THE EFFECT OF SAUCE OF L. LEUCOCEPHALA SEEDS (LEUCAENA LEUCOCEPHALA [LAM.] DE WIT) ON LOWERING BLOOD GLUCOSE IN HYPERGLYCEMIC MICE

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ABSTRACT

Objective: The research to investigate that the source of Leucaena leucocephala can lower blood glucose levels in hyperglycemic mice.

Methods: In this study, 42 mice were divided into 7 groups each consisted of 6 mice: Normal Group I, Group II (metformin positive control), Group III (negative control), Group IV (sauce of L. leucocephala at a dose of 0.1 ml), Group V (sauce of L. leucocephala at a dose of 0.2 ml), Group VI (sauce of L. leucocephala with doses of 0.4 ml), and Group VII (sauce of L. leucocephala at a dose of 0.8 ml). The dosage of Group II, IV, VI, V, and VII was orally administered. Blood glucose levels in mice were observed during normal conditions and after administration of 200 mg/kg bw alloxan intra-peritoneally (alloxan-induced mice). On day 0, 3, 7, and 14, blood was taken from the tail and glucose levels were measured with a glucometer.

Results: Data were analyzed by one-way analysis of variance followed by LSD test. Based on the results, the sauce at a dose of 0.8 ml was able to lower the blood glucose levels up to normal blood glucose levels.

Conclusion: There was not significantly different between the groups given sauce of L. leucocephala at a dose of 0.8 ml with the group given metformin (positive control).

Keywords: Hyperglycemic, Leucaena leucocephala [Lam.] de Wit, Alloxan, Sauce.

INTRODUCTION

Most Indonesian people's dietary habit does not pay attention to the nutrition balance which could bring negative consequences such as the rise of degenerative diseases, including diabetes mellitus; this disease is a disorder of carbohydrate metabolism which is characterized by high blood glucose levels. One of the characteristics of early diabetes is quickly feeling hungry then made them eat a lot [1]. Diabetes mellitus disease usually lasts for a lifetime. A recent study from the International Diabetes Federation in 2012 revealed that people with diabetes mellitus worldwide reached 371 million people. Whereas, Indonesia entered the order of seven countries with most diabetics. The first rank is China with 92.3 million people, India 63 million, 24.1 million, Brazil 13.4 million, Russia 12.7 million, Mexico 10.6 million, and Indonesia with 7.6 million people diabetic patients [2].

During this treatment, diabetes mellitus is usually performed by given oral antidiabetics or with insulin injection. However, both treatments are less accessible to Indonesians as well as because of the economic factors are also due to the adverse side effects of treatment with insulin or orally, then many medicinal plants are developed to treat this disease as a cheaper alternatives for alternative healing [3]. Medicinal plants that have potential to be developed are associated with a decreasing blood’s glucose level that is Leucaena leucocephala. Part of this plant that serves to lowering blood’s glucose levels are the seeds [4,5].

Research on the effect of antidiabetic L. leucocephala has been carried out previously, in which extracts from L. leucocephala has active compounds such as flavonoids that can lower blood glucose [6]. Based on previous studies, methanol extract from L. leucocephala with doses of 0.25 g/kg BW given in oral mice, which were propagated with aloe vera in the dose of 100 mg/kg BW and 500 mg/kg BW [8].

In addition to having many benefits, L. leucocephala have also been known to have toxin mimosine compounds [9]. Mimosine can cause growth slowdown, hair loss, swollen thyroid gland, and reduce fertility [10]. However, based on the study conducted by DitA, mimosine levels can be minimized optimally through the fermentation process [11]. Sauce is one of the forms of preparation made through the fermentation process. Based on this description, in this study, a hypoglycemic impression test of sauce of L. leucocephala in mice that was first encouraged by alloxan where alloxan was a diabetogenic agent with a cheaper price compared with streptozotocin. The results of this study are expected to be used as information by the public about the effect of antidiabetic sauce of L. leucocephala.

MATERIALS AND METHODS

Materials
The material used in this research is sauce made from Leucaena leucocephala obtained from previous research [12]. The sauce composition can be seen in Table 1. The experimental animals used were male mice (Mus musculus) DDY strain 2-3 months old with average weight of 20–30 g of 42 heads. While the ingredients for hypoglycemic activity test are water, alloxan tetrahydrate, and metformin.

