Contents lists available at IOCS

Science Midwifery

journal homepage: www.midwifery.iocspublisher.org

Magnesium and insulin leaf combination on glucose, low-density lipoprotein and malondialdehyde levels in wistar rats with diabetes mellitus

Astari Nurisani[1], Ana Hidayati Mukaromah[2], Purwanto AP[3], Mamay[4]

[1,4] Department of Medical Laboratory Technology, STIKes Karsa Husada Garut, Garut, Indonesia

[2,3] Fakultas Ilmu Keperawatan dan Kesehatan, Universitas Muhammadiyah Semarang, Semarang, Indonesia

ARTICLE INFO

Article history:

Received March 23, 2023
Revised April 13, 2023
Accepted April 20, 2023

Keywords:

Diabetes mellitus
Insulin Leaf
Glucose
Magnesium

ABSTRACT

Diabetes mellitus is a metabolic disease characterized by chronic hyperglycemia due to impaired insulin secretion, insulin receptor sensitivity and/or both. Hyperglycemia can trigger oxidative stress conditions that can increase the risk of micro and macrovascular complications. Preventive efforts need to be done such as giving supplements and traditional therapies that have low side effects, namely magnesium and insulin leaves. This research is experimental type with post test only control group. The samples in this study were 30 male wistar rats weighing 170-300g which had healthy conditions adapted for 7 days, given standard feed and given ad libitum drinking water. The results showed that the glucose level in the treatment group 3 when compared to the negative control group had a percentage decrease of 26.49%. LDL and MDA levels were lower in the combination treatment group of magnesium and insulin leaves when compared to the negative control group. The ANOVA results showed that there was a significant difference between the groups in the glucose test results ($p < 0.05$), and there was no significant difference between the groups in the LDL and MDA results ($p > 0.05$). The conclusion of this study was that the best results when compared to negative controls were the combination of magnesium at a dose of 300 mg and insulin leaves at a dose of 300 mg/KgBB had a decreased percentage of LDL levels by 52.38% and MDA levels of 29.11%, while for glucose levels the combination of magnesium doses 300 mg and insulin leaves 75 mg/KgBW had a percentage reduction of 26.49%

Corresponding Author:

Astari Nurisani,
Department of Medical Laboratory Technology,
STIKes Karsa Husada Garut,
Jl. Subyadinata No.7, Garut, 44150, Indonesia,
Email: nurisani.astari@gmail.com

INTRODUCTION

Diabetes mellitus is a metabolic disease characterized by chronic hyperglycemia due to disturbances in carbohydrate, fat, and protein metabolism due to defects in insulin hormone secretion due to damage to pancreatic β-cells or due to impaired insulin receptor sensitivity. Damage to β-pancreatic cells is associated with oxidative stress due to an imbalance between oxidants and antioxidants in

the body. (American Diabetes Association, 2015) Hyperglycemia triggers oxidative stress conditions that occur when the production of reactive oxygen species (ROS) exceeds antioxidants in the body. Oxidative stress generated by ROS has been reported as a cause of T2DM and the progressive development of its long-term complications of both micro and macrovascular complications (Prawitasari, 2019).

Insulin resistance in DM will trigger dyslipidemia, namely increased LDL cholesterol, increased triglycerides, decreased HDL cholesterol, and the presence of small dense LDL. (JS, 2001) LDL cholesterol is one type of lipoprotein composed of a hydrophibic core containing triglycerides and cholesterol esters wrapped by hydrophilic phospholipids, free cholesterol and apolipoproteins, especially B-100. LDL is responsible for transporting cholesterol from the liver to the tissues. LDL is more atherogenic in that it easily adheres to the inner wall of blood vessels, forms plaques and causes blockage of blood vessels, thus triggering atherosclerosis which can risk becoming a complication of cardiovascular disease (Badimon & Vilahur, 2012; JS, 2001).

Dyslipidemia conditions in DM can lead to overproduction of ROS which can further result in mitochondrial DNA damage and β-pancreatic cell malfunction. Overproduction of ROS can stimulate the oxidation of LDL (LDLox) which cannot be recognized by LDL receptors and attacks the lipid components of cell membranes resulting in lipid peroxidation. The disruption of LDLox uptake by macrophages in the blood vessel wall can lead to the development of atherosclerotic plaques. (Bajaj & Khan, 2014) Increased ROS is a marker of oxidative stress. Increased free radicals cause lipid peroxidation of cell membranes will also increase the end product, namely Malondialdehyde (MDA). (Asha et al., 2017)

Malondialdehyde (MDA) is a dialdehyde compound that is the end product of lipid peroxidation in the body. High MDA concentrations indicate oxidation processes in cell membranes. Erythrocyte and plasma MDA levels have been used as markers of free radical-induced tissue damage in vivo. The more chemically stable nature of MDA makes this compound more often used as a marker of oxidative stress than other compounds. MDA is a stable and accurate measurement component of lipid peroxidation and has helped explain the role of oxidative stress in a number of diseases (Mulianto, 2020).

