and increases glucose uptake in the muscle so that it can lower blood glucose levels. It has been investigated also that Hsp 32 may drive the insulin sensitivity evidenced by a decrease in blood glucose levels (Nicolai et al 2009).

Hsp 72 and Hsp synthesis 32 will trigger the synthesis Adiponectin (cytokines from fat cells) that play a role in inhibiting insulin resistance (Gupte et al 2009). Hsp 72 inhibits the activation of JNK (June N-terminal kinase) receptors so that insulin resistance does not occur and insulin-induced glucose uptake can run. In addition, Hsp 32 also will inhibit JNK activation receptors and concomitant increase in Hsp 32, there will be an increase in GLUT-4 glucose transporter that serves as the muscles. The increase of GLUT-4 will increase glucose uptake in muscle. 80-90% is done by setting the levels of glucose uptake by skeletal muscle. Therefore, heating therapy in skeletal muscle is expected to have an influence in the management of Diabetes Mellitus (Molina 2006).

CONCLUSION

Heating therapy can lower blood glucose levels in mice (*Mus musculus*) and can be used as an alternative therapy with Diabetes Mellitus Type 2. Heating therapy effective temperature is 37°C.

REFERENCES

- Braunwald E, Fauci AS, Kasper DL, Hauser SL, Longo DL, Jameson JL (2001). Harrison's Principles of Internal Medicine, 15th ed, New York, McGraw-Hill Professional Publishing
- Chung J, Nguyen AK, Henstridge DC, Holmes AG, Chan MH, Mesa JL, Lancaster GI, Southgate RJ, Bruce CR, Duffy SJ, Horvath I, Mestrlil R, Watt MJ, Hooper PL, Kingwell BA, Vigh L, Hevener A, Febbraio MA (2008). HSP72 protects against obesity-induced insulin resistance. *Proc Natl Acad Sci USA* 105, 1739-1744
- Cockram CS (2000). The epidemiology of diabetes mellitus in the Asia-Pacific region. *Hong Kong Med J* 6, 43-52
- Gupte AA, Bomhoff GL, Swerdlow RH, Geiger PC (2009). Heat treatment improves glucose tolerance and prevents skeletal muscle insulin resistance in rats fed a high-fat diet. *Diabetes* 58, 567-578
- Guyton AC and Hall JE (2000). Textbook of Medical Physiology, 10th ed, Philadelphia, WB Saunders Company
- Kementerian Kesehatan Republik Indonesia (2010). Tahun 2030 Prevalensi Diabetes Mellitus di Indonesia Mencapai 21,3 Juta Orang. Available from <http://www.depkes.go.id/index.php/berita/press-release/414-tahun-2030-prevalensi-Diabetes-melitus-di-indonesia-mencapai-213-juta-orang.html>. Accessed September 21, 2011
- Kurucz I, Morva A, Vaag A, Eriksson KF, Huang X, Groop L, Koranyi L (2002). Decreased expression of heat shock protein 72 in skeletal muscle of patients with type 2 diabetes correlates with insulin resistance. *Diabetes* 51, 1102-1109
- Molina PE (2006). Endocrine Physiology, 2nd ed, New York, McGraw-Hill Medical
- Nakatani Y, Kaneto H, Kawamori D, Hatazaki M, Miyatsuka T, Matsuoka TA, Kajimoto Y, Matsuhisa M, Yamasaki Y, Hori M (2004). Modulation of the JNK pathway in liver affects insulin resistance status. *J Biol Chem* 279, 45803-45809
- Ndisang JF and Jadhav A (2009). Heme oxygenase system enhances insulin sensitivity and glucose metabolism in streptozotocin-induced diabetes. *Am J Physiol Endocrinol Metab* 296, E829-E841
- Nicolai A, Li M, Kim DH, Peterson SJ, Vanella L, Positano V, Gastaldelli A, Rezzani R, Rodella LF, Drummond G, Kusmic C, L'Abbate A, Kappas A, Abraham NG (2009). Heme oxygenase-1 induction remodels adipose tissue and improves insulin sensitivity in obesity-induced diabetic rats. *Hypertension* 53, 508-515
- Nishizawa J, Nakai A, Matsuda K, Komeda M, Ban T, Nagata K (1999). Reactive oxygen species play an important role in the activation of heat shock factor 1 in ischemic-reperfused heart. *Circulation* 99, 934-941
- Oktarini R (2010). Pengaruh ekstrak herba anting-anting (*Acalypha australis* L.) terhadap kadar glukosa darah mencit Balb/C induksi streptozotocin. Bachelor Thesis. Universitas Sebelas Maret, Surakarta
- Pederson BA, Schroeder JM, Parker GE, Smith MW, DePaoli-Roach AA, Roach PJ (2005). Glucose metabolism in mice lacking muscle glycogen synthase. *Diabetes* 54, 3466-3473
- Prieto PG, Cancelas J, Villanueva-Peñacarrillo ML, Valverde I, Malaisse WJ (2004). Plasma D-glucose, D-fructose and insulin responses after oral administration of D-glucose, D-fructose and sucrose to normal rats. *J Am Coll Nutr* 23, 414-419
- Tenhunen R, Marver H, Pimstone NR, Trager WF, Cooper DY, Schmid R (1972). Enzymatic degradation of heme. Oxygenative cleavage requiring cytochrome P-450. *Biochemistry* 11, 1716-1720
- Zainuddin M (1999). Metodologi Penelitian Kefarmasian dan Kesehatan, Surabaya, Airlangga University Press
- Zuo L, Christofi FL, Wright VP, Liu CY, Merola AJ, Berliner LJ, Clanton TL (2000). Intra- and extracellular measurement of reactive oxygen species produced during heat stress in diaphragm muscle. *Am J Physiol Cell Physiol* 279, C1058-C1066