Bali Medical Journal (*Bali Med J*) 2017, Volume 3, Number 3 (IBL Conference 2017 Special Issue): S113-S116
P-ISSN.2089-1180, E-ISSN.2302-2914

Hypoglycemic and antioxidant effects of *Syzygium polyanthum* leaves extract on alloxan induced hyperglycemic Wistar Rats

Sri Wahjuni,[1] I Wayan Wita[2]

ABSTRACT

Introduction: Hyperglycemia is a common signature of Diabetes Mellitus (DM) which could define as fasting blood glucose higher than 110 mg/dL or 2-hour post prandial blood glucose higher than 180 mg/dL. One of the consequences of hyperglycemia is increased rate of the oxidative process that could potentially damage many organs. The damage could be assessed by measuring 8-Hydroxy-2 Deoxyguanosine (8-OHdG) that represent the damage of the plasma membrane. In this study, we aimed to evaluate the efficacy of *Syzygium polyanthum* leaves extracts as antioxidant agent on hyperglycemic Wistar rats.

Methods: 40 Wistar rats were used in this study. Hyperglycemic state was achieved by administration of alloxan for two weeks for each rat. 3 Kg of *S.polyanthum* leaves was used for extraction which yields 0,730 ethanol extract. The rats were divided into positive control, P1 (alloxan only), P2 (Alloxan+ 0.5 mg kg^{-1} body weight Z polyanthum extract), P3 (Alloxan+ 2.0 mg kg^{-1} body weight Z polyanthum extract), P4 (Alloxan+ 5.0 mg kg^{-1} body weight *S.polyanthum* extract), P5 (Alloxan+ 0.18 mg/day/ kg body weight glibenclamide).

Result: The result shown that the rats which received *S.polyanthum* extract had significantly lower blood glucose level (65.91 % lower than control) and lower level of 8-OHdG (50.20% lower than control). The effective dose to optimally lower blood glucose and 8-OHdG was found at 5,0 mg kg^{-1}.

Conclusion: *S.polyanthum* ethanolic leaves extract was an effective anti-hypoglycemic and antioxidant agent with optimal dose found at 5,0 mg kg^{-1}.

Keywords: *Syzygium polyanthum leaves extract; blood glucose; 8-Hydroxy-2 Deoxyguanosine (8-OHdG)*
Cite This Article: Wahjuni, S., Wita, I.W. 2017. Hypoglycemic and antioxidant effects of *Syzygium polyanthum* leaves extract on alloxan induced hyperglycemic Wistar Rats. *Bali Medical Journal* 3(3): S113-S116. DOI:10.15562/bmj.v3i3.755

[1]Departement of Chemistry, Faculty of Mathematics and Natural Science Udayana University Campus Bukit Jimbaran street 80361 Bali Indonesia
[2]Clinical Internal Departement, Faculty of Medicine, Udayana University, PB. Sudirman Street Denpasar Bali Indonesia

*Corresponding Author: Sri Wahjuni, Departement of Chemistry, Faculty of Mathematics and Natural Science Udayana University Campus Bukit Jimbaran street 80361 Bali Indonesia
sriwahjunimanuaba@gmail.com

Received: 2017-07-17
Accepted: 2017-07-20
Published: 2017-07-20

INTRODUCTION

Diabetes mellitus (DM) is one of the most prevalent non-communicable diseases worldwide and contribute significantly to the chronic disease related mortality and morbidity.[1] The signature mark of DM is elevated blood glucose which is defined as fasting glucose level higher than 110 mg/dl or more than180 mg/dl for 2-hours post prandial glucose. Hyperglycemia has been proved to play a direct role in many of diabetic-related complication such as retinopathy, nephropathy, atherosclerosis, diabetic foot, and neuropathy.[2,3] The mechanisms that link hyperglycemia with these complications include induction of subclinical inflammation, the formation of glycation product, as well as increased production of free radicals which result in oxidative stress. Hyperglycemia increases free radicals productions through polyol pathway, hexosamine pathway, and by inflammation induced by advanced glycation end product (AGE). [4]