Patients with DM who have been diagnosed can then carry out therapy to reduce blood glucose levels and other risk factors with diet and exercise together and treatment with oral drugs or require insulin injections. The first step that must be taken in managing DM is non-pharmacological management, in the form of following a healthy diet that is in accordance with the needs of calories and nutrients such as vitamins and minerals, as well as increasing physical activity and regular physical exercise (Pusat Data dan Informasi Kementerian Kesehatan RI, 2020; Soelistijo et al., 2015). Fulfillment of vitamin and mineral needs can be by taking supplements that function to increase the intake of vitamins and minerals needed by the body and prevent the body from the risk of experiencing malnutrition. DM can be controlled by consuming the right vitamins and minerals, such as chromium, zinc, and magnesium. Chromium and zinc are included in the micromineral group, while magnesium is included in the macromineral whose needs are needed in large quantities, therefore it is necessary to add magnesium by consuming supplementation to meet its needs. (Kimball et al., 2017)

The second step that can be taken to control glucose levels is by pharmacological intervention with oral hypoglycemic drugs (OHO) and or insulin injections. The use of OHO must be consumed for life so that it requires high medical costs and long-term use of OHO can pose a risk of serious side effects. (Decroli, 2019) Metformin is one of the OHO that is widely used by DM patients because it is considered the safest with minimal side effects. (Pontarolo et al., 2015) Synthetic drugs have serious side effects so that safer and more effective therapies are needed to control DM. Therefore, since the last few years, therapeutic attention began to turn to herbal plants that have the potential to be used as medicines. (Baroni et al., 2008) One of the plants used in the community today to help reduce blood glucose levels is insulin leaf (Smallanthus sonchifolius). This plant, also known as

yacon, has a high content of polyphenols and fructooligosaccharides that can reduce blood glucose levels in DM. (Aditya & Adifa, 2016) The combination of magnesium therapy and insulin leaves is expected to control glucose levels more effectively and have minimal side effects so as to reduce complications that occur in DM.

Soltani's research (2007) showed a decrease in blood glucose and a significant increase in magnesium in type 2 DM rats given magnesium therapy at a dose of 10 g/L MgSO4 for 8 weeks. (Soltani et al., 2007) Sari's research (2015) showed a decrease in blood glucose levels, total cholesterol and triglycerides in type 2 DM rats given insulin leaf therapy at a dose of 300 mg/kgBB for 2 weeks. (Sari et al., 2015) Based on the above background, research will be conducted on the effect of magnesium capsules and insulin leaf capsules (Smallanthus sonchifolius) on glucose levels, LDL cholesterol and MDA in Wistar rats with DM.

RESEARCH METHOD

This research is experimental type with post test only control group design. The treatment is to give a combination of variations in magnesium concentration and insulin leaves, while what is measured as the end result is the level of glucose, LDL cholesterol and MDA in Wistar rats with DM. This research was conducted at the Laboratory of Animal Testing, University of Muhammadiyah Semarang.

The samples in this study were male wistar rats injected with streptozotocin 1 time at a dose of 55 mg/kg BW with inclusion criteria, namely age 6-8 weeks, body weight 170-300 grams and healthy conditions (not disabled and active), having glucose levels >200 mg/dL. Exclusion criteria are male Wistar rats sick during the adaptation period, do not have high blood glucose levels after being injected with streptozotocin. The sampling technique used was simple random sampling. Determination of sample size using the provisions of the calculation formula (t - 1) (r - 1) ≥ 15, with the minimum number of samples per group is 3 rats. In this study there were 6 control groups, namely negative control (K1), positive control using metformin (K2), control Mg dose 150 mg (K3), control Mg dose 300 mg (K4), control insulin leaf dose 75 mg / kg BW (K5), control insulin leaf dose 300 mg / kg BW (K6) and 4 treatment groups, namely the combination treatment of Mg dose 150 mg and insulin leaf dose 75 mg / kg BW (P1), combination treatment of Mg dose 150 mg and insulin leaf dose 300 mg/Kg BW (P2), combination treatment of Mg dose 300 mg and insulin leaf dose 75 mg/Kg BW (P3) and combination treatment of Mg dose 300 mg and insulin leaf dose 300 mg/Kg BW (P4).

Animal Preparation

The criteria of wistar were put into cages in the laboratory with three mice in each cage. The process of adaptation of mice adjusts to the new environment for seven days. Rats were fasted for 12 hours and injected with streptozotin once at a dose of 55 mg/Kg BW. After 72 hours (three days), blood was taken from the retroorbital plexus to check blood glucose and ensure that the results of blood glucose levels were above 200 mg/dL so that the rats could be categorized as hyperglycemia. The glycemic rats were then divided into four treatment groups and six control groups.