Tool
The tools used in the study were a syringe, oral sonde, cage of mice as well as food and beverage outlets, an analytical balance, pipette, cotton, scissors, glucometers and test strips glucose, flask, and glassware.

Methods
Hypoglycemic activity test
a. All mice maintained for 1 week (7 days), given the same food and drinks.

b. On the 7th day, each mouse that will be used for research is fasted for 16 h, then taken early blood samples by cutting the tail of the mouse. Levels obtained are normal blood glucose levels.
Data analysis
Data were analyzed by normality test and homogeneity test. Data were analyzed by one-way analysis of variance (ANOVA) test if the data were normal and homogenous. If the data are not normal and homogenous, then the data are analyzed by Kruskal–Wallis test. If there is a significant difference in one-way ANOVA test, then it is continued with the smallest significant differential (BNT) test. Moreover, if there is significant difference in Kruskal–Wallis test, then it is continued with Mann–Whitney test.

RESULTS
Before use in the experiment, all mice from each group were weighed, then maintained for 1 week. Then, all the mice were given treatment according to the division of the group for 14 days. On day 0 (hyperglycemia), 3, 7, and 14, weigh the mice to obtain data of weight.

DISCUSSION
From Tables 2 and 3, it can be seen that before treatment hyperglycemic mice of Group II, IV, V, VI, and VII decreased body weight on day 0 and after treatment Group III hyperglycemic mice lost weight on day 7. This is due to the depletion of fat cells and proteins to meet energy needs because they cannot be met from glucose metabolism. In Groups II, III, IV, and V, there is an increase in blood glucose levels which averaged 130–158 mg/dL. However, in Group IV, mice given sauce of 0.1 ml until day 7 continued to increase blood glucose levels. Increased blood glucose levels were also experienced by Group III (negative control), in which the group was not treated.

Based on these data, it can be seen that sauce of L. leucocephala with a dose of 0.8 ml can lower blood glucose levels of mice to normal. This can be seen from the statistical analysis of the difference of blood glucose level of mice before and after treatment, where blood glucose level from Group III was 130.7 mg/dL and still categorized hyperglycemic. In Group VI mice, there was a decrease in glucose levels, where the average blood glucose level was 97.8 mg/dL, and was said to have achieved normal blood glucose levels. In Group VII there is no significant difference with Group II which is positive group of mice given metformin decreased blood glucose levels to reach normal blood glucose levels. Where the mean blood glucose levels of the positive group on day 14 were 102.2 mg/dL. In Group IV, mice given sauce of L. leucocephala with a small dose of 0.1 ml, blood glucose levels continued to increase until day 14, where the average blood glucose level from Group IV was 151.7 mg/dL. Meanwhile, Groups V and VI continued to decrease blood glucose levels but were still categorized in hyperglycemic states because the mean blood glucose levels of Groups V and VI were 129.7 mg/dL and 120.6 mg/dL. For Group III (negative control), blood glucose levels decreased when compared with day 7, where the average blood glucose level from Group III was 130.7 mg/dL and still categorized hyperglycemic. In Group VI mice, there was a decrease in glucose levels, where the average blood glucose level was 97.8 mg/dL, and was said to have achieved normal blood glucose levels.
causing increased blood glucose levels; whereas, flavonoids found in *L. leucocephala* have antioxidant activity that can inhibit free radicals so that *L. leucocephala* has hypoglycemic effect [8].

**CONCLUSION**

Sauce of *L. leucocephala* with dose 0.2 ml, 0.4 ml, and 0.8 ml can decrease blood glucose level of hyperglycemic mice. However, only sauce with a dose of 0.8 ml alone can reduce blood glucose to normal levels.

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**REFERENCES**

1. Jariyah MP. The Effect of Hypoglycemic Pedada Flour (*Sonneratia caseolaris* (L.) ENG) in Alloxan-Induced Wistar Rats. Surabaya: National Development University; 2013. p. 4.

