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Effect of Papaya Leaf Extract (*Carica Papaya* L.) on Two-Hour Postprandial Blood Glucose in Male Wistar Rats (*Rattus Norvegicus*) Induced by Streptozotocin and Nicotinamide

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ABSTRACT

Diabetes mellitus (DM) is a metabolic disease that involves inappropriately elevated blood glucose levels. Suspicion of diabetes mellitus can be done by checking two hours postprandial blood glucose. Glimepiride is a drug used in the treatment of diabetes mellitus that belongs to the sulfonylurea group. However, like other sulfonylureas, the administration of glimepiride in patients with diabetes mellitus can trigger side effects in the form of hypoglycaemia due to increased insulin secretion. One of the plants that can be utilized as an antidiabetic drug is papaya leaf. Research in the form of experimental Randomized Controlled Trial (RCT) with Pretest-Posttest Only Group Design research design using male wistar strain white rats for 38 days, with 35 rats. The administration of papaya leaf extract at a dose of 150mg/kg and 200mg/kg in group IV and V rats is proven to have the small effect of lowering two hours postprandial blood glucose in male Wistar rats. In conclusion depend on the dose the administration of papaya leaf had a small effect in changing two-hours postprandial blood glucose.

KEYWORDS: Papaya Leaf Extract, Diabetes Mellitus, Two-Hours Postprandial Blood Glucose.

ARTICLE DETAILS

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INTRODUCTION

Changes in lifestyle affecting changes in eating behaviour patterns can lead to the onset of disease. Diabetes mellitus (DM) is a metabolic disease that involves inappropriately elevated blood glucose levels. DM has several categories, including type 1, type 2, maturity-onset diabetes of the young (MODY), gestational diabetes, neonatal diabetes, and secondary causes due to endocrinopathies, steroid use, etc. Type 1 and 2 DM are the main sub-types, each with different pathophysiology, presentation, and management, but both have the potential for hyperglycaemia (1). Diabetes is one of the global health emergencies with the fastest increase in disease incidence, this was confirmed by the International Diabetes Federation (IDF) in the 10th edition of the Atlas. In the 10th edition of the Atlas of the International Diabetes Federation in Indonesia, it is estimated that the adult diabetic population aged between 20-79 years is 19,465,100 people out of a total adult population aged 20-79 years of 179,720,500, so that when calculated from these two figures more than 1/10 have diabetes (2).

Regardless of the specific type of diabetes, its complications involve microvascular, macrovascular, and neuropathic problems. Microvascular and macrovascular complications vary according to the degree and duration of uncontrolled diabetes, including nephropathy, retinopathy, neuropathy, and Atherosclerotic Cardiovascular Disease (ASCVD) events, especially when associated with other comorbidities such as dyslipidaemia and hypertension (1)

Suspicion of diabetes mellitus can be done by checking Two Hours Postprandial Blood Glucose (3). Glimepiride is a drug used in the treatment of diabetes mellitus which belongs to the sulfonylurea group (4). However, like other sulfonylureas, the administration of glimepiride in patients with diabetes mellitus can trigger side effects in the form of hypoglycaemia due to increased insulin secretion (5). Therefore, the development of antidiabetic drugs without side effects can be an innovation in diabetes management. One of the plants that can be utilized as an antidiabetic drug is papaya leaf. Phytochemical compounds have been identified in papaya leaves, mostly in the flavonoid class such as apigenin, catechin, deoxyquercetin, hesperitin,

isorhamnetin, kaempferol, myricetin, naringenin, protocatechuic acid, quercetin, and rutin. Flavonoid compounds are antioxidant compounds and are thought to restore insulin receptor sensitivity in pancreatic beta cells, thus causing a decrease in blood glucose levels in rats (6). Radical compounds derived from amine compounds have a very long termination stage. Alkaloids and tannins can also inhibit glucose absorption in the intestine. So that the presence of flavonoids, alkaloids and tannins has a beneficial effect on the state of diabetes mellitus (7).