Reagent Preparation

The concentration of STZ made was 20 mg/mL by way of preparation, namely dissolving 600 mg of STZ in 30 ml of citrate buffer (50 mmol sodium citrate, pH 4.5) which was made before the study for a maximum of 15 minutes. The solution was stored in a 30 ml Falcon bottle and wrapped in aluminum foil. Storage of STZ solution is carried out at low temperature (4 °C).

The insulin leaves used are capsules containing 100% inuslin leaf extract under the brand "Insulmaxs". One capsule contains 500 mg of insulin leaf extract. The dosage of insulin leaves used was 75 mg/Kg BW and 300 mg/Kg BW. The average body weight of mice is 200 grams.

The magnesium used is a Mg supplement under the brand "Life Extension" which contains 500 mg of Mg in one capsule. The dose of Magnesium that will be given is 150mg and 300mg. The conversion factor for calculating drug doses from humans (70 kg) to rats (200 gr) is 0.018 and the volume of administration for optimization in rats is 2 ml/rat

All supplements or drugs that have been weighed according to the calculation will then be suspended using 0.5% NaCMC solution. Hot water entered to in the mortar as much as 50 mL, then sprinkle 500 mg of NaCMC in a manner slowly. Mix until homogeneous, marked by no longer visible white powder and the mixture is in the form of a gel . Add hot water little by little while stirring until the volume of the solution becomes 100 mL .

Treatment Process

At this stage, for 28 days the rats were given standard feed using BR-I feed as much as 20g/day and given therapy to lower blood glucose and LDL levels using magnesium and insulin leaves orally using a gastric tube per day. After 28 days, the rats were taken for blood to be examined for glucose, LDL and MDA for monitoring after being given therapy. The dosage of magnesium and insulin leaves in each group was varied with the following provisions:

Table 1. Dosage of magnesium and insulin leaves in groups

Group	Code	Na.CMC 0.5%	Metformin (mg/Kg BW/ day)	Magnesium (mg/Kg BW/ day)	Insulin Leaves (mg/Kg BW/ day)
Control (-)	K1	2ml	-	-	-
Control (+)	K2	-	500	-	-
Control Mg 1st	K3	-	-	150	-
Control Mg 2nd	K4	-	-	300	-
Control insulin leaves 1st	K5	-	-	-	75
Control insulin leaves 2nd	K6	-	-	-	300
Treatment 1st	P1	-	-	150	75
Treatment 2nd	P2	-	-	150	300
Treatment 3th	P3	-	-	300	75
Treatment 4th	P4	-	-	300	300

Glucose, LDL and MDA assesment

Blood was taken from the retroorbital plexus of male Wistar rats as much as 3 ml and put into a clean tube. The blood was then incubated for 30 minutes at room temperature, then centrifuged until serum was obtained and glucose, LDL and MDA levels were examined. Glucose measurement was carried out by an enzymatic method, namely glucose oxidation para aminophenazone (GOD-PAP) which can be measured photometrically. Measurement of serum LDL cholesterol levels using the direct method with the principle of enzymatic colorimetry. MDA measurements were carried out using the thiobarbituric acid (TBA) test method which can be measured spectrophotometrically.

RESULTS AND DISCUSSIONS

The overall research stage starts from the sample adaptation process, sample induction, suspense making, treatment process, specimen taking on the sample, specimen measurement and result analysis. The results of the study are presented in tables and figures.

Characteristics of the Research Subjects

The results of blood glucose levels of Wistar rats after three days of injection with STZ dose of 55 mg/Kg BW can be seen in Table 2.

Table 2. Mean glucose level after 72 hours of STZ injection

Group	Average Glucose Level (mg/dL)
K1	328,67 ± 78,75
K2	445,00 ± 58,51
K3	355,33 ± 117,64
K4	486,00 ± 65,64
K5	413,00 ± 66,34
K6	473,67 ± 27,79
P1	462,00 ± 38,20
P2	496,33 ± 17,90
P3	438,00 ± 46,70
P4	466,33 ± 28,29

Table 1 shows the results of the average glucose levels in each group in Wistar rats have glucose levels above 200 mg/dL so that it can be categorized as Wistar rats experiencing diabetic conditions.

Glucose Level Analysis

The results of the examination of serum glucose levels of Wistar rats are presented in Figure 1.