Current therapy of DM relies on hypoglycemic agents to lower blood glucose level which ranged from insulin sensitizing agents like metformin and thiazolidinedione to insulin and insulin secretagogue like sulfonylurea and glinid.[5] However, these agents only focus on lowering blood glucose and just have little or no effect on oxidative stress that directly results from hyperglycemia and the main mechanism that lead to the diabetic-related complications.[6] Some studies have shown that some agents like thiazolidinedione could increase internal antioxidant capacity, but these reports were still conflicting. Furthermore, its adverse effects such as hepatotoxicity also hinder its clinical application.[7]

Syzygium polyanthum is a plant that belongs to Myrtaceae family that commonly consumed in Indonesia and Malaysia. Several studies had proved that its extract holds anti-hyperglycemic potential that could be applied as a new therapy for DM. Furthermore, due to the rich tannins, flavonoids and terpenoids content, it also a potential sources of the antioxidant agent. This combination could offer a new perspective in anti-diabetic treatment and hold potency to more efficient way to not only control the hyperglycemia but also to effectively prevent the development of diabetic complications.[8]

However, both the effects of *S.polyanthum* extract have not been investigated thoroughly, especially its antioxidant effect. Therefore, this study aimed to study both its hypoglycemic and antioxidant effect of *S.polyanthum* ethanol leaves extract.

METHODS

A true experimental study with pre- and post-test control group design was conducted to evaluate the effectiveness of *S.polyanthum* leaves extract as a hypoglycemic and antioxidant agent. 40 Wistar rats were used in this study which grouped into five groups namely P0 (positive control), P1 (negative control), P2 (alloxan-induced diabetes + 0.5 mg kg^{-1} *S.polyanthum* leaves extract), P3 (alloxan-induced diabetes + 2.0 mg kg^{-1} *S.polyanthum* leaves extract), P4 (alloxan-induced diabetes + 5.0 mg kg^{-1} *S.polyanthum* leaves extract), and P5 (alloxan-induced diabetes + 125 mg/kg glibenclamide). The alloxan was used to induce hyperglycemic condition in the rats by applying it for one week for each rat except those that included in the negative control group. The blood glucose level and 8-OHdG level were assessed at Laboratory of Organic Chemistry Departement of Chemistry and Analytical Science faculty of Udayana University.

The *S.polyanthum* extract was obtained by macerating 3 kg of *S.polyanthum* leaves in 1000 ml ethanol for 48 hours. The resulting solution then filtered and evaporated using rotary vacuum evaporator. The resulting product then analyzed using gas chromatography-mass spectrometry (GC-MS) to unveil its content. The extract was fed to the rats by using a feeding tube.

Table 1 **Compounds detected in ethanol extract of *S.polyanthum* leaves**

Peak number	Retention(minute)	Area (%)	Molecular formula	Compounds name
1	18.93	37.55	0	Phytol
2	19	4.90	0	2-Hexadecene, 3,7,15-tetramethyl
3	19.19	6.82		(z)-1,3-phytadiene
4	19.38	14.06		Cyclopentane, 1-ethyl
5	20.22	5.70	0	Cis-1,3-Dideuterio-1,3-cyclohexana
6	21.62	1497	0,	phytol, acetate
7	25.32	5.17		1,3-dimethyl-4-aza phenanthrene
8	28.13	10.84	0	1-methyl-2 phenylindole-2-Ethylacridine

Table 2 **Blood Glucose Level Before and After Treatment**

Treatment	Observation blood glucose level (mg/dL)	
	Pretest	Posttest
P_0	109.35 ± 3.03	105.76 ± 2.53
P_1	320.33± 3.06	218.60 ± 3.23
P_2	257.21± 2.19	173.59 ± 2.99
P_3	249.41± 2.77	128.59± 1.88
P_4	248.61± 2.07	110.56 ± 1.68
P_5	340.61± 2.73	151.69 ± 2.03

