NUSANTARA BIOSCIENCE
Vol. 15, No. 2, pp. 143-148
November 2023

ISSN: 2087-3948
E-ISSN: 2087-3956
DOI: 10.13057/nusbiosci/n150202

Effectiveness test of *Diploknema oligomera* extracts to the decrease in glucose levels in alloxan-induced BALB/c male mice

SITI FAIZAH, KARTINI ERIANI♥, ROSNIZAR
Department of Biology, Faculty of Mathematics and Natural Sciences, Universitas Syiah Kuala. Jl. Syech Abdur Rauf No.3, Banda Aceh 23111, Aceh, Indonesia. Tel.: +62-651-741-0248, ♥email: kartini_eriani@unsyiah.ac.id

Manuscript received: 30 December 2022. Revision accepted: 4 June 2023.

Abstract. *Faizah S, Eriani K, Rosnizar. 2023. Effectiveness test of* Diploknema oligomera *extracts to the decrease in glucose levels in alloxan-induced BALB/c male mice. Nusantara Bioscience 15: 143-148.* People with diabetes mellitus usually consume chemical drugs to lower their blood glucose levels which cause side effects and require a reasonably high cost. Therefore, alternative natural medicines (phytopharmaceuticals) that are safer and more affordable are needed. Punti fruit (*Diploknema oligomera* H.J Lam) has the potential as an antihyperglycemic agent that can lower glucose levels in the blood because the punti fruit contains flavonoid compounds, tannins, terpenoids, and saponins. These secondary metabolites can reduce blood glucose levels. This research used an experimental method with a completely randomized design, six treatments, and four replications. This study used 24 BALB/C (*Mus musculus* Linnaeus, 1758) male mice grouped into six treatments, namely normal control treatment (P1), negative control by giving alloxan 150 mg/kg bw (P2), positive control by giving glibenclamide dose of 0.65 mg/kg body weight (bw) (P3), ethanol extract of punti fruit dose of 250 mg/kg bw (P4), ethanol extract of punti fruit dose of 500 mg/kg body weight (P5) and ethanol extract of punti fruit dose of 1,000 mg/kg bw (P6). The results proved that administering punti extract (P3, P4, P5) for 14 could reduce blood glucose levels in male diabetic mice with a percentage reduction of 33.6%, 37.4%, and 52.7%, and treatment-positive control decreased blood glucose by 32.6%. The conclusion showed that the ethanol extract of punti fruit at a dose of 1,000 mg/kg bw could effectively reduce blood glucose levels in diabetic mice compared to a dose of 250 mg/kg bw, 500 mg/kg bw and 0.65 mg/kg glibenclamide kg bw. Therefore, punti fruit can be used as a drug candidate to be developed as phytopharmaceuticals in treating diabetes mellitus.

Keywords: Antioxidant, blood glucose, diabetes mellitus, hyperglycemia, hypoglycemia

INTRODUCTION

Hyperglycemia is one of the causes of Diabetes Mellitus (DM). It is a degenerative disease that causes the most deaths in the world. Based on the International Diabetes Federation (IDF), there were 425 million people with diabetes worldwide in 2007, and Indonesia was ranked seventh with 10.7 million DM sufferers in 2019. The number increased by 19.5 million in 2021 and was ranked fifth worldwide (IDF 2021; Sun et al. 2022); hyperglycemia of DM sufferers is caused by diet. Widiyanto and Rahayu's (2019) research result on 17 respondents at the Sidomulyo Health Center in Pekanbaru indicated that 51.5% of respondents had irregular eating patterns. Rahati et al. (2014) stated that consuming foods containing excess carbohydrates has a high risk of diabetes mellitus. The high consumption of carbohydrates results in a buildup of glucose in the blood (Hawash et al. 2020), which causes the pancreas to work harder to produce insulin (Rahman et al. 2021). If this happens continuously, the pancreas will be damaged, and insulin cannot be produced optimally. These conditions lead to diabetes mellitus (Feinman et al. 2015).

Moreover, people with diabetes mellitus must lower their blood glucose levels by consuming chemical drugs, such as glibenclamide, acarbose, and metformin. However, metformin has side effects: impaired liver and kidney function and digestive disorders, such as flatulence, vomiting, abdominal pain, diarrhea, and constipation (Ghosal and Ghosal 2019). Likewise, glibenclamide's side effects include diarrhea, nausea, vomiting, and constipation (Rani et al. 2014). In addition, chemical drugs take longer and are quite expensive to treat hyperglycemia. Therefore, alternative therapeutic solutions from plants that are safer, more affordable, and effective in dealing with hyperglycemia in diabetes mellitus patients are needed.

One of the plants with the potential to an antihyperglycemic is punti (*Diploknema oligomera* H.J. Lam). Punti contains secondary metabolites, such as phenolic compounds, flavonoids, saponins, tannins, and terpenoids (Furqan and Putri 2020). These secondary metabolites can lower blood glucose (Lisdiani et al. 2022) by increasing insulin secretion (Martin and Ramos 2021) and/or regenerating pancreatic β cells (Ghorbani et al. 2019). In addition, these secondary metabolites also lower blood glucose by increasing insulin sensitivity through the stimulation of insulin receptors (Xu et al. 2018) on insulin target cells, such as adipose cells (Kang and Chiang 2022), liver cells, and muscle cells (Hu et al. 2014).