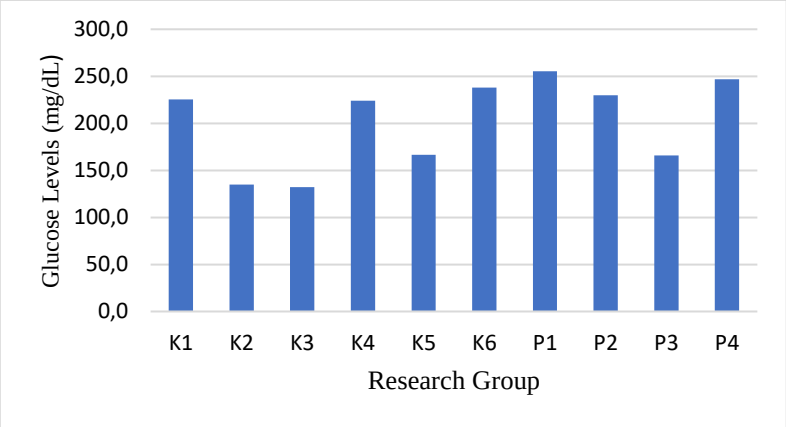

Figure 1. Average glucose level in DM wistar rats

The results show the average results of the glucose parameter examination for each group. The average glucose level in group K1 was 225.5 mg/dL. The K2 group had a glucose level of 134.8 mg/dL. Groups that had lower glucose levels compared to K1 were K3, K4, K5 and P3 with a percentage reduction in glucose levels of 41.42%; 0.68%; 26.24%; and 26.49%, respectively. Some groups had higher glucose levels compared to group K1, namely groups K6, P1, P2 and P4 with a percentage increase in glucose levels of 5.48%; 13.22%; 1.89%; and 9.43%, respectively.

The statistical test results show that in the data normality test using the Shapiro-Wilk test, all groups obtained a p-value ≥ 0.05, which means that the data were normally distributed. Data analysis was continued by calculating the similarity of data variation (homogeneity) using Levene's Test, the results showed that the p-value ≥ 0.05, namely 0.111, which means that the data variation is the same. Data analysis was continued using the One Way ANOVA test, the test results showed a p-value on glucose examination of 0.000 ($p \leq 0.05$) so there was a significant difference between groups with glucose levels in Wistar rats with DM. Further test (Post Hoc) was conducted to see the difference in the results of each group.

Analysis of LDL-cholesterol levels

The results of the examination of serum LDL-cholesterol levels of Wistar rats are presented in Figure 2.

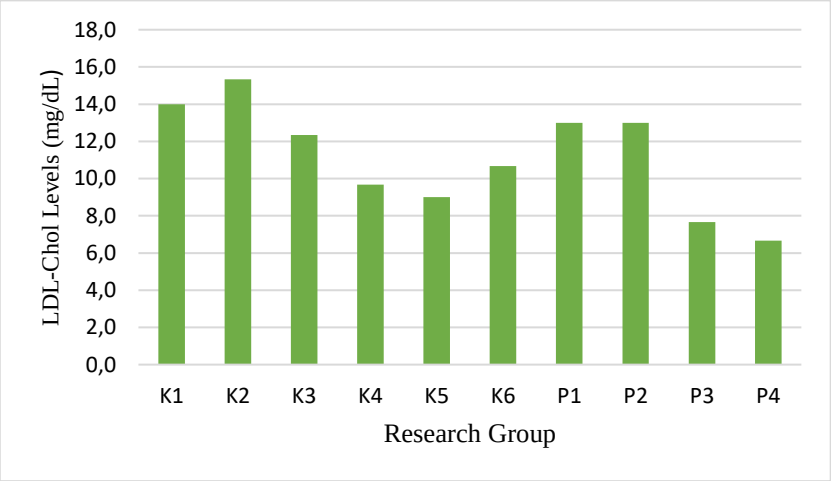

Figure 2. Average LDL-cholesterol levels in DM wistar rats

Figure 2 shows the average results of the examination of LDL-cholesterol levels in each group. The average LDL level in group K1 is 14.0 mg/dL, the average LDL level in group K2 is 15.3 mg/dL. The results of LDL levels of Wistar rats with DM in groups K3, K4, K5, K6, P1, P2, P3 and P4 have lower levels compared to K1 with a percentage decrease in LDL levels respectively by 11.91%; 30.95%; 35.71%; 23.81%; 7.14%; 7.14%; 45.24%; and 52.38%.

The statistical test results show that in the data normality test using the Shapiro-Wilk test, all groups obtained a p-value ≥ 0.05, which means that the data were normally distributed. Data analysis was continued by calculating the similarity of data variation (homogeneity) using Levene's Test, the results showed that the p-value ≥ 0.05, namely 0.070, which means that the data variation is the same. Data analysis was continued using the One Way ANOVA test, the test results showed the p-value in the LDL-cholesterol examination was 0.187 (p ≥ 0.05), it can be interpreted that there is no significant difference between groups with the results of LDL-cholesterol levels in Wistar rats with DM.

MDA Level Analysis

The results of the examination of serum MDA levels of Wistar rats are presented in Figure 3.

.