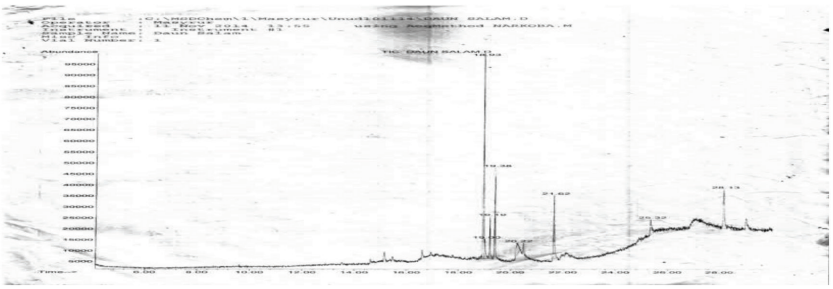

Figure 1 Chromatogram of the active compounds ***Syzygium polyanthum*** leaves extract

RESULTS

The leaves of *S.polyanthum* were successfully extracted and analyzed using GC-MS. The result of GC-MS analysis of showed eight peaks of compounds with different retention (t_R), shown in Figure 1 with a retention time (tR), peak area (%), and molecule weights presented in Table 1. The retention time showed 18.93 which indicates a function to modulate the transcription factor peroxisome proliferator-activated receptor –alpha (PPAR-α) retinoid receptor X (RX). Eight major compounds contained in the extract are shown in Table 1 which majority is the precursor of chlorophyll. The highest peak was found to be the phytol group.

The animal analysis showed that the *S.polyanthum* extract effectively decreased blood glucose with the extent of hypoglycemic effect was in linear proportion to its dosage (Table 2). The most significant drop in mean plasma glucose level was observed at the dosage 5.0 mg kg^{-1} which decrease blood glucose level 65.91% than control. It hypoglycemic effect was found to be higher compared to glibenclamide at 0,18mg/day/200g body weight. One-way ANOVA and posthoc analysis showed that the results were statistically significant.

Furthermore, the antioxidant effect of *S.polyanthum* leaves extracts also proved in in this study (Table 3). The results showed a steady decrease in the 8-OHdG concentration in linear proportion to the extract dosage. The anti-oxidant effect of this extract also proved to be significantly higher than glibenclamide.

DISCUSSION

Diabetes mellitus is a chronic debilitating disease that affects approximately 422 million people in

Published by DiscoverSys | *Bali Med J* 2017; 3(3): S113-S116 | doi: 10.15562/bmj.v3i3.755

Table 3 8-OHdG Level Before and After Treatment

Treatment	Observation of 8-OHdG level (ng/dL)	
	Pretest	Posttest
P_0	6.35 ± 0.33	3.76 ± 0.15
P_1	6.75± 0.46	4.60 ± 0.34
P_2	6.56± 0.19	4.59 ± 0.71
P_3	6.41± 0.07	3.69 ± 0.48
P_4	6.38± 0.47	3.41 ± 0.18
P_5	6.40± 0.45	3.61 ± 0.17

2014.[1] It grave consequences mostly caused by its complication which mainly affect macrovascular and microvascular. Macro-vascular complication includes atherosclerosis, stroke, and acute myocardial infarction while the microvascular complication manifests as neuropathy, retinopathy, nephropathy as well as the diabetic foot which contribute to most diabetic-related morbidity.[2] With its health burden, it is important to find and evaluate new agent for diabetic therapy that addresses not only the hyperglycemia but also the oxidative stress that resulted from it.

The oxidative stress that resulted from the hyperglycemic condition is deeply entrenched in the pathogenesis of diabetic related complication.[9] The primary mechanisms of increased oxidative stress in hyperglycemic condition are due to increased rate of polyol pathway, increased protein kinase C activation, increased intracellular AGE formation, and increased hexosamine pathway activation.[10] The resulting free radicals damage cellular compartments and leading to cellular dysfunction, apoptosis and low-grade inflammation, all of which play the central role in the pathogenesis of microvascular and macrovascular dysfunction.

S.polyanthum leaves are commonly consumed in South-East Asia region including Indonesia.[8] However, besides as food, it holds several therapeutic potentials for several diseases. It also proved to have the cytotoxic effect that could be exploited as an anti-cancer agent. It also had some antimicrobial and antioxidant effect.[11] However, its hypoglycemic activity is only recently revealed.

