

Activity Test Of Ethanol Extract Of Green Sugarcane Stem (*Saccharum Officinarum* Linn) As Antidiabetic In Male Mice

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ABSTRACT

Diabetes Mellitus is a metabolic disorder characterized by an increase in blood sugar levels caused by the body's inability to produce insulin. One of the non-pharmacological therapies is by utilizing herbal plants such as green sugarcane stalks (*Saccharum officinarum* L.). Research objective: to find out whether administration of green sugarcane stem extract can reduce alloxan-induced blood glucose levels in mice. Research method: experimental using male mice as test animals totaling 25 animals which were divided into 5 groups, namely the negative control group, positive control, a sample group of 100 mg/kg, 125 mg/kg, and 250 mg/kg. Data were analyzed by one-way ANOVA test and continued with the LSD test. The results of the study at a dose of 125 mg/kg BW optimally reduce blood glucose level

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1. INTRODUCTION

Diabetes mellitus is a metabolic disorder characterized by hyperglycemia due to insulin deficiency or due to insulin resistance. Symptoms include frequent urination, thirst, and hunger (Putra, et al, 2017).

So far, the treatment that has been carried out for diabetics is by using insulin and administering oral anti-diabetic drugs. The use of OAD (Oral Antidiabetes) routinely in the long term can cause unwanted side effects. This encourages the search for alternative drugs that are safe and have relatively smaller side effects. One of them is by utilizing plants that can be useful as anti-diabetics.

One of the plants that is efficacious as an antidiabetic is sugar cane (*Saccharum officinarum* Linn). Sugarcane (*Saccharum officinarum* Linn) contains anthocyanins, flavonoids and their derivatives, and phenolic acids are type of natural antioxidant. (Wibawa, et.al, 2021).

One of the contents of sugarcane (*Saccharum officinarum* Linn) is anthocyanin which is efficacious as an anti-diabetic by inducing the hormone insulin in the pancreas. Anthocyanins are pigments that give red, blue, or purplish colors to flowers, fruits, and vegetables (Abdullah, 2017).

2. METHODS

This research is using experimental method. Phytochemical screening and activity tests of green sugar cane stem ethanol extract were carried out. The simplicial powder was extracted by maceration using 96% ethanol.

3. RESULTS

The results of the phytochemical screening of flavonoid compounds from the ethanol extract of green sugar cane stems were carried out with concentrated HCl reagent and Mg powder producing a yellowish red color. Saponins were tested by observing 1 cm high foam which lasted for 10 minutes. Observation of tannins by adding 1% FeCl₃ positive reaction was indicated by the presence of color greenish black (Pangemanan, Suryanto & Yamlean, 2020), The alkaloid test was tested using dragendorf and mayer reagents. Anthocyanins were tested by mixing with 2 M HCl and heating. From the results of the phytochemical screening, the following results were obtained:

Table 1. Results of the Phytochemical Screening of Green Sugar Cane Stem Ethanol Extract.

Test	Procedure	Result	Note
Alkaloid (mayer)	Extract + HCl 2N + 2-3 drops of mayer reagent	No white precipitate	-
Alkaloid (dragendorf)	Extract + HCl 2N + 2-3 drops of dragendorf reagent	There is a red precipitate	+
Flavonoid	Extract + 10 ml water boiled for 5 minutes, strained and taken 5 ml + 0,1 g powder Mg + 1 ml HCl concentrated + 2 ml C ₅ H ₁₂ O	Orange	+
Saponin	Extract + 1 drop of 2N HCl was shaken vigorously	Foam was formed	+
Tannin	Extract + 100 ml distilled water to boil for 3 minutes, filtered. Take 2 ml + 1-2 drops of FeCl ₃ .	Blackish green	+
Anthocyanin	Method 1: extract + 10 ml distilled water + 2M HCl heated for 2 minutes	Dark red	+
	Method 2: extract + 10 ml distilled water + NaOH drop by drop.	Green	A slight blue discoloration occurs at the top of the test tube solution.