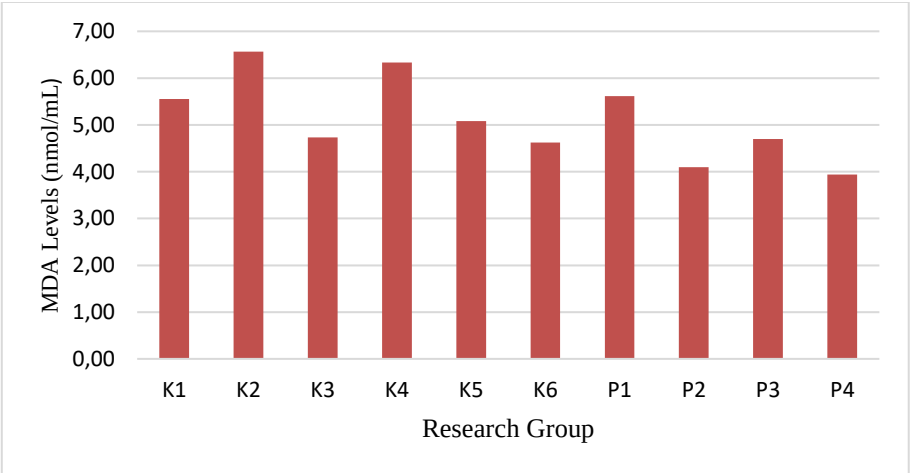

Figure 3. Average MDA levels in DM wistar rats

Figure 3 shows the average results of the MDA parameter examination for each group. The average MDA level in group K1 was 5.55 nmol/mL. The average MDA level in group K2 was 6.57 nmol/mL. Group K4 and P1 have higher average MDA levels than K1 with a percentage increase in MDA levels of 18.25%; 14.11%; and 1.08%, respectively. The average results of MDA levels in groups K3, K5, K6, P2, P3 and P4 have lower average results than group K1 with a percentage decrease in MDA levels, which are respectively 14.83%; 8.46%; 16.69%; 26.17%; 15.37%; and 29.11%.

The statistical test results show that in the data normality test using the Shapiro-Wilk test, all groups obtained a p-value ≥ 0.05, which means that the data is normally distributed. Data analysis was continued by calculating the similarity of data variation (homogeneity) using Levene's Test, the results showed that the p-value ≤ 0.05, namely 0.015, which means that the data variation was not the same. Data analysis continued using the One Way ANOVA test with the Brown-forsythe alternative test because the results of the data homogeneity test were not the same. This test is used to test the average difference in data between two or more groups. The results of the One-way ANOVA test showed a p-value in the MDA examination of 0.082 (p ≥ 0.05), which means that there is no significant difference between groups with the results of MDA levels in Wistar rats with DM. Alternative test results using Brown-forsythe showed a p-value of 0.149 (p ≥ 0.05), which means that there is no significant difference between groups with the results of MDA levels in wistar rats with DM.

Discussion

Diabetes mellitus is a chronic syndrome characterized by an increase in blood glucose levels (hyperglycemia) and increased urinary secretion due to insulin deficiency, the effect of insulin action and / or both. (American Diabetes Association, 2015) This study used Wistar rats that were made to experience diabetic conditions with a single medium dose injection using STZ which was characterized by an average blood glucose level > 200 mg / dL. The use of a single intermediate dose was carried out in this study because it can cause impaired insulin secretion. Hikmah (2015) mentioned that the toxic effect can be more stable in inducing diabetic conditions in rats(Hikmah et al., 2015)

Combination of Magnesium Supplement and Insulin Leaf on Glucose Levels in Wistar Rats with DM

The control group giving supplementation with a single dose that has the largest lower glucose levels when compared to K1 occurs in groups K3 and K5 with a percentage reduction in glucose levels which is 41.44% and 26.14% respectively. Group K3 was given a single dose of magnesium therapy of 150 mg, the administration of magnesium can reduce blood glucose levels because magnesium plays a role in maintaining homoestatic blood glucose along with the activity of factors involved in insulin sensitivity. Magnesium can work with tyrosine kinase enzymes in increasing insulin receptors so as to increase insulin sensitivity and make it easier for glucose to enter cells, and magnesium is a cofactor of various enzymes for glucose oxidation. .(Wang et al., 2013) Research conducted by Liu (2020) concluded that magnesium supplementation has a positive effect on insulin receptor activity and insulin sensitivity in rats with DM conditions. (H. Liu et al., 2020) Group K5 was given a single dose of insulin leaf therapy of 75 mg / kg BW. Insulin leaves can reduce blood glucose levels because it has an antidiabetic effect by inhibiting the process of glycogenolysis and glyconeogenesis, so it can increase liver insulin sensitivity in a state of insulin resistance. This is in line with the research of Satoh (2013) which states that the effect of yacon administration to lower blood glucose is most likely due to utilizing the effect on liver insulin sensitivity. (Satoh et al., 2013).