Based on this background, researchers want to conduct research on the effect of papaya leaf extract (Carica Papaya L.) on changes in changing two hours postprandial blood glucose in male Wistar rats induced by streptozotocin and nicotinamide.

MATERIALS AND METHODS

The type of research used is experimental research Randomized Controlled Trial (RCT) with Pretest-Posttest Only Group Design using male wistar white rats to see the effect of papaya leaf extract on changes in changing two hours postprandial blood glucose. The samples used in the study were 35 male wistar rats aged 2-3 months with a body weight of 200-250gram, adapted for 2 weeks before the study as an adjustment. At the beginning of the study were divided into 5 groups consisting of 1 normal control group, 1 positive control group, 1 negative control group and 2 treatment groups. Group I is a normal control group that is only given standard food and drinks. Group II is a negative control group that was given STZ-NA induction on day 15 and then given standard food and drinks for 14 days. Group III is a positive control group that is given STZ-NA induction on day 15 which is then given glimepiride at a dose of 0.04mg/200gr and standard food and drinks for 14 days. Group IV is treatment group 1 which is given STZ-NA induction on day 15 which is then given papaya leaf extract at a dose of 150mg/kg and standard food and drinks for 14 days. Group V is treatment group 2 which is given STZ-NA induction on day 15 which is then given papaya leaf extract at a dose of 200mg/kg and standard food and drinks for 14 days.

Blood sampling of experimental animals is done three times and then will be measured using a blood glucose meter, Easy Touch GCU. The first test of two-hour postprandial blood glucose was done on the first day in the lateral vein of the rat's tail. First, the tail of the rat is cleaned first using an alcohol swab then cut slightly using a razor blade, then the blood glucose strip that has been attached to the device is then attached until the number appears on the monitor layer of the glucometer. The second test was done after diabetes induction on day 25. Before blood sampling, rats will be fed and waited for 2 hours to check blood glucose levels. Blood was taken through the lateral vein of the rat's tail. The third or last test is done after the administration of papaya leaf extract and glimepiride for 14 days and the blood taken is the lateral vein of the tail of the rat.

The data that has been collected from the sampling results will be analysed to determine the effect of papaya leaf extract on changing two hours postprandial blood glucose in male Wistar rats induced by Streptozotocin and Nicotinamide The data that has been obtained is then tested for normality with the Shapiro-Wilk test with the condition that $p>0.05$. Then, if the data is normally distributed and homogeneous, a significance difference test will be conducted between the control group and the treatment group using One Way ANOVA. Then the Least Significant Difference (LSD) post hoc test will be carried out to analyse the relationship between the 5 groups. If the data obtained is not normally distributed, the Kruskal-Wallis H non-parametric test will be carried out. Furthermore, calculations are carried out using Effect Size Cohen's D to determine the comparison of papaya leaf extract administration between groups.

RESULTS

Table 1. (Description of Changing Two Hours Postprandial Blood Glucose Level Ratio)

Test	GROUP	Mean ± SD	Minimum	Maximum
1	I	101.43 ± 8.753	90	114
	II	97.57 ± 8.886	84	107
	III	97.29 ± 12.553	83	123
	IV	109.14 ± 11.127	88	119
	V	108.00 ± 11.902	100	134
2	I	102.29 ± 20.353	65	129
	II	315.14 ± 210.758	126	601
	III	383.43 ± 214.830	133	601
	IV	370.43 ± 185.453	138	601
	V	411.85 ± 176.625	140	601
3	I	113.29 ± 29.284	93	178
	II	317.00 ± 228.731	121	601
	III	373.29 ± 233.259	113	601
	IV	434.86 ± 198.138	144	601
	V	423.71 ± 223.340	98	601

Table 2. (Normality Test Results)

GROUP	Test	Results
GROUP I	1	0.758
	2	0.359
	3	0.002
GROUP II	1	0.496
	2	0.062
	3	0.014
GROUP III	1	0.089
	2	0.065
	3	0.032
GROUP IV	1	0.151
	2	0.403
	3	0.034
GROUP V	1	0.003
	2	0.461
	3	0.016

*Significant if p > 0.05

It is known that the results of the normality test in all groups obtained several results stating that the p value <0.05, namely in test 1 group V, test 3 in all groups, which means that there is inequality in the study group, so it can be stated that the distribution is not normal.