Green sugarcane stem extract was tested by Thin Layer Chromatography (TLC) using a silica gel plate G60 F254 and the mobile phase of n-butanol: acetic acid: water (4:1:5) was observed under UV light at 366 nm. The chromatogram results are shown in the image below:



Figure 1. Thin Layer Chromatography Results with a Wavelength of 366 nm

Treatment of samples and experimental animals

Sugarcane stems simplicia was macerated using 96% ethanol for 5 days and re-macerated for 7 days. Extracts were prepared with concentrations of 100 mg/kg, 125 mg/kg, and 250 mg/kg. Negative control CMC Na 0.5% and positive control Glibenclamide 3 mg/kg BW. Administration of the extract to mice was suspended in CMC Na 0.5%.

Measurement of blood sugar levels was carried out 6 times, namely before alloxan induction, after alloxan induction (initial) and after drug administration at the 2nd, 4th, 6th and 24th hours. Mice

were fasted for 16 hours while still being given a drink and then injected with alloxan solution with dose of 120 mg/kg body weight. The results of the activity test can be seen in table 2 below:

Table 2. Results of the Average Decrease in Blood Sugar Levels.

Groups	Decreased Blood Sugar Levels (mg/dL)					
	T0	T2	T4	T6	T24	P
Kontrol (-)	150,4	140,6	111,8	120,4	154,8	44
Kontrol (+)	154,6	143,8	127,6	110,6	95,6	59
Ekstrak 1	148,2	132,4	127,4	118,4	103,2	45
Ekstrak 2	160,2	149,8	139,2	123,2	105,4	54,8
Ekstrak 3	155,2	143	132,2	115	102,6	52,6

Information :

Extract 1: 100 mg/kgBB

Extract 2: 125 mg/kgBB

Extract 3: 250 mg/kgBB

T0 : Beginning

T2 : 2nd hour

T4 : 4th hour

T6 : 6th hour

Q: Decrease

Based on Table 2, it can be seen that the results of measuring blood sugar levels were carried out in 1 day for 24 hours. Blood sugar was checked at 2, 4, 6, and 24 hours. Blood sugar levels on the third day after alloxan induction were the initial blood sugar levels. The results of research conducted in a day obtained the results of a decrease in blood sugar levels in mice due to the influence of the test extract. The research results are displayed in the form of average and standard deviation.

The negative control group of diabetic mice given 0.5% CMC-Na suspension experienced an increase in blood sugar levels of 44 ± 28.8 mg/dL for 24 hours, and the positive control group of diabetic mice was given glibenclamide suspension at a dose of 3 mg/kg BW mice. experienced a decrease of 59 ± 10.79 mg/dL.

The 100 mg/kg BW dose group of mice with diabetes who were given green sugarcane stem extract suspension was able to lower blood sugar by 45 ± 17.39 mg/dL in a day. The 125 mg/kg BW dose group of mice with diabetes who were given a suspension of green sugarcane stem extract was able to reduce daily blood sugar by 54.8 ± 15.12 mg/dL. Whereas for the 250 mg/kgBB group, mice with diabetes were given green sugarcane stem extract suspension to lower blood sugar in a day by 52.6 ± 9.63 mg/dL.

The results of the phytochemical screening showed the presence of alkaloids, tannins, saponins, flavonoids, and anthocyanins. From the research data that has been done, it appears that the decrease in blood glucose in mice is due to green sugarcane stems having anthocyanin compounds as anti-diabetic activity. Anthocyanin as an antidiabetic can help induce the production of insulin hormone from pancreatic cells, this ability is demonstrated by binding to pancreatic beta cells which then reduces the level of cell saturation (Wibawa, Andila, Lugrayasa & Sujarwo, 2021). Anthocyanin acts as an antidiabetic by protecting pancreatic beta cells from oxidative stress.

The normality test data with Shapiro Wilk showed normality with a significance of more than 0.05 ($p > 0.05$), for the homogeneity test a significant value of 0.053 was greater than 0.05 (homogeneous). The variance similarity test was carried out using the Levene test with the same data variance results. The results of the ANOVA test showed that there was a significant difference in decreasing blood sugar levels between the five study groups. The LSD (least significant difference) test showed that the green sugar cane extract at 125 kg/kg body weight did not have a significant difference with the positive control.

4. CONCLUSION

From the results of the study, it can be concluded that the optimal dose of green sugar cane ethanol extract is at a dose of 125 mg/kg BW with a sig value > 0.05 which is comparable to the positive control of glibenclamide with a decrease of 54.8 mg/dL.

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