The treatment group with a combination of magnesium supplements and insulin leaves produced lower glucose levels than K1 only in the P3 group, namely the percentage reduction in

glucose levels by 26.49%. The P3 group was given a combination therapy of magnesium dose of 300 mg and insulin leaves dose of 75 mg/kg BW. The decrease can occur because insulin leaves containing FOS can increase the absorption of magnesium in the intestine so that magnesium in the blood can work together with insulin leaves to increase glucose sensitivity (Putri & Suriadi, 2015; Wang et al., 2013).

Groups P1, P2 and P4 have higher glucose levels compared to K1, this may be due to the different stress responses where the genes of each individual are different. According to Juster and Marin (2011) genetics can affect stress hormone levels, namely cortisol. (Saputra et al., 2018) Stress conditions in rats can also be caused by the sonde process which involves physical stress in the form of handling and restraint, insertion of rigid iron tubes or flexible plastic from the mouth to the stomach and gastric distension. The stress response also differs depending on the solvent administered. More viscous solvents induce more stress than less viscous solvents (water). In addition, the handling process included in the sonde process, namely the removal of the experimental animals, can increase the serum concentration of the hormone cortisol which mediates the stress response and regulates carbohydrate and protein metabolism so that it can increase blood glucose levels automatically. All rats in the study were well conditioned to minimize this, but some conditions cannot be avoided. (Dewi et al., 2017; Saputra et al., 2018)

Combination of Magnesium Supplement and Insulin Leaf on LDL Level in Wistar Rats with DM

Insulin resistance in DM will cause dyslipidemia characterized by an increase in LDL levels, triglycerides and a decrease in HDL levels. LDL which is atherogenic can trigger atherosclerosis conditions that can increase the risk of coronary heart disease complications in DM. (Laakso, 2010) The results showed LDL levels in the control group with a single dose (K3-K6) and the combined treatment group of two supplements (P1-P4) had lower LDL levels compared to the negative control (K1). The group that had the lowest levels compared to K1 occurred in the P3 and P4 groups with a percentage reduction in LDL levels of 45.24% and 52.38%, respectively. Treatment group 3 and treatment 4 were given the same magnesium dose combination of 300 mg, while the dose of insulin leaves given was 75 mg / kg BW for treatment 3 and 300 mg / kg BW for treatment 4.

Giving the combination with the same high dose can significantly reduce LDL levels. This can occur because insulin leaves containing FOS as a prebiotic that can produce SCFA can increase the absorption of minerals such as magnesium in the digestive tract and the uptake of FFA in adipose tissue. (Caetano et al., 2016) Good magnesium absorption can increase the amount of magnesium in the body, so that the role of magnesium as a cofactor for various enzymes can run well. In addition, in adipose tissue magnesium acts as an anti-inflammatory factor that can reduce IL-1 and TNF secretion so as to reduce the risk of atherosclerosis. (Gommers et al., 2016)

Combination of Magnesium Supplement and Insulin Leaf on MDA Level in Wistar Rats with DM

The toxic effect of STZ in inducing rats into DM is selective to pancreatic beta cells. STZ administration can trigger DNA damage which ultimately causes pancreatic beta cell necrosis through depletion of cellular energy stores. STZ administration causes the formation of free radicals and increases MDA significantly reducing the activity of antioxidant enzymes (Husna et al., 2019).

The results of MDA levels in the control group that had lower MDA levels compared to K1 were K3, K5 and K6 with a percentage reduction in MDA levels of 14.83%; 8.46%; and 16.69%, respectively. Treatment groups that have lower MDA levels compared to K1 are P2, P3 and P4 with a percentage reduction in MDA levels of 26.17%; 15.37% and 29.11%, respectively. The highest percentage reduction in MDA levels was found in group P4 which was given a combination of magnesium dose of 300 mg and insulin leaves dose of 300 mg/Kg BW. The group given a dose of 300 mg/kg BW insulin leaves in single dose therapy or combination dose therapy showed a decrease of >15%. This can occur because insulin leaves contain polyphenol compounds and flavonoid

compounds that function as antioxidants so that the production of free radicals in the body can be reduced. Flavonoids also play a role in repairing damage to pancreatic beta cells so that the pancreas can re-secrete insulin (Aditya & Adifa, 2016).

The combination of therapy with magnesium supplementation can increase the percentage of reduction in higher MDA levels because magnesium plays a role in improving mitochondrial function and reducing lipid peroxides so that it can reduce oxidative stress. (M. Liu & Jr, 2020) Analysis of MDA levels is a good and stable free radical analysis and its levels can be found in various biological fluids including serum as done in this study. MDA levels can also be analyzed in various organs related to the formation of free radicals due to hyperglycemia such as the pancreas, heart, liver and kidneys, its stable levels in isolated samples and is a specific product of fat peroxidation make it possible to determine the level of oxidative stress that occurs in certain organs. (Mulianto, 2020)

Limitations in this study are the level of stress in research animals that cannot be avoided due to the repeated sonde process, thus affecting blood glucose levels and the analysis of MDA levels which is only carried out from serum specimens, not along with organs related to the formation of free radicals due to hyperglycemia.