Table 3. (Kruskal Wallis H Test Results)

GROUP	Results
GROUP I	0.795
GROUP II	0.001
GROUP III	0.001
GROUP IV	0.001
GROUP V	0.009

*Significant if p < 0.05

Based on the results of the Kruskal-Wallis H test above in all groups at the asymp value. Significant results obtained p < 0.05, which means there are differences in groups II to V. And insignificant in group I. And not significant in group I.

Table 4. (Mann-Whitney U Test Results 1 and 2)

GROUP	Results
GROUP I	0.665
GROUP II	0.002
GROUP III	0.002
GROUP IV	0.002
GROUP V	0.002

*Significant if p < 0.05

In the Mann-Whitney U test between test 1 and 2, it was found that the differences in group II to group V obtained p < 0.05, which means that the changing two hours postprandial blood glucose in the 1st and 2nd tests were significantly different. Whereas in group I the results obtained p> 0.05 which means there is no significant difference in blood glucose levels 2 hours postprandial in tests 1 and 2.

Table 5. (Mann-Whitney U Test Results 1 and 3)

GROUP	RESULTS
GROUP I	0.522
GROUP II	0.002
GROUP III	0.003
GROUP IV	0.002
GROUP V	0.035

*Significant if p < 0.05

In the Mann-Whitney U test between test 1 and 3, it was found that the differences in group II to group V obtained p < 0.05, which means that the two-hours postprandial blood glucose in the 1st and 3rd tests were significantly different. Whereas in group I the results obtained p> 0.05 which means there is no significant difference in two-hours postprandial blood glucose l in tests 1 and 3.

Table 6. (Mann-Whitney U Test Results 2 and 3)

GROUP	RESULTS
GROUP I	0.798
GROUP II	0.948
GROUP III	0.894
GROUP IV	0.564
GROUP V	0.897

*Significant if p < 0.05

In the Mann-Whitney U test between test 2 and 3, the results obtained p> 0.05, which means that there is no significant difference in the overall group of 2-hour postprandial blood glucose levels in the 2nd and 3rd tests.

Table 7. (Cohen's D Effect Size Test Results)

GROUP	Cohen's d formula (M2-M1) √((SD1 2 + SD2 2) : 2)	Sample	Results	Interpretation 0.20 = small 0.50 = medium 0.80 = large
1 and 2	$\frac{(243.24 - 105.67)}{142.514129}$	7	0.965308	The calculation result of cohen's d is 0.965308 which means there is a big influence of STZ and NA induction on two-hours postprandial blood glucose levels of male wistar rats in group II. It can be concluded that STZ and NA induction can cause an increase in two-hours postprandial blood glucose levels in male Wistar rats.
2 and 3	$\frac{(284.67 - 243.24)}{210.775785}$	7	0.19656	The result of calculation cohen's d is 0.19656, meaning that the effect of glimepiride administration at a dose of 0.04 mg/200gr in group III provides no effect size on changes in two-hours postprandial blood glucose levels in male wistars.
2 and 4	$\frac{(304.81 - 243.24)}{203.892367}$	7	0.301973	The result of calculation cohen's d is 0.301973, meaning that the effect of giving papaya leaf extract at a dose of 150mg / kg in group IV provides a small effect size on changes in two-hours postprandial blood glucose levels in male wistars.
2 and 5	$\frac{(314.52 - 243.24)}{208.528267}$	7	0.341824	The result of calculation cohen's d is 0.341824, meaning that the effect of giving papaya leaf extract at a dose of 200mg / kg in group V provides a small effect size on changes in two-hours postprandial blood glucose levels in male wistars.
3 and 4	$\frac{(304.81 - 284.67)}{214.039983}$	7	0.094095	The result of calculation cohen's d is 0.094095, meaning that the effect of giving papaya leaf extract at a dose of 150mg / kg in group IV does not provide an effect size on changes in two-hours postprandial blood glucose levels in male wistars given glimepiride at a dose of 0.04mg / kg in group III.
3 and 5	$\frac{(314.52 - 284.67)}{218.460649}$	7	0.136638	The result of calculation cohen's d is 0.136638, meaning that the effect of giving papaya leaf extract at a dose of 200mg / kg in group V does not provide an effect size on changes in two-hours postprandial blood glucose levels in male wistars given glimepiride at a dose of 0.04mg / 200gr in group III.
4 and 5	$\frac{(314.52 - 304.81)}{211.827102}$	7	0.045839	The result of calculation cohen's d is 0.045839, meaning that the effect of giving papaya leaf extract at a dose of 150mg / kg in group IV does not provide an effect size on changes in two-hours postprandial blood glucose levels in male wistars given papaya leaf extract at a dose of 200mg / kg in group V.