Several studies have investigated various plants' activity with secondary metabolites in reducing blood glucose levels. For example, Njogu et al. (2018) reported that *Kigelia africana* (Lam.) Benth. leaves have hypoglycemic activity due to phytochemicals detected in leaf powder.

These phytochemical compounds include phenolics, saponins, tannins, flavonoids, and alkaloids. Likewise, Morada et al. (2016) found that tannins extracted from *Sonneratia alba* Sm. leaves (16 mg/g body weight) were given to hyperglycemic mice for 6 hours and 12 hours; it showed a significant decrease in blood glucose levels (39,6% and 56,4%). Meanwhile, Nardia et al. (2020) reported the ethanol extract of *Persea americana* Mill. seeds contain polyphenols, tannins, flavonoids, triterpenoids, quinones, monoterpenoids, and sesquiterpenoids. Furthermore, these phytochemicals proved in these secondary metabolites could lower blood glucose in diabetic mice.

Another study conducted by Song et al. (2022) revealed that terpenoid compounds extracted from the bark of *Dillenia indica* L could stimulate insulin receptors so that they have the potential as antihyperglycemic agents. Meanwhile, research by Latief et al. (2021) showed that the ethanol extract of Sungkai leaves (*Peronema canescens* Jack) contains alkaloids, steroids, terpenoids, flavonoids, saponins, tannins, and phenolics. These phytochemicals could reduce glucose levels by inhibiting the α-glucokinase enzyme, increasing antioxidant enzyme activities, and regenerating pancreatic β cells. It is in line with research by Yuliana et al. (2021) that the ethanol extract of *karamunting* (*Rhodomyrtus tomentosa* (Aiton) Hassk.) leaves could reduce blood glucose levels in diabetic rats. The *karamunting* leaves contain phenolic compounds, such as flavonoids, saponins, tannins, steroids, and triterpenoids (Dona et al. 2020). Matusin et al. (2021) examined the methanol extract of Dillenia excelsa leaves. These leaves have antihyperglycemic properties because of the triterpenoid compound effects: flavonoids, tannins, and phenolics (Lisdiani et al. 2022). Meanwhile, Furqan and Putri's (2020) found that the ethanol extract of punti contains flavonoids, terpenoids, phenolics, tannins, and saponins. However, punti as an antihyperglycemic agent has not been studied scientifically. Therefore, research on punti (*D. oligomera*) as an antihyperglycemic is paramount so that the findings can later provide information regarding the treatment of diabetes mellitus.

MATERIALS AND METHODS

Sample extractions

Punti powder of 150 g was soaked in one liter of 96% ethanol and then allowed to stand for three days in a glass container protected from light. Then it was filtered with filter paper to get the filtrate. The filtrate was then concentrated using a rotary evaporator at 60°C for ± 3 hours.

Management of experimental animals

The experimental animals used in this study were 24 BALB/c male mice weighing 20-30 g. All experimental animals were given water and all feed-4 based on body weight (ad libitum) and placed in cages 20 cm x 13 cm x 10 cm covered with sawdust. The cage was enclosed with iron wire measuring 35 cm x 35 cm and cleaned every two days.

Diabetes induction in experimental animals

Before intraperitoneal induction of alloxan monohydrate (Sigma-Aldrich, USA), BALB/c male mice fasted for 24 hours. Alloxan with a 150 mg/kg BW dose was dissolved in 1 mL of 0.9% NaCl (Widastuti et al. 2017). After 72 hours of induction, the blood glucose levels of BALB/c male mice were checked with the EasyTouch GCU. The positive control and treatment groups included blood glucose values above > 200 mg/dl.

Experimental animal treatments

Experimental animals were divided into six groups consisting of four BALB/c male mice. The normal control group was given water and all feed-4 (P1). In comparison, alloxan monohydrate (P2) induced the negative control group. The positive control group was induced by alloxan monohydrate and glibenclamide administration of 0.65 mg/kg bw (P3). Groups P4, P5, and P6 were induced by alloxan and given punti extracts orally at a dose of 250 mg/kg bw for P4, 500 mg/kg bw for P5, and 1,000 mg/kg bw for P6. All treatments were carried out at 10.00 WIT for 14 days.

Measurement of glucose levels

Blood samples were taken by cutting a small part of BALB/c male mice's tails. The tails were cleaned first with cotton given 70% alcohol. Then, the blood from the tail vein was dripped onto the strips attached to the Easy Touch GCU glucometer. Blood measurements were carried out on days 0, 4, 11, and 18. Blood sugar levels on day zero were glucose levels after fasting for 16 hours before alloxan was induced. Meanwhile, on the fourth day, it was diabetic blood sugar levels after alloxan induction and recovery blood sugar levels after treatment of punti extracts and glibenclamide administration on the 11^{th} and 18^{th} days.

Data analysis

Blood glucose levels obtained were analyzed using the SPSS 25 software with a one-way Analysis of Variance (ANOVA) test. The ANOVA test aims to determine differences in blood glucose levels between treatments. If there is a significant difference, proceed with Duncan's multiple range test at a 5% confidence level (Eriani et al. 2021).