2. Rosalina. The Threat of Diabetes in Indonesia Increases. Tempo Sep 2013.

3. Widowati L, Dzulkarnaen B. Sa’roni Medicinal Plants for Diabetes Mellitus. Mirror World Medicine. Center for Pharmaceutical Research and Development, Research and Development of Health Department of the Republic of Indonesia; 1997. p. 53-7.

4. Adenhigbe SA, Adetunji OA, Odenyi MA. Antimicrobial and pharmaceutical properties of the seed oil of *Lecanuca leucocephala* (Lam.) De Wit (Leguminosae). Afr J Biomed Res 2011;14:64.

5. Talubmook C, Buddhakala N. Hypoglycemic and hypolipidemic properties of leaf extracts from *Phyllanthus acidus* (L.) Skeels., *Lecanuca leucocephala* (Lam.) De wit. and *Psidium guajava* (L.) in streptozotocin-induced diabetic rats. GTSF Int J Biosci 2013;2:30.

6. Sandra F. Effect of Seed Extract of (*Leucaena leucocephala* Lam) De Wit) on Blood Glucose Level and Histopathology Image of Pancreas β Cells of Male White Rat Induced Streptozotocin (Thesis). Jakarta: Pharmacy Faculty of Pancasila University; 2009.

7. Tiara. Influence of Flavonoid Fraction from Extra Methanol Seed (*Leucaena leucocephala* L.) to Blood Glucose Level of Mice Induced by Alloxan (Thesis). Jakarta: Faculty of Pharmacy Universitas Pancasila; 2004.

8. Hardani A. Mimosine Toxicity of *Corcyra cephalonica* Stainton Larvae (Pyralidae, Lepidoptera) (Thesis). Bogor: Bogor Pertanian University; 1984. p. 5.

9. Prasetyo E. Glycemic Index Test and Analysis of Macro Nutrient Content from Soy Sauce Based (*Lecanuca leucocephala* (Lam) de Wit) (Thesis). Jakarta: Faculty of Pancasila University; 2014.

| Time (day) | Group I | Group II | Group III | Group IV | Group V | Group VI | Group VII |
|-----------|---------|----------|-----------|----------|---------|----------|-----------|
| Initial   | 27.8±2.2 | 27.4±1.6 | 26.4±2.1 | 26.4±1.7 | 27.9±1.9 | 27.9±1.3 | 27.1±1.9 |
| 0         | 29.3±2.6 | 24.9±2.1 | 29.8±2.1 | 25.5±1.4 | 26.4±1.9 | 27.3±1.2 | 26.9±1.5 |
| 3         | 29.3±3.0 | 26.5±1.6 | 30.1±1.3 | 26.8±1.9 | 27.5±2.8 | 27.2±1.7 | 26.6±1.4 |
| 7         | 31.8±1.4 | 27.2±1.2 | 26.8±1.5 | 29.8±2.7 | 30.2±4.1 | 28.3±2.8 | 24.2±2.7 |
| 14        | 34.0±2.2 | 28.6±1.3 | 25.2±1.4 | 32±2.7   | 32.2±5.8 | 27.7±3.9 | 23.2±1.6 |

| Time   | Group I | Group II | Group III | Group IV | Group V | Group VI | Group VII |
|--------|---------|----------|-----------|----------|---------|----------|-----------|
| Initial | 81.3±10.2 | 90.5±8.7 | 79±12.9 | 80±12.5 | 85.2±15.2 | 75.8±10.1 | 85.8±12.3 |
| 0      | 91.7±7.2 | 158±2.3 | 130±9.1 | 136.5±19.1 | 156.8±15.3 | 155.2±19.4 | 144±15.2 |
| 3      | 82.7±10.9 | 130.5±17.5 | 137.5±10.6 | 140±9.7 | 137.2±11.9 | 138.8±12.6 | 137.3±12.7 |
| 7      | 77.2±5.1 | 123±13.3 | 142.2±7.7 | 140±8.8 | 134.8±11.3 | 132.2±28.5 | 99.8±9.4 |
| 14     | 87.5±12.7 | 102.2±11.9 | 130.7±14.8 | 151.7±9.3 | 129.7±12.9 | 120.7±26.5 | 97.8±8.1 |

**Table 2: Mean weight data of mice (g)**

**Table 3: Mean data on blood glucose levels of mice (mg/dL)**