DISCUSSION

The administration of papaya leaf extract at a dose of 150mg/kg and 200mg/kg in male Wistar rats is proven to reduce two-hours postprandial blood glucose with a small effect compared to the administration of glimepiride at a dose of 0.04mg/200gram. The content of papaya leaf extract has several active compounds that function as antidiabetics, namely alkaloids, tannins, flavonoids, and saponins. The effect of alkaloid compounds in papaya leaf extract can trigger an increase in insulin secretion in pancreatic beta cells. Alkaloids also function to increase glucose uptake in the blood to be carried into cells (8). Tannin compounds are

compounds that can accelerate glucose absorption and have protective properties against pancreatic beta cells by binding free radicals (9). Flavonoids work by reducing glucokinase activity, liver gluconeogenesis, and glycogenolysis resulting in a decrease in blood sugar levels. Flavonoids also increase glucose uptake in the liver (10).

The content of saponin compounds in papaya leaf extract is effective in reducing blood sugar levels by inhibiting the alpha glucosidase enzyme. This enzyme is an enzyme that plays a role in converting carbohydrates into glucose. Therefore, inhibition of the alpha glucosidase enzyme can result in a decrease in blood sugar levels (11).

STZ is an induction of diabetes that can destroy β cells in the pancreas. This was proven by checking the second two-hours postprandial blood glucose after STZ-NA induction. In group II, the average two-hours postprandial blood glucose was 315.14 + 210,758, group III with an average of 383.43 + 214,830, group IV with an average of 370.43 + 185,453, and group V with an average of 411.85 + 176,625. This is also supported by the results of group I which is a normal group that does not give STZ-NA induction with an average two-hours postprandial blood glucose level of 102.29 + 20.353. Administration of STZ and NA can cause disruption and death of pancreatic beta cells. In anticipation of a significant impact, it is necessary to administer NA 15 minutes before STZ induction to protect pancreatic beta cells (12), (13), (14).

Toxic streptozotocin enters pancreatic beta cells through glucose transporter 2 (GLUT 2). The structure of STZ, which has similarities with glucose molecules, makes it easy for it to enter pancreatic beta cells. The entry of STZ into pancreatic beta cells triggers cell death triggered by DNA methylation. This triggers necrosis in pancreatic beta cells. Pancreatic beta cell death can also be caused by activation of cell repair enzymes such as nuclear poly ADP-ribose, nitric oxide production and the formation of free radicals such as hydrogen peroxide (12).

There are differences in the state of hyperglycaemia in rats due to the mechanism of STZ induction of pancreatic beta cell receptors in each individual male Wistar rat. Moderate hyperglycaemia is triggered by DNA damage which results in the release of nitric oxide to release free radicals and trigger damage to pancreatic beta cells. Severe hyperglycaemia occurs due to activation of poly ADP-ribosylation triggered by DNA damage, resulting in a decrease in ATP followed by a decrease in secretion and synthesis of insulin (15).