CONCLUSION

Based on the results of the study Combination of Magnesium and Insulin Leaves (*Smallanthus sonchifolius*) on Glucose Levels, Low Density Lipoprotein and Malondialdehyde in Wistar Rats with Diabetes Mellitus" it can be concluded that: There are statistical differences in glucose levels in each group, and there is a decrease in LDL levels and MDA in DM rats given high doses of magnesium and insulin leaf combination therapy. So that the combination of magnesium and insulin leaves can be used as an alternative therapy in reducing LDL and MDA levels to reduce the risk of complications of dyslipidemia in DM. Further research is recommended to analyze MDA levels in several organs related to the formation of free radicals due to hyperglycemia in DM

References

Aditya, M., & Adifa, D. P. (2016). Potensi Yacon (Smallanthus sonchifolius) sebagai Agen Antidiabetes. *Majority*, *5*(3), 68–72.

American Diabetes Association. (2015). Standards of Medical Care in Diabetes. *Diabetes Care Journal*, *38*(1), S1–S94.

Asha, K., Singal, A., Sharma, S. B., Arora, V. K., & Anggarwal, A. (2017). Dyslipidemia & Oxidative Stress in Patients of Psoriasis: Emerging cardiovascular risk factors. *Indian J Med Res*, *146*, 708–713. https://doi.org/10.4103/ijmr.IJMR

Badimon, L., & Vilahur, G. (2012). LDL-cholesterol versus HDL-cholesterol in the atherosclerotic plaque: Inflammatory resolution versus thrombotic chaos. *Annals of the New York Academy of Sciences*, *1254*(1), 18–32. https://doi.org/10.1111/j.1749-6632.2012.06480.x

Bajaj, S., & Khan, A. (2014). Antioxidants and diabetes. *Indian Journal of Endocrinology and Metabolism*, *16*(Suppl 2), S267-71. https://doi.org/10.4103/2230-8210.104057

Baroni, S., Suzuki-Kemmelmeier, F., Caparroz-Assef, S. M., Cuman, R. K. N., & Bersani-Amado, C. A. (2008). Effect of crude extracts of leaves of Smallanthus sonchifolius (yacon) on glycemia in diabetic rats. *Revista Brasileira de Ciencias Farmaceuticas/Brazilian Journal of Pharmaceutical Sciences*, *44*(3), 521–530. https://doi.org/10.1590/S1516-93322008000300024

Caetano, B. F. R., de Moura, N. A., Almeida, A. P. S., Dias, M. C., Sivieri, K., & Barbisan, L. F. (2016). Yacon (Smallanthus sonchifolius) as a Food Supplement: Health-promoting Benefits of Fructooligosaccharides. *Nutrients*, *8*(436), 1–13. https://doi.org/10.3390/nu8070436

Decroli, E. (2019). *Diabetes Melitus Tipe 2* (A. Kam, Y. P. Efendi, G. P. Decroli, & A. Ramhadi (eds.)). Pusat Penerbitan Bagian Ilmu Penyakit Dalam - Fakultas Kedokteran Universitas Andalas.

Dewi, I. K., Wulan, A. J., Ayu, P. R., Kedokteran, F., Lampung, U., Anatomi, B., Kedokteran, F., Lampung, U., Klinik, B. P., Kedokteran, F., & Lampung, U. (2017). Pengaruh Pemberian Ekstrak Etanol Kulit Manggis

Terhadap Kadar Glukosa Darah Tikus Putih Yang Diberi Paparan Gelombang Elektromagnetik Handphone Periode Kronik The Effect of the Ethanol Extract of Mangosteen Peel On Blood Glucose Level In the Rat Exposed. *Medula*, *7*(4), 164–170.

Gommers, L. M. M., Hoenderop, J. G. J., Bindels, R. J. M., & De Baaij, J. H. F. (2016). Hypomagnesemia in type 2 diabetes: A vicious circle? *Diabetes*, *65*(1), 3–13. https://doi.org/10.2337/db15-1028

Hikmah, N., Dewi, A., Shita, P., & Maulana, H. (2015). Rat Diabetic Blood Glucose Level Profile with Stratified Dose Streptozotocin (SD-STZ) and Multi Low Dose Streptozotocin (MLD-STZ) Induction Methods. *The Journal of Tropical Life Science*, *5*(1), 30–34.

Husna, F., Suyatna, F. D., Arozal, W., & Purwaningsih, E. H. (2019). Model Hewan Coba pada Penelitian Diabetes. *Pharmaceutical Sciences and Research (PSR)*, *6*(3), 131–141.