RESULTS AND DISCUSSION

The results of the 6 treatments showed blood glucose levels in mice for 18 days of treatment, as shown in Figure 1. After 72 hours of alloxan induction, the blood glucose examination with a dose of 150 mg/kg body weight to the treatment (P2, P3, P4, P5, and P6) showed that blood glucose levels increased by > 200 mg/dL on the 4^{th} day. The highest increase in blood glucose levels was found in the negative control treatment on day 11 and day 18, with an average of 442.5 ± 66.28 mg/dL and 455.50 ± 38.46 mg/dL. Duncan's 5% confidence level test showed that the

average glucose levels on day 11 and day 18 in the negative control treatment were significantly different from the other treatments (Table 1).

The positive control treatment with glibenclamide of 0.65 mg/kg in diabetic mice showed a decrease in blood glucose levels of 401.75 ± 58.60 mg/dL on day 11 and continued to decrease to 269.75 ± 38.87 mg/dL on day 18, reduced by 32.9%. In addition, the results of Duncan's multiple range test with a 5% confidence level showed that the average blood glucose levels on day 11 and day 18 in the positive control treatment significantly differed from the negative control treatment (Table 1).

The examination of blood glucose levels in diabetic mice after adding punti fruit ethanol extract in the treatments (P4, P5, and P6) showed a decrease in blood glucose levels on the 11th and 18th days by 33.6%, 37.4%, and 52.7%, respectively. The percentage value of decreasing blood glucose was measured by subtracting the average blood glucose levels from day 11 to day 18. The reduction results were then divided by 100% to obtain the percentage value of decreasing blood glucose levels (Chandrika et al. 2006). The Duncan test with a 5% confidence level proved that the punti fruit ethanol extract treatment (P4, P5, P6) significantly differed from the negative control treatment (Table 1).

The admission of punti ethanol extract to diabetic mice in treatment (P6) for 14 days showed a higher decrease by 52.7% compared to the positive control treatment (32.9%). For other treatments (P4 and P5), it was 33.6% and 37.4%. These percentages indicated treatment 6 had the highest blood glucose reduction compared to the positive control treatment (P4 and P5). The Duncan test with a 5% confidence level also revealed that, on average, blood glucose levels at P6 significantly differed from the positive control treatments (P4 and P5) (Table 1).

Discussion

This study proved that mice induced with alloxan starting from 150 mg/kg showed an increase in blood glucose levels >200 mg/dL on the 4th day. That aligns with Widastuti et al. (2017) reported that alloxan at 150 mg/kg increased blood glucose levels in mice >200 mg/dL on day 4. The increase in blood glucose levels is due to the alloxan mechanism. Alloxan's structure is similar to glucose; it can enter pancreatic β cells via glucose transporter 2 (GLUT2) (Berraaouan et al. 2015). In addition, alloxan also perforates the pancreatic β cells through the cell membrane because the structure of alloxan consists of substituted N atoms bound to lipophilic alkyl groups, easily penetrating all parts of the cell membrane. Furthermore, the lipophilic nature of alloxan leads to free radical toxicity (Ighodaro et al. 2017), which occurs through Reactive Oxygen Species (ROS). ROS undergo dismutation into hydrogen peroxide (H_2O_2) and hydroxyl radicals (*OH). Hydroxyl radicals are more reactive, so these radicals destroy pancreatic β cells (Eguchi et al. 2021), leading to reduced insulin production (Udia et al. 2016) and hyperglycemia (Abdulgani et al. 2014).

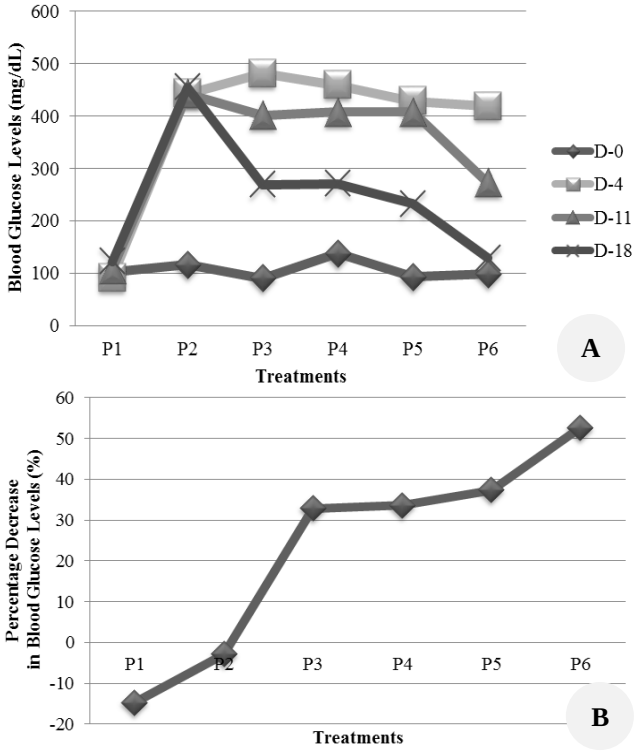

Figure 1. Blood glucose levels of 1 to 6 treatments A. Average blood glucose levels during the 18 days of treatment, B. Percentage reduction in average blood glucose levels from day 11 to day 18

