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The effect of fermented soybean flour towards blood glucose level of mice (*Mus musculus*)

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Abstract

Diabetes mellitus (DM) is a set of symptoms that arise in a person characterized by blood glucose levels blood glucose levels exceeding normal values (hyperglycemia) and there is a disturbance of insulin metabolism. Patients with diabetes mellitus are unable to secrete insulin in sufficient amounts, use insulin effectively or both. Long-term diabetes mellitus can cause pathological abnormalities macrovascular and microvascular pathological abnormalities. This study used 3 groups treatment groups with each group consisted of 10 mice. Before the research was carried out, first mice were first adapted to the laboratory atmosphere for seven days. Mice that can pass the adaptation period, divided into five groups randomly, fasted overnight for 10-12 hours. Soybean tempeh flour (*Glycine Max L.Merrill*) lowers blood glucose levels of adult male Swiss Webster adult male mice that are induced glucose. Soybean tempeh flour (*Glycine max L. Merrill*) dose of 18,050 mg/kgBB has the potential equivalent to glibenclamide (positive control) in reduce blood glucose levels male Swiss Webster mice induced by glucose.

Keywords: Fermented; Soybean; Blood; Glucose; Mice

1. Introduction

Diabetes mellitus (DM) is a set of symptoms that arise in a person characterized by blood glucose levels blood glucose levels exceeding normal values (hyperglycemia) and there is a disturbance of insulin metabolism. Patients with diabetes mellitus are unable to secrete insulin in sufficient amounts, use insulin effectively or both. Long-term diabetes mellitus can cause pathological abnormalities macrovascular and microvascular pathological abnormalities [1]. Signs and early symptoms that are often complained of DM patients are thirst, a lot of urination, hunger, weakness in the body, and weight loss [12].

Diabetes mellitus is one of one of the biggest threats to global health and the number of people with DM is increasing rapidly worldwide. According to report from the World Health Association (WHO) regarding the study of DM populations in various countries, Indonesia ranked 4th in 2000 with number of people with DM as many as 8.4 million after India, China and the United States United States. WHO estimates an increase in the number of people with DM in Indonesia to 21.3 million by 2030 [5]. The magnitude of incidence, prevalence, and complications of DM illustratethe importance of prevention and early management of the disease. DM management is most effective in the early stages before symptoms develop or prediabetes [7].

Prediabetes is characterized by fasting blood glucose levels between 100-125 mg/dL. Prediabetes is one of the risk factors risk factor for heart and blood vessels. Prediabetes condition can be improved by changing sedentary life, losing weight, managing diet, and doing regular exercise [5]. One of the food ingredients that associated with improving the

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condition of prediabetes through lowering blood glucose levels is soy. The habit of consumption of legumes, especially soybean has a protective effect against DM type 2[3]. Isoflavone content provides hypoglycemic effect. This content contained in processed soy products among others: tempeh, tofu, soygurt, and soy milk [11].

Soybeans are plants that can be processed into tempeh through the process of fermentation process. Soybeans and tempeh have isoflavone compounds. Tempe consumption consumption is expected to continue to increase in line with efforts to increase consumption of protein consumption and in accordance with the purchasing power society[13]. Tempeh is a product of fermented product that has high nutritional value. Isoflavone content in processed soybean processed products such as tempeh is higher compared to unprocessed soybeans [2]. Fresh tempeh can be stored for one to two days at room temperature without much experience a reduction in its quality properties. After two days, tempeh will experience decay process and can no longer be consumed. To overcome this problem, tempeh can be preserved in the form of tempeh flour [14].

Tempeh flour has many benefits, including easy mixing with carbohydrate sources to enrich nutritional value, easy to store, or easily processed into fast food. With the presence of tempeh flour, the nutritional value of a food will increase. Making tempeh flour is done as a solution to increase the nutritional value of nutritional value in low-protein foods. Tempeh flour is useful as a substrate in low protein foods. The purpose of making tempeh flour is to increase the selling value of tempeh and flour diversification, increase nutritional content for low protein foods low protein foods, and improve the nutrition of the Indonesia, especially the middle class to the bottom. Tempeh flour can be applied to every type of food both side dishes and snacks. Examples of snacks that can be made from tempeh flour are satay donuts, brownies, tiramisu, and others [13].

2. Material and methods

This study used 3 groups treatment groups with each group consisted of 10 mice. Before the research was carried out, first mice were first adapted to the laboratory atmosphere for seven days. Mice that can pass the adaptation period, divided into five groups randomly, fasted overnight for 10-12 hours. After fasting, the test animals that had been grouped, blood samples were taken from the from the tail of the mice to measure initial blood glucose level (T0). Then all treatment groups were given the drug tested in three doses, positive control positive control, and negative control. After 30 minutes, 50% glucose was given orally as much as 2mL. Venous blood collection from the tail of mice was repeated after treatment at 15, 30, 60, 90, and 120 minutes. The data measured was the decrease in blood glucose levels at minutes 15, 30, 60, 90 and 120. Data analysis using One-way ANOVA with $\alpha = 0.05$. The level of meaning or significance of the results analysis was tested with Tukey HSD test with p value <0.05.

3. Results and discussion

From the results of the average blood glucose of all groups at each minute obtained that the blood glucose of mice after treatment at the 15th and 30th minutes showed an increase. This is because glucose loading still occurs so that glucose levels in the blood are still high. There was a decrease at minutes 60, 90 and 120. But in the KN group, blood glucose still increased at minute 60 and began to show decreased at minutes 90 and 120. This is because when blood glucose increased beyond the normal limit, the speed of pancreatic insulin secretion also increases so that the blood glucose level back to its control value. Glucose that absorbed by the intestine will be converted into glycogen and stored in the liver [9].