The administration of glimepiride at a dose of 0.04mg/200gr did not have an effect in reducing two-hours postprandial blood glucose levels. The ineffectiveness of glimepiride in this study can be caused by various things. The dose of glimepiride administration in the research of Sartika and Rahardiantini (2022) was 0.2mg / kg, 0.4mg / kg, 0.8mg / kg and obtained the highest blood sugar reduction rate at a dose of 0.8mg / kg. While the dose of glimepiride used in this study is 0.04mg/200gr. In addition, the administration of glimepiride is often combined with metformin to produce optimal effects in reducing blood sugar levels (17).

The administration of STZ-NA to rats affects the two-hours postprandial blood glucose levels of rats. This is evidenced by the second two-hours postprandial blood glucose test after STZ-NA induction. In group II, the average two-hours postprandial blood glucose was 315.14 + 210.758, group III with an average of 383.43 + 214.830, group IV with an average of 370.43 + 185.453, and group V with an average of 411.85 + 176.625. After being given glimepiride treatment and papaya leaf extract (Carica Papaya L.), the third two-hours postprandial blood glucose was checked and the average results of group II were 317.00 + 228,731, group III with an average of 373.29 + 233,259, group IV with an average of 434.86 + 198,138, and group V with an average of 423.71 + 223,340.

The administration of papaya leaf extract at doses of 150mg/kg and 200mg/kg in groups IV and V has little effect on changes in two-hours postprandial blood glucose in group II. This is in accordance with previous research by Ismukada, et al (2020) who said the optimal dose of papaya leaf extract in reducing blood sugar levels was 1000mg/kg.

The content of active compounds in papaya leaf extract in the form of alkaloids, tannins, saponins and flavonoids can cause hypoglycaemia effects. Alkaloids and tannins work by stimulating the release of insulin in pancreatic beta cells. While saponin and flavonoid compounds trigger the release of somatostatin to suppress the process of glucagon formation so that blood glucose levels decrease (19).

Comparison of two-hours postprandial blood glucose between the whole group showed a relatively small effect and did not even have a significant effect when the Cohen's D effect size test was carried out. So it can be concluded that the administration of papaya leaf extract at a dose of 150mg/kg and 200mg/kg has the potential to reduce two-hours postprandial blood glucose levels with a small effect. This is in accordance with research (Pudyawanti et al., 2018) by giving papaya leaf extract at a dose of 100mg/kg can reduce fasting blood sugar levels (19). However, researchers could not find research on papaya leaf extract that could reduce two-hours postprandial blood glucose levels.

Comparison between two-hours postprandial blood glucose groups IV and V showed insignificant results despite the difference in dose of papaya leaf extract. However, two-hours postprandial blood glucose in rats showed a decrease in accordance with the increase in the dose of papaya leaf extract in the tested group.

CONCLUSION

The results of this study showed that the administration of papaya leaf extract at a dose of 150mg/Kg

and 200mg/Kg for 14 days has a small effect in reducing 2-hour postprandial blood sugar levels in the blood of male Wistar rats.

REFERENCES

I. Sapra A, Bahndari P. Diabetes. StatPearls [Internet]. 2023; Available from: https://www.ncbi.nlm.nih.gov/books/NBK551501/

II. IDF. International Diabetes Federation Atlas. 10th ed. Vol. 102, Diabetes Research and Clinical Practice. 2021. 147–148 p.

III. Kardika IBW, Herawati S, Yasa IWPS. Preanalitik dan Interpretasi Glukosa Darah untuk Diagnosis Diabetes Melitus. Univ Udayana. 2018;1–14.

IV. Trerattanavong K, Tadi P. Glimepiride. StatPearls. 2023;

V. Basit A, Riaz M, Fawwad A. Glimepiride: Evidence-based facts, trends, and observations. Vasc Health Risk Manag. 2012;8(1):463–72.