Table 1. Analysis of average blood glucose levels in BALB/c male mice during the treatments

Treatments	Blood glucose levels (mg/dL) ($\bar{x}$ ± SD)				Decrease %
	0 day	4th day	11th day	18th day	
P1	102.75 ± 16.68	91.750 ± 22.90	107.25 ± 27.60^{a}	123.00 ± 27.30^{a}	-14.68
P2	116.25 ± 30.33	443.00 ± 77.59	442.50 ± 66.28^{c}	455.50 ± 38.46^{c}	-2.9
P3	90.00 ± 7.300	481.25 ± 34.1	401.75 ± 58.60^{b}	269.75 ± 38.87^{b}	32.9
P4	137.75 ± 42.5	458.75 ± 96.2	408.00 ± 90.80^{b}	270.75 ± 81.70^{b}	33.6
P5	93.50 ± 28.55	428.50 ± 26.51	408.00 ± 90.80^{b}	233.00 ± 44.02^{b}	37.4
P6	98.50 ± 11.26	418.75 ± 9.630	272.75 ± 78.40^{a}	129.00 ± 59.50^{a}	52.7

Note: Numbers followed by different letters in the same column state significant difference at α of 0,05 (Duncan). P1 = Normal control, P2 = Negative control, P3 = Positive control, P4 = Mice given *punti* fruit extract 250 mg/kg BW, P5 = Mice given punti fruit extract 500 mg/kg BW, P6 = Mice given punti fruit extract 1,000 mg/kg BW

Adding glibenclamide to diabetic mice in the positive control treatment decreased the average blood glucose level due to its mechanism. Glibenclamide could lower blood glucose through sulfonylurea 1 (SUR1) receptor inhibition, which closes the ATP-sensitive potassium channels (KATP) in the pancreatic β cell membrane. That causes depolarization of the pancreatic β-cell membrane and Ca^{2+} channels opening. As a result, ca^{2+} ions enter the cell and increase the intracellular Ca^{2+} ions. The increasing Ca^{2+} in pancreatic β cells will stimulate insulin-carrying granules; therefore, insulin is released, and blood glucose decreases (Yau et al. 2021).

In this study, punti fruit was extracted using ethanol solvent. According to Yusnawan (2013), using polar solvents (ethanol) for maceration can produce a diverse group of secondary metabolites. Furthermore, polar solvents include alcohol groups generally used to extract secondary metabolites in plants. Polar solvents can increase cell permeability and penetrate the cell, resulting in extraction of more endocellular secondary metabolites, both polar and less polar compounds, than non-polar solvents (Seidel 2012). Research by Azmir et al. (2013) reported that ethanol, as a polar solvent, can attract polyphenolic compounds, tannins, and terpenoids. Meanwhile, research conducted by Hikmawanti et al. (2021) proved that ethanol solvent could attract phenolic and flavonoid compounds in the *katuk* leaf plant (*Sauropus androgynus* (L.) Merr.). Likewise, Furqan and Putri's research (2020) reported that ethanol solvents produced saponin, tannin, phenolic, and flavonoid compounds in punti fruit plants.

The administration of punti ethanol extract to diabetic mice in the treatments (P4, P5, and P6) reduced the average blood glucose level. That is presumably due to the effects of the flavonoids, terpenoids, saponins, and phenolic compounds in the ethanol extract of punti. Xu et al. (2018) reported that saponin compounds could increase insulin sensitivity through the stimulation of insulin receptors (insulin receptor substrate-1 (IRS-1). IRS-1 stimulation activates phosphoinositide 3-kinase (PI3K) phosphorylation and protein kinase B (PKB)/AKT activation. PKB/AKT activation can also be done by phenolic flavonoids (Deng et al. 2020) and terpenoids (Song et al. 2022). Hu et al. (2014) argued that IRS-1 stimulation with PI3K and AKT activation could cause GLUT4 translocation to cell membranes, increasing glucose uptake in liver and skeletal muscle cells and decreasing blood glucose.

Saponins and phenolic compounds also lower blood glucose by stimulating adenosine 5′ monophosphate-Activated Protein Kinase (AMPK), causing glucose absorption in adipose tissue and affecting blood glucose reduction (Xu et al. 2018; Kang and Chiang 2022). Meanwhile, tannin lowers blood glucose with its ability to inhibit glucose-6-phosphatase, increases glucose absorption in hepatocyte cells and myoblast cells, and acts as an inhibitor of the α-glucosidase enzyme that can delay glucose absorption in the small intestine wall (Sheikh et al. 2019). Saponin and flavonoids also can inhibit the α-glucosidase enzyme (Sohretoglu et al. 2018; Nabil et al. 2019).