Table 1 Fasting blood glucose level before treatment (mg/dL)

	TT1	**TT2**	**TT3**	**KP**	**KN**
Mice 1	114	144	104	109	100
Mice 2	112	120	112	110	112
Mice 3	109	120	118	118	106
Mice 4	120	123	126	113	111
Mice 5	116	114	120	114	116

Table 2 Mean blood glucose after treatment (mg/dL)

	TT1	TT2	TT3	KP	KN
15 minutes	154 ± 4.73	163 ± 11.03	160 ± 7.59	155 ± 2.54	145 ± 9.50
30 minutes	161 ± 6.55	165 ± 6.36	170 ± 3.27	166 ± 1.81	157 ± 6.26
60 minutes	155 ± 4.23	154 ± 6.77	152 ± 4.07	151 ± 3.29	163 ± 3.20
90 minutes	150 ± 3.23	143 ± 7.69	142 ± 3.70	136 ± 5.43	161 ± 2.26
120 minutes	144 ± 2.06	135 ± 6.60	132 ± 6.42	1.24 ± 3.55	155 ± 2.74

From the above results, it is found that at 15th minute TT2 group against KN group and at the 30th minute TT3 group against the KN group group showed a significant difference significant. But because at minute 15 and 30th minute blood glucose levels still increased, the result of significance that will be discussed further are at 60th, 90th, and 120th minutes because at minutes, the blood glucose level has decreased. At the 60th minute, the TT1 and TT2 groups in the experimental results have showed a decrease but the results of Tukey HSD test results did not show significant difference to the KN group, this means that the TT1 and TT2 groups have not proven the effect of reducing blood glucose statistically. Group TT3 group showed a significant difference significant difference to the KN group, thismeans that TT3 group has the effect of of lowering blood glucose.

At 90 minutes the TT1 group did not showed no difference to KN group, TT2 and TT3 groups group showed a very significant difference to the KN group, this means that TT2 and TT3 groups have the effect of lowering blood glucose. TT2 and TT3 groups compared with the KP group showed no difference, this means that the TT2 and TT3 groups have the same the same blood glucose lowering potential with the KP group. At the 120th minute, the TT1 and TT2 group compared with KN group group showed the effect of lowering blood glucose which is statistically significant, but when compared with the KP group showed a very significant difference (p=0.000) which indicates that the lowering effect of the TT1 and TT2 groups were not better than KP group. TT3 group when compared to the KN group group showed the effect of lowering blood glucose blood glucose lowering effect that is very significant statistically statistically significant with a value of p=0.000, and when compared to the KP group group showed no difference (non significant), thus concluding the potential to reduce blood glucose by TT3 group is the same as the KP group.

Previous research on tempe flour reduces blood glucose profile in Wistar male white rats induced by alloxan. induced by alloxan was seen to decrease blood glucose every week in 3 weeks weeks of research showed that in the treatment group given tempe flour compared to the negative control group showed a very significant difference (p (p=0.000)[10]. Soybeans contain isoflavones which has a hypoglycek effect. Isoflavones contained in soybeans have the effect of increasing insulin sensitivity insulin sensitivity[8], improve insulin secretion [6], block intestinal glucose absorption [3]and is an antioxidant. The process of fermentation of soybeans into tempeh causes an increase in isoflavone [2].

The study found that through processing both fermentation and non-fermentation, isoflavone compounds can be undergo transformation into free isoflavones called aglycones. These aglycones have higher activity. The aglycone compounds are genistein, glycitein, and daidzein which are associated with lowering blood glucose blood glucose. Genistein and daidzein act as antihyperglycemia through glucokinase (GK) activation mechanism, inhibition of glucose-6-phosphatase (G6pase), phospoenol pyruvate carboxykinase (PEPCK), fatty acid synthase (FAS), β-oxidation and carnitine palmitoyltransferase (CPT) in the liver [16]. Isoflavones in soy protect cells from cytokine pre-inflammation, damage fat induction and apoptosis. Isoflavones can also stimulate the endurance of beta cells and lower blood glucose by activating the PPAR (peroxisome-proliferator activated receptor), a nuclear receptor that participates in the regulation of blood glucose and insulin action [16]. Through the process of further fermentation, the aglycone compound will transform to produce new compounds. The result of further transformation of this aglycone compound produce compounds that have higher biological activity. Factor-II (6,7,4' tri-hydroxy isoflavone) has better antioxidant activity thanantioxidant activity than daidzein and genistein in Soybean [15]. This antioxidant activity can overcome free radicals caused by state of hyperglycemia [4].

4. Conclusion

Soybean tempeh flour (*Glycine Max* L.Merrill) lowers blood glucose levels of adult male Swiss Webster adult male mice that are induced glucose. Soybean tempeh flour (*Glycine max* L. Merrill) dose of 18,050 mg/kgBB has the potential equivalent to glibenclamide (positive control) in reduce blood glucose levels male Swiss Webster mice induced by glucose.

Compliance with ethical standards

Disclosure of conflict of interest

The authors declare no conflict of interest.

Statement of informed consent

Informed consent was obtained from all individual participants included in the study.

Author Contributions

All authors planned, designed the work, and supervised all the processes.

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