The blood glucose lowering effect of *S.polyanthum* is still poorly understood. Nevertheless, several mechanisms are thought to be involved. Widyawati et.al showed that methanol extract of *S.polyanthum* could inhibit a α-glucosidase enzyme which yields an effect similar to acarbose.[8] Furthermore, it also enhances the expression and translocation of GLUT-4 which increase glucose uptake by muscle and adipose tissues. The flavonoid compounds of the extract also could help regenerate the dysfunctional pancreatic β-cell which could improve glucose control by optimizing insulin production.[12]

Unlike its hypoglycemic property, the antioxidant effect of *S.polyanthum* is only mentioned in one study by Widyawati et al.[8] It was stated that its antioxidant properties mainly due to its flavonoids content that has strong antioxidant properties. However, our research confirms this notion by showing a lower level of 8-OHdG in *S.polyanthum* extract-fed rats. This antioxidant property could offer great benefit in preventing the complication of DM which is mainly caused by oxidative stress.

However, despite the extent of our findings and facts from previous researchers, the clinical application of *S.polyanthum* extract is still needed to be evaluated carefully. Toxicology research is needed to assess its dosage range before it could administer to human. Furthermore, elucidation and isolation of its active components are also necessary to enable mass production while avoiding adverse reaction as well as interaction with another therapeutic agent.

CONCLUSION

To conclude, the ethanol leaves extract of *S.polyanthum* was showed significant antihyperglycemic and antioxidant properties, especially at 5.0 mg Kg^{-1}bw dosage. Thus, it made it as a potential candidate for the new anti-diabetic agent. However, further studies are needed to evaluate its dosage range and isolate the active compounds.

ACKNOWLEDGEMENT

The authors would like to thank Prof. Dr. dr. A. A. G. Sudewa Djelantik, Sp.PK and Prof. Drh. N. Mantik Astawa, Ph.D. for their invaluable supervision in this study.

REFERENCES

1. World Health Organization. Diabetes: Facts sheet. World Health Organization, Geneva, 2016
2. Fowler MJ. Microvascular and Macrovascular Complications of Diabetes. Clinical Diabetes 2008 Apr; 26(2): 77-82
3. Papatheodorou K, Banach M, Edmonds M, Papanas N, and Papazoglou D. Complications of Diabetes. Journal of Diabetes Research 2015;1:1-5
4. A. Ullah, A. Khan, I. Khan, Diabetes Mellitus and Stress Oxidative: A Concise Review. Saudi Pharmaceutical Journal, 2016; 24: 547-55
5. Pang J, Chan DC, Watts GF. Origin and therapy for hypertriglyceridemia in type 2 diabetes. World Journal of Diabetes. 2014;5(2):165-175
6. Hamilton SJ, Watts GF. Atherogenic dyslipidemia and combination pharmacotherapy in diabetes: recent clinical trials. Rev Diabet Stud. 2013;10:191–203
7. Alemán-González-Duhart D, Tamay-Cach F, Álvarez-Almazán S, Mendieta-Wejebe JE. Current Advances in the

Biochemical and Physiological Aspects of the Treatment of Type 2 Diabetes Mellitus with Thiazolidinediones. PPAR Research. 2016;2016:7614270.
8. Widyawati T, Adlin Yusoff N, Asmawi MZ, Ahmad M. Antihyperglycemic Effect of Methanol Extract of Syzygium polyanthum (Wight.) Leaf in Streptozotocin-Induced Diabetic Rats . Nutrients. 2015;7(9):7764-7780
9. Giacco F, Brownlee M. Oxidative stress and diabetic complications. Circulation research. 2010;107(9):1058-1070.
10. Maiese K. New Insights for Oxidative Stress and Diabetes Mellitus. Oxidative Medicine and Cellular Longevity. 2015;2015:875961.
11. Kusuma IW, Kuspradini H, Arung ET, Aryani F, Min YH, Kim JS, Kim Y. Biological Activity and Phytochemical Analysis of Three Indonesian Medicinal Plants, Murraya koenigii, Syzygium polyanthum and Zingiber purpurea. Journal of Acupuncture and Meridian Studies 2011;4(1):75-79
12. Jananie R.K., Priya V., Vijayalakshmi K. Determination of bioactive components of Cynodon dactylon by GC-MS analysis. N. Y. Sci. J. 2011;4:16–20.