Flavonoid also increases insulin secretion (Khedher et al. 2018) through the effect of regenerating pancreatic β cells (Ghorbani et al. 2019), which in turn lowers blood glucose (Kasmawati et al. 2019). Flavonoids carry out the regeneration mechanism of pancreatic β cells from the flavone group. Flavones consist of rings C (C2, C3) and B (C4), which are thought to stimulate glucagon-like peptide-1 (GLP-1) (Wang et al. 2018). The activation of GLP-1 leads to PI3K and AKT phosphorylation (Campbell and Drucker 2013). In addition, Phosphorylated AKT will increase the expression of the pancreatic duodenum homeobox-1 (PDX-1) gene. This gene can regulate the regeneration of pancreatic β cells through the processes of proliferation, differentiation, and transdifferentiation (Seeberger et al. 2014; Guney and Lorberbaum 2020). In addition, the pancreatic β cell regeneration activity can increase the amount of insulin secretion, decreasing blood glucose (Campbell and Drucker 2013).

Furthermore, phenolic can act as an antioxidant by protecting pancreatic β cells from damage caused by alloxan (Adiba et al. 2021). Phenolic has hydroxyl groups of one or more aromatic ring structures (Singh et al. 2015). Based on the number of hydroxyl groups, it is grouped into several compounds, such as tannins and flavonoids (Singh et al. 2016). Phenolic compounds, such as flavonoids with hydroxyl groups (OH^-), can donate hydrogen atoms (H) to stabilize free radicals (Vo et al. 2019). In addition, free radicals are unstable and highly reactive compounds (Kia et al. 2020) that can cause cell damage (Yu et al. 2012). Phenolic can deactivate free radicals, stopping damage to pancreatic β cells (Hasanah 2016) and decreasing blood glucose levels (Golovinskaia and Wang 2023).

The Duncan test results with a 5% confidence level in the punti ethanol extract treatment (P6) showed a significant difference from other treatments (P4 and P5). The content of secondary metabolites in P4 (dose 250 mg/kg bw) and P5 (500 mg/kg bw) was at a low concentration to improve the performance of pancreatic β cells so that the reduction in blood glucose levels had not yet reached the maximum effect. Nasution (2015) argued that administering the dose required an optimal condition to achieve the desired effect. Thus, the dose of punti ethanol extract to best reduce blood glucose levels is 1,000 mg/kg body weight.

The Duncan test with a 5% confidence level on the average blood glucose level in the punti ethanol extract treatment (P6) and the positive control treatment (P3) also indicated a significant difference. Glibenclamide only works to lower blood glucose (Liem et al. 2015) through the release of insulin by pancreatic β cells (Yau et al. 2021) without increasing the regeneration of pancreatic β cells (Ibrahim et al. 2021).

In conclusion, the ethanol extract of punti of 1,000 mg/kg body weight improved blood glucose levels in diabetic mice by 52.7% compared to Glibenclamide administration (32.9%). Therefore, the ethanol extract of punti at a dose of 1,000 mg/kg BW works more optimally in lowering blood glucose than glibenclamide. This study's findings prove that punti can be used as a drug candidate in treating diabetes mellitus.

ACKNOWLEDGEMENTS

The authors are grateful to Indonesia's Ministry of Education, Culture, Research and Technology of the Republic of Indonesia for providing grants for Masters Thesis Research in 2022 with contract number 83/UN11.2.1/PT.01.03/DPRM/2022.

REFERENCES

Abdulgani N, Trisnawati I, Aunurohim, Hidayati D, Aisyatussoffi N, Arifiyanto A. 2014. Snakehead (*Channa striata*) extracts treatment towards hyperglycemic mice (*Mus musculus*) blood glucose levels and pancreatic histology structure. J Appl Environ Biol Sci 4: 1-6.

Adiba HMP, Arifullah M, Nazahatul AA, Sirikitputtisak T, Klaiklay S, Chumkaew P, Chewchanwuttiwong S, Norhazlini MZ, Zulhazman H. 2021. Total phenolic content and antioxidant activity of limestone endemic Araceae species, *Alocasia farisii*. Earth Environ Sci 842: 1-6. DOI: 10.1088/1755-1315/842/1/012064.

Azmir J, Zaidul ISM, Rahman MM, Sharif KM, Mohamed A, Sahena F, Omar AKM. 2013. Techniques for extraction of bioactive compounds from plant materials: A review. J Food Eng 117 (4): 426-436. DOI: 0.1016/j.jfoodeng.2013.01.014.

Berraaouan A, Abderrahim Z, Hassane M, Abdelkhaleq L, Mohammed A, Mohamed B. 2015. Evaluation of protective effect of cactus pear seed oil (*Opuntia ficus-indica* L. Mill.) against alloxan induced diabetes in mice. Asian J Trop Med A 8: 532-537. DOI: 10.1016/j.apjtm.2015.06.013.

Campbell JE, Drucker DJ. 2013. Pharmacology, physiology, and mechanisms of incretin hormone action. Cell Metabolism 17: 819-837. DOI: 10.1016/j.cmet.2013.04.008.

Chandrika UG1, Wedage WS, Wickramasinghe SMDN1, Fernando WS. 2006. Hypoglycaemic action of the flavonoid fraction of *Artocarpus heterophyllus* leaf. Afr J Tradit 3: 42-50.

Deng N, Zheng B, Li T, Liu RH. 2020. Assessment of the phenolic profiles, hypoglycemic activity, and molecular mechanism of different highland barley (*Hordeum vulgare* L.) varieties. Intl J Mol Sci 21: 1175. DOI: 10.3390/ijms21041175.