VI. Hariono M, Julianus J, Djunarko I, Hidayat I, Adelya L, Indayani F, et al. The future of carica papaya leaf extract as an herbal medicine product. Molecules. 2021;26(22).

VII. Hariyono P, Patramurti C, Candrasari DS, Hariono M. An integrated virtual screening of compounds from Carica papaya leaves against multiple protein targets of SARS-Coronavirus-2. Results Chem [Internet]. 2021 Jan;3(January). Available from: https://linkinghub.elsevier.com/retrieve/pii/S2211715621000187

VIII. Behl T, Gupta A, Albratty M, Najmi A, Meraya AM, Alhazmi HA, et al. Alkaloidal Phytoconstituents for Diabetes Management: Exploring the Unrevealed Potential. Molecules. 2022;27(18).

IX. Widiastuti TC, Rahayu TP, Lestari A, Kinanti AP. Uji Aktivitas Antidiabetes Kombinasi Ekstrak Terstandar Daun Salam (Syzigium polyanthum Walp.) dan Daun Ganitri (Elaeocarpus ganitri Roxb.) Pada Tikus Putih (Rattus norvegicus) Jantan Galur Wistar yang Diinduksi Streptozotosin. JPSCR J Pharm Sci Clin Res. 2023;8(1):92.

X. Al-Ishaq RK, Abotaleb M, Kubatka P, Kajo K, Büsselberg D. Flavonoids and their anti-diabetic effects: Cellular mechanisms and effects to improve blood sugar levels. Biomolecules. 2019;9(9).

XI. Palupi DA, Armita II, Sugiarti L. Pengaruh Pemberian Kombinasi Ekstrak Daun Pepaya(Carica Papaya L.) Dan Aktivitas Fisik Terhadap Kadar Glukosa Darah Mencit Diabetes Melitus Tipe II. Cendekia J Pharm. 2023;7(1):89–96.

XII. Husna F, Suyatna FD, Arozal W, Purwaningsih EH. Model Hewan Coba pada Penelitian Diabetes. Pharm Sci Res [Internet]. 2019 Dec 31;6(3):131–41. Available from: https://scholarhub.ui.ac.id/psr/vol6/iss3/1/

XIII. Cruz PL, Moraes-Silva IC, Ribeiro AA, Machi JF, de Melo MDT, dos Santos F, et al. Nicotinamide attenuates streptozotocin-induced diabetes complications and increases survival rate in rats: role of autonomic nervous system. BMC Endocr Disord. 2021;21(1):1–10.

XIV. Kamal S, Margono, Hidayah N, Rohmayanti, Luthfiyati H. Dosis Streptozotocyn Mempengaruhi Mortalitas Mencit Balb-C Dalam Proses induksi Hewan Model Diabetes Mellitus. Univ Res Colloq. 2017;1–6.

XV. Szkudelski T. The mechanism of alloxan and streptozotocin action in B cells of the rat pancreas. Physiol Res. 2001;50(6):537–46.

XVI. Sartika L, Rahardiantini I. Pengaruh Glimepirid terhadap Penurunan Glukosa Darah pada Mencit Diabetes-Disfungsi Ginjal. J Ilm Medicam. 2022;8(2):104–9.

XVII. Zhu H, Zhu S, Zhang X, Guo Y, Shi Y, Chen Z, et al. Comparative efficacy of glimepiride and metformin in monotherapy of type 2 diabetes mellitus: Meta-analysis of randomized controlled trials. Diabetol Metab Syndr. 2013;5(1):1–11.

XVIII. Ismukada TISBSDR. Antidiabetic activity of papaya leaf extract (Carica Papaya L.) isolated with maceration method in alloxan-induces diabetic mice. Syst Rev Pharm. 2020;11(9):774–8.

XIX. Pudyawanti PE, Astuti MD, Adhie NR, Hidayat IW. Ektrak Daun Pepaya Sebagai Anti DM Tipe 2. Pros APC (Annual Pharm Conf. 2018;3(1):97–102.