JS, F. (2001). Obesity. In Braunwald. E, Fauci. A, Harrison (Ed.), *principles of internal medicine* (15th ed., pp. 2152–2180). Mc-Graw-Hill.

Kimball, S. M., Emery, J. C. H., & Lewanczuk, R. Z. (2017). Effect of a vitamin and mineral supplementation on glycemic status: Results from a community-based program. *Journal of Clinical and Translational Endocrinology*, *10*(November), 28–35. https://doi.org/10.1016/j.jcte.2017.11.002

Laakso, M. (2010). Cardiovascular Disease in Type 2 Diabetes From Population to Man to Mechanisms. *Diabetes Care Journal*, *33*(2), 442–449. https://doi.org/10.2337/dc09-0749

Liu, H., Li, N., Jin, M., Miao, X., Zhang, X., & Zhong, W. (2020). Magnesium Supplementation Enhances Insulin Sensitivity and Decreases I nsulin Resistance in Diabetic Rats. *Iranian Journal of Basic Medical Sciences*. https://doi.org/10.22038/ijbms.2020.40859.9650

Liu, M., & Jr, S. C. D. (2020). Magnesium, Oxidative Stress, Inflammation, and Cardiovascular Disease. *Antioxidants*, *9*(907), 1–31.

Mulianto, N. (2020). Malondialdehid sebagai Penanda Stres Oksidatif pada Berbagai Penyakit Kulit. *Cermin Dunia Kedokteran*, *47*(1), 39–44.

Pontarolo, R., Cristina, A., Sanches, C., Wiens, A., Perlin, C. M., Tonin, F. S., Hiemisch, H., Borba, L., Lenzi, L., & Tavares, S. (2015). Pharmacological Treatments for Type 2 Diabetes. *INTECH*, *6*, 147–184.

Prawitasari, D. S. (2019). Diabetes Melitus dan Antioksidan. *KELUWIH: Jurnal Kesehatan Dan Kedokteran*, *1*(1), 48–52. https://doi.org/10.24123/kesdok.v1i1.2496

Pusat Data dan Informasi Kementerian Kesehatan RI. (2020). *Infodatin 2020 - Diabetes-Melitus* (pp. 1–10). P2PTM, Kementerian Kesehatan RI.

Putri, D. D. N., & Suriadi. (2015). Pengaruh Daun Insulin (Smallanthus sonchifolius) Terhadap Penurunan Kadar Glukosa Darah pada Tikus Diabetes Melitus. *Jurnal Proners*, *3*(1). https://doi.org/http://dx.doi.org/10.26418/jpn.v3i1.21329

Saputra, N. T., Suartha, N. I., & Dharmayudha, A. A. G. O. (2018). Agen Diabetagonik Streptozotocin untuk Membuat Tikus Putih Jantan Diabetes Mellitus. *Buletin Veteriner Udayana*, *10*(2), 116–121. https://doi.org/10.24843/bulvet.2018.v10.i02.p02

Sari, F. R., Hendarto, H., Muqorrobin, A., R, C. A. H., & Amelia, E. (2015). Insulin Leaves (Smallanthus sonchifolius) Dry Extract Improves Blood Glucose and Lipid Profile in Aloxan-Induced Rat. *Asian Jr. of Microbiol. Biotech. Env. Sc*, *17*(2), 405–408.

Satoh, H., Audrey Nguyen, M. T., Kudoh, A., & Watanabe, T. (2013). Yacon diet (Smallanthus sonchifolius, Asteraceae) improves hepatic insulin resistance via reducing Trb3 expression in Zucker fa/fa rats. *Nutrition and Diabetes*, *3*, e70-6. https://doi.org/10.1038/nutd.2013.11

Soelistijo, S., Novida, H., Rudijanto, A., Soewondo, P., Suastika, K., Manaf, A., Sanusi, H., Lindarto, D., Shahab, A., Pramono, B., Langi, Y., Purnamasari, D., & Soetedjo, N. (2015). Konsesus Pengelolaan Dan Pencegahan Diabetes Melitus Tipe2 Di Indonesia. In *Perkeni*. PB Perkeni.

Soltani, N., Keshavarz, M., & Dehpour, A. R. (2007). Effect of oral magnesium sulfate administration on blood pressure and lipid profile in streptozocin diabetic rat. *European Journal of Pharmacology*, *560*(2–3), 201–205. https://doi.org/10.1016/j.ejphar.2006.12.020

Wang, J., Persuitte, G., Olendzki, B. C., Wedick, N. M., Zhang, Z., Merriam, P. A., Fang, H., Carmody, J., Olendzki, G. F., & Ma, Y. (2013). Dietary Magnesium Intake Improves Insulin Resistance among Non-Diabetic Individuals with Metabolic Syndrome Participating in a Dietary Trial. *Nutrients*, *5*(10), 3910–3919. https://doi.org/10.3390/nu5103910