Dona R, Furi M, Suryani F. 2020. Determination of total phenolic, flavonoid and antioxidant activity tests of extracts and fractions of karamunting (*Rhodomyrtus tomentosa* (Aiton) Hassk) leaves. Indon Pharm Res J 9: 72-78. DOI: 10.51887/jpfi.v9i2.975.

Eguchi N, Vaziri ND, Dafoe DC, Ichii H. 2021. The role of oxidative stress in pancreatic β Cell dysfunction in diabetes. Intl J Mol Sci 22: 1-18. DOI: 10.3390/ijms22041509.

Eriani K, Hasanah U, Fitriana R, Sari W, Yunita Y, Al Azhar. 2021. Antidiabetic potential of methanol extract of flamboyant (*Delonix regia*) flowers. Biosaintifika 13: 185-194. DOI: 10.15294/biosaintifika.v13i2.30080.

Feinman RD, Pogozelski WK, Astrup A, Bernstein RK, Fine EJ, Westman EC, Accurso A, Frassetto L, Gower BA, McFarlane SI, Nielsen JV, Krarup T, Saslow L, Roth KS, Vernon MC, Volek JS, Wilshire GB, Dahlqvist A, Sundberg R, Childers A, Morrison K, Manninen AH, Dashti HM, Wood RJ, Wortman J, Worm N. 2015. Dietary carbohydrate restriction as the first approach in diabetes management: Critical review and evidence base. Nutrition 31: 1-13. DOI: 10.1016/j.nut.2014.06.011.

Furqan M, Putri NA. 2020. Isolation of phenolic compounds from punti fruit (*Diploknema oligomera* H.J.Lam) by FT-IR analysis. J Health Care Technol Med 6: 2615-109. DOI: 10.33143/jhtm.v6i2.1198.

Ghorbani A, Rashidia R, Nick RS. 2019. Flavonoids for preserving pancreatic beta cell survival and function: A mechanistic review. Biomed Pharm 111: 947-957. DOI: 10.1016/j.biopha.2018.12.127.

Ghosal S, Ghosal S. 2019. The side effects of metformin - a review. Diabetes Metab Disord 6: 1-7. DOI: 10.24966/DMD-201X/100030.

Golovinskaia O, Wang CK. 2023. The hypoglycemic potential of phenolics from functional foods and their mechanisms. Food Sci Human Well 12: 986-1007. DOI: 10.1016/j.fshw.2022.10.020.

Guney MA, Lorberbaum DS. 2020. Pancreatic β cell regeneration: To β or not to β. Curr Opin Physiol 14: 13-20. DOI: 10.1016/j.cophys.2019.10.019.

Hasanah U. 2016. Profil of beta pancreatic cell in rats are given kimpul (*Xanthosoma sagittifolia* (L.) Schott.). [Thesis]. Universitas Sebelas Maret, Surakarta, Indonesia.

Hawash ZAE, Farrag AA. Eldamaty HSE. 2020. Effect of using sweet potato powder on diabetic rats. J Home Econ 36: 1-18. DOI: 10.21608/jhe.2020.109290.

Hikmawanti NPE, Fatmawati S, Asri AW. 2021. The effect of ethanol concentrations as the extraction solvent on antioxidant activity of katuk (*Sauropus androgynus* (L.) Merr.) leaves extracts. IOP Conf Ser Earth Environ Sci 755 (1): 012060. DOI:10.1088/1755-1315/755/1/012060.

Hu X, Wang S, Xu J, Wang DB, Chen Y, Yang GZ. 2014. Triterpenoid saponins from *Stauntonia chinensis* ameliorate insulin resistance via the AMP-activated protein kinase and IR/IRS-1/PI3K/Akt pathways in insulin-resistant HepG2 cells. Intl J Mol Sci 15: 10446-10458. DOI: 10.3390/ijms150610446.

Ibrahim KG, Audu BB, Mainasara AS, Abubakar MB. 2021. Effect of Co-administration of glibenclamide and aqueous calyx extract of Hibis*cus sabdariffa* on oxidative stress markers in Streptozotocin-Induced diabetic rats. Niger J Physiol Sci 36: 77-84.

Ighodaro OM, Adeosun AM, Akinloye OA. 2017. Alloxan-induced diabetes, a common model for evaluating the glycemic-control potential of therapeutic compounds and plants extracts in expereimental studies. J Med 53: 365-374. DOI: 10.1016/j.medici.2018.02.001.

International Diabetes Federation (IDF). 2021. IDF Diabetes Atlas (10th edition). Diabetes Atlas, Belgium. https://diabetesatlas.org/.

Kang BB, Chiang BH. 2022. A novel phenolic formulation for treating hepatic and peripheral insulin resistance by regulating GLUT4-mediated glucose uptake. J Tradit Complement Med 12: 195-205. DOI: 10.1016/j.jtcme.2021.08.004.

Kasmawati H, Ruslin, Ihsan S, Hasnawati, Suryani, Pertama BA, Farida SA. 2019. Antidiabetic activity of ethanol extract of soni leaves (*Dillenia celebica* Hoogland.) on blood glucose levels and pancreatic histology of male wistar rats glucose-induced. Indo Am J Pharm Sci 6: 13649-13659. DOI: 10.5281/zenodo.3346991.

Khedher MRB, Hammami M, Arch JRS, Hislop DC, Eze D, Wargent ET, Kępczyńska MA, Zaibi MS. 2018. Preventive effects of *Salvia officinalis* leaf extract on insulin resistance and inflammation in a model of high fat diet induced obesity in mice that responds to rosiglitazone. Peer J 9: 14-22. DOI: 10.7717/peerj.4166.

Kia FJ, Wibowo JP, Elachouri M, Masumi R, Jouneghani SJ, Abolhassanzadeh Z, Lorigooini Z. 2020. Battle between plants as antioxidants with free radicals in human body. J Herbmed Pharmacol 9: 191-199. DOI: 10.34172/jhp.2020.25

Latief M, Sari PM, Fatwa LT, Tarigan IL, Rupasinghe HPV. 2021. Antidiabetic activity of *sungkai* (*Peronema canescens* Jack) leaves ethanol extract on the male mice induced alloxan monohydrate. Pharmacol Clin Pharm Res 6: 64-74. DOI:10.15416/pcpr.v4i3.31666.

Liem S, Yuliet, Khumaidi A. 2015. Antidiabetic activity test of combination of glibenclamide and bay leaf extract (*Syzygium polyanthum* wight.) To alloxan induced mice (*Mus musculus*). Galenika J Pharm 1: 42-47. DOI: 10.22487/j24428744.2015.v1.i1.4831.

Lisdiani, Susanto D, Manurung, H. 2022. Phytochemical screening and antioxidant activity of methanol extract of *Dillenia excelsa* leaf. Biodiversitas 23: 3827-3835. DOI: 10.13057/biodiv/d230760.

Martin MA, Ramos S. 2021. Dietary flavonoids and insulin signaling in diabetes and obesity. J Cells 10: 2-22. DOI: 10.3390/cells10061474.

Matusin AHA, Ghani N IA, Ahmad N. 2021. Pancreatic islet regenerative capability of *Dillenia excelsa* in alloxan-induced diabetic rats. J Appl Pharm Sci 11: 121-129. DOI:10.7324/JAPS.2021.110315.

Morada NJ, Metillo EB, Uy MM, Oclarit JM. 2016. Toxicity and hypoglycemic effect of tannin-containing extract from the mangrove tree *Sonneratia alba* Sm. Bull Environ Pharmacol Life Sci 5: 58-64.

Nabil M, Ghaly NS, Kassem IAA, Grace MH, Melek FR. 2019. Two triterpenoid saponins with α-glucosidase inhibitory activity from *Harpullia pendula* seed extract. Pharmacog J 11: 1386-1390. DOI: 10.5530/pj.2019.11.214.

Nardia L, Listerb IN, Girsang E, Fachriald E. 2020. Hypoglycemic effect of avocado seed extract (*Persean americana* Mill) from analysis of oral glucose tolerance test on *Rattus norvegicus* L. Am Sci Res J Eng Technol Sci 65: 2313-4410.

Nasution A. 2015. Clinical Pharmacokinetics. USU Press, Medan. [Indonesian]

Njogu SM, Arika WM, Machocho AK, Ngeranwa JJN, Njagi ENM. 2018. In vivo hypoglycemic effect of *Kigelia africana* (Lam): Studies with alloxan-induced diabetic mice. J Evid-Based Integr Med 23: 1-10. DOI: 10.1177/2515690X18768727.

Rahati S, Shahraki M, Arjomand G, Shahraki T. 2014. Food pattern, lifestyle and diabetes mellitus. Intl J High Risk Behav Addict 10: 1-5. DOI: 10.5812/ijhrba.8725.

Rahman MDS, Hossain KS, Das S, Kundu S, Adegoke EO, Rahman MDA, Hannan MDA, Uddin MDJ, Pang MG. 2021. Role of insulin in health and disease: An update. Intl J Mol Sci 22: 2-19. DOI: 10.3390/ijms22126403.

Rani M, Yadav S, Choudhary S, Sharma S, Pandey SM. 2014. Incidence of hypoglycemia and other side effects in patients of type 2 diabetes mellitus treated with glimepiride versus glibenclamide. Intl J Health Sci Res 4: 68-72.

Seeberger KL, Anderson SJ, Ellis CE, Yeung TY, Korbutt GS. 2014. Identification and differentiation of PDX1 β-cell progenitors within the human pancreatic epithelium. World J Diabetes 1: 59-68. DOI:10.4239/wjd.v5.i1.59.

Seidel V. 2012. Initial and bulk extraction of natural product isolation. Methods Mol Biol 864: 27-41. DOI: 10.1007/978-1-61779-624-1_2.

Sheikh Y, Chanu MB, Mondal G, Manna P, Chattoraj A, Deka DC, Talukdara NC, Borah JC. 2019. Procyanidin A2, an anti-diabetic condensed tannin extracted from *Wendlandia glabrata*, reduces elevated G-6-Pase and mRNA levels in diabetic mice and increases glucose uptake in CC1 hepatocytes and C1C12 myoblast cells. Royal Chem Soc 9: 17211-17219. DOI: 10.1039/c9ra02397f.

Singh JP, Kaur A, Shevkani K, Singh N. 2015. Influence of jambolan *(Syzygium cumini)* and xanthan gum incorporation on the physicochemical, antioxidant and sensory properties of gluten-free eggless rice muffins. Intl J Food Sci Technol 50: 1190-1197. DOI: 10.1111/ijfs.12764.

Singh JP, Kaur A, Singh N, Nim L, Shevkani K, Kaur H, Arora DS. 2016. In vitro antioxidant and antimicrobial properties of jambolan *(Syzygium cumini)* fruit polyphenols. Food Sci Technol 65: 1025-1030. DOI:10.1016/j.lwt.2015.09.038.

Sohretoglu D, Sari S, Barut B, Özel A. 2018. Discovery of potent α-glucosidase inhibitor flavonols: Insights into mechanism of action through inhibition kinetics and docking simulations. Bioorganic Chem 79: 257-264. DOI: 10.1016/j.bioorg.2018.05.010.

Song BR, Alam MB, Lee SH. 2022. Terpenoid-rich extract of *Dillenia indica* L. bark displays antidiabetic action in insulin-resistant C2C12 cells and STZ-induced diabetic mice by attenuation of oxidative stress. Antioxidants 11: 2-19. DOI: 10.3390/antiox11071227.

Sun H, Saeedi P, Karuranga S, Pinkepank M, Ogurtsova K, Duncan, Bruce B. 2022. IDF diabetes atlas: Global, regional and country-level diabetes prevalence estimates for 2021 and projections for 2045. Diabetes Res Clin Pract 183: 1-13. DOI: 10.1016/j.diabres.2021.109119.

Udia PM, Takem LP, Ufot UF, Antai AB, Owu DU. 2016. Insulin and alpha amylase levels in alloxan-induced diabetic rats and the effect of *Rothmannia hispida* (K. Schum) Fagerl leaf extract. J Phytopharmacol 1: 1-5. DOI: 10.31254/phyto.2016.5101.

Vo QV, Nam PC, Thong NM, Trung NT, Phan CTD, Mechler A. 2019. Antioxidant Motifs in Flavonoids: O−H versus C−H Bond Dissociation. ACS Omega 4: 8935-8942. DOI: 10.1021/acsomega.9b00677.

Wang Y, Wang A, Alkhalidy H, Luo J, Moomaw E, Neilson AP, Liu D. 2018. Flavone hispidulin stimulates glucagon-like peptide-1 secretion and ameliorates hyperglycemia in streptozotocin-induced diabetic mice. J Mol Nutr Food Res 64: 1-12. DOI: 10.1002/mnfr.201900978.

Widastuti, Endang L, Sutyarso, Gregorius NS, Mahmud R, Mohammad K. 2017. Ameliorative properties of crude diosgenin from costus speciosus and taurine on testicular disorders in alloxan-induced diabetic mice. Biomed Pharmacol J 10: 09-17. DOI: 10.13005/bpj/1075.

Widiyanto J, and Rahayu S. 2019. Effect of diet on the incidence of diabetes mellitus at the Sidomulyo Inpatient Health Center, Pekanbaru City. Proceedings of ScienceTeKes, UMRi MIPAKes National Seminar 1: 1-7.

Xu J, Wang S, Feng T, Chen Y, Yang G. 2018. Hypoglycemic and hypolipidemic effects of total saponins from *Stauntonia chinensis* in diabetic db/db mice. J Cell Mol Med 22: 6026-6038. DOI: 10.1111/jcmm.13876.

Xu Y, Rashwan AK, Ge ZW, Li YT, Ge HJ, Li JX, Xie JH, Liu SY, Fang J, Cheng KJ. 2022. Identification of a novel α-glucosidase inhibitor from *Melastoma dodecandrum* Lour. fruits and its effect on regulating postprandial blood glucose. Food Chem 399: 133999. DOI: 10.1016/j.foodchem.2022.133999.

Yau B, Hocking S, Andrikopoulos S, Kebede MA. 2021. Targeting the insulin granule for modulation of insulin exocytosis. Biochem Pharmacol 194: 114821. DOI: 10.1016/j.bcp.2021.114821.

Yu Y, Fan SM, Song JK, Tashiro SI, Onodera S, Ikejima T. 2012. Hydroxyl radical (*OH) played a pivotal role in oridonin-induced apoptosis and autophagy in human epidermoid carcinoma A431 Cells. Biol Pharm Bull 35: 2148-2159. DOI: 10.1248/bpb.b12-00405.

Yuliana I, Asnawati , Ulfah M, Suhartono E. 2021. Antidiabetic potential of *karamunting* leaves ethanolic extract as a natural herb: Blood glucose levels and pancreatic islets histomorphology on diabetic rats model. Jurnal Kedokteran Dan Kesehatan Indonesia 12: 250-256. DOI: 10.20885/JKKI.Vol12.Iss3.art9.

Yusnawan E. 2013. The effectiveness of polar and non polar fractions of *Ageratum conyzoides* l. to control peanut rust disease and phytochemical screenings of secondary metabolites. Jurnal Hama dan Penyakit Tumbuhan Tropika 13: 159-166. [Indonesian]