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Efficacy of Kyllinga monocephala Rottb. Plant Extract in Lowering the Blood Glucose Level of Mice

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Abstract

Medicinal plants are among the oldest products mankind has utilized. In folk times, herbal drugs have helped many people in treating their illnesses. In our time today, many plants which are proven to be effective through intensive laboratory experiments are being processed and registered to be an approved medicine.

Kyllinga monocephala Rottb. is a common weed found abundantly in open-grasslands. Some may think that it is useless but this type of weed contains active compounds which has antihyperglycemic effect.

The study conducted a simple experiment to prove if the plant really possesses such properties. The study aimed to find out if Kyllinga monocephala Rottb. plant extract reduces the blood glucose level of mice. Specifically, the study identifies the active compounds present in the plant extract; and determines the antihyperglycemic potential of annuang plant extract at different concentrations.

This study would serve as a springboard for future researchers who are capable of doing intensive and careful medical studies regarding the plant.

Keyword : Kyllinga monocephala Rottb., blood glucose level, hyperglycemia

1. Introduction

1.1 Background of the Study

Hyperglycemia is the technical term for high blood glucose level. It is characterized by the derangements in carbohydrate, protein and fat metabolism caused by the complete or relative insufficiency of insulin secretion[1]. Diabetes is a disease which is characterized by hyperglycemia. It is a major degenerative disease today. It also causes complications which include hypertension, atherosclerosis and micro-circulatory disorders[2].

There are already methods of preventing high blood glucose related ailments but average and low class people could hardly afford to take these kinds of medications. The use of indigenous materials found in the society such as Kyllinga monocephala Rottb. might be one of the solution for some

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ailments.

This plant belongs to the family of Cyperaceae is can also be referred to as Cyperus kyllinga[3]. Its common names in English are whitehead spikesedge, white-flowered kyllinga or nut grass[4]. In Philippines, it can be called as anuang, bolobotones, busikad, katutu, mutha or sudsud[5].

[Fig. 1] shows the image of Kyllinga monocephala Rottb. The left side shows the picture of the whole plant. The right side shows a closer look. This plant is more or less glabrous, arising from creeping rootstocks. The stems are usually solitary measuring 10 to 40 centimeters in height. The leaves averagely measures 15 centimeters in length and 3.5 millimeters wide. This plant has spikes which are colored white. It is ovoid and measures 8 to 13 millimeters long. The numerous spikelets of this plant measures from 3 to 3.5 millimeters long, the flowering glume distinctly winged along the keel. The nut is up to 1.5 millimeters long[3].

[Fig. 1] Kyllinga monocephala Rottb

In folk medicine, it is also prescribed for fistula, pustules, tumors, measles and other intestinal affections. This plant has aromatic use for indigestion. It can also be used as a treatment for rheumatic problems, stomach and intestinal joint pain. Decocted leaves of this plant can stimulate hair growth and may be also used to ease fever. The roots of this plant are used as diuretic, demulcent, refrigerant and antipyretic[3][6][7].

The methanolic extract of the leaves of Kyllinga monocephala Rottb. was proven to have analgestic activity in a study conducted by Amor, Quanica and Perez in 2009. It was evaluated using acetic acid-induced writhing test on mice. Results show that the extract significantly reduced the number of

writhes in mice by half[8]. The significant analgesic activity was observed by following a bioassay-guided fractionation scheme.

Based on the above information, the researcher decided to conduct a study on the efficacy of Kyllinga monocephala Rottb. plant extract in lowering the blood glucose level of mice.

1.2 Significance of the Study

This study gives information to the people regarding the usefulness of the plant.

This also serves as a literature for future researchers who are interested in studying about Kyllinga monocephala Rottb. and blood glucose level. This ordinary plant may help people with diabetes if further studies will be conducted by skilled medical researchers.

1.3 Scope and Limitations of the Study

The study focuses only on the effectiveness of Kyllinga monocephala Rottb. plant extract in lowering the blood glucose level of mice. The variables evaluated were the specific active compounds through phytochemical analysis and the blood glucose level reduction using different concentrations of the plant extract through Glucose Visual Reagent Test.

The experiment used fifty-four (54) male white mice divided into six (6) treatments of tree mice per group with three (3) replicates each.

Analysis of Variance (ANOVA) and Duncan Multiple Range Test were used as statistical tools to identify the significant differences among treatment means and their interactions at 0.01 and 0.05 levels of significance, respectively.

2. Materials and Methods

2.1 Collection and Preparation of Research Materials, Tools and Equipment

Five (5) kilograms of Kyllinga monocephala Rottb. plants were collected and fifty-four (54) male white mice (three-month old weighing eighteen to twenty-two (18 to 22) grams were obtained from its source one (1) week before the start of the study.

Other materials that were needed in the study were fifty-four (54) mice water dispensers, blender, weighing scale, basin, beaker and graduated cylinder were also obtained.

Ninety (90) milligrams of Metformine hydrochloride, pricking needles, glucometer, glucostrips, and distilled water, hand gloves and face masks were purchased from the pharmaceutical store prior to conduct of the study.

2.2 Research Procedure/Experimental Design

Fifty-four (54) all male mice were pre-weighed using weighing scale to determine the daily feeding ratio based on the five (5) percent body weight. They were divided into six (6) treatments with three (3) replications having three (3) mice each to replicate. Mice in all groups were fed by normal pellets and had an access to water ad libitum. After one (1) week, initial blood samples were obtained by pricking the tail vein of the mice using Glucose Visual Reagent Test. Small amount of blood samples were put on the glucostrips and blood glucose level were measured using the glucometer. On the second week, the mice were induced in order to have high blood glucose, followed by another blood glucose count using glucometer through Glucose Visual Reagent Test. Varying amounts of K. monocephala Rottb.plant extract were orally administered to the experimental groups. Blood glucose level count (milligrams/deciliter) on supplemental extract were assessed through Glucose Visual Reagent Test four (4) hours after the administration of the extract.

Five (5) kilograms of Kyllinga monocephala Rottb. were washed, cleaned and air-dried for twelve (12) hours at room temperature. After air-drying, the plant was chopped into smaller uniform sizes. The chopped material was subjected to fifteen (15) minutes boiling at a ratio of 2:1 : two (2) parts of water and one (1) part of chopped plant material until one-half (1/2) of the liquid was left, then cooled. The liquid was strained using a fine mesh cloth strainer. The resulting filtrate was rotary evaporated to obtain crude extract. The obtained juice was placed in a sterilized glass bottles ready for dilution, experimentation and phytochemical analysis, respectively.

The experiment used six (6) treatments with three (3) replications having three (3) mice in each replicate. The duration of the experimental activities was one hundred twenty-seven (127) days that began on April 15, 2010 and ended on August 20, 2010.

The six (6) treatments used in the study were as follows:

Treatment A – 100%=30 milliliters of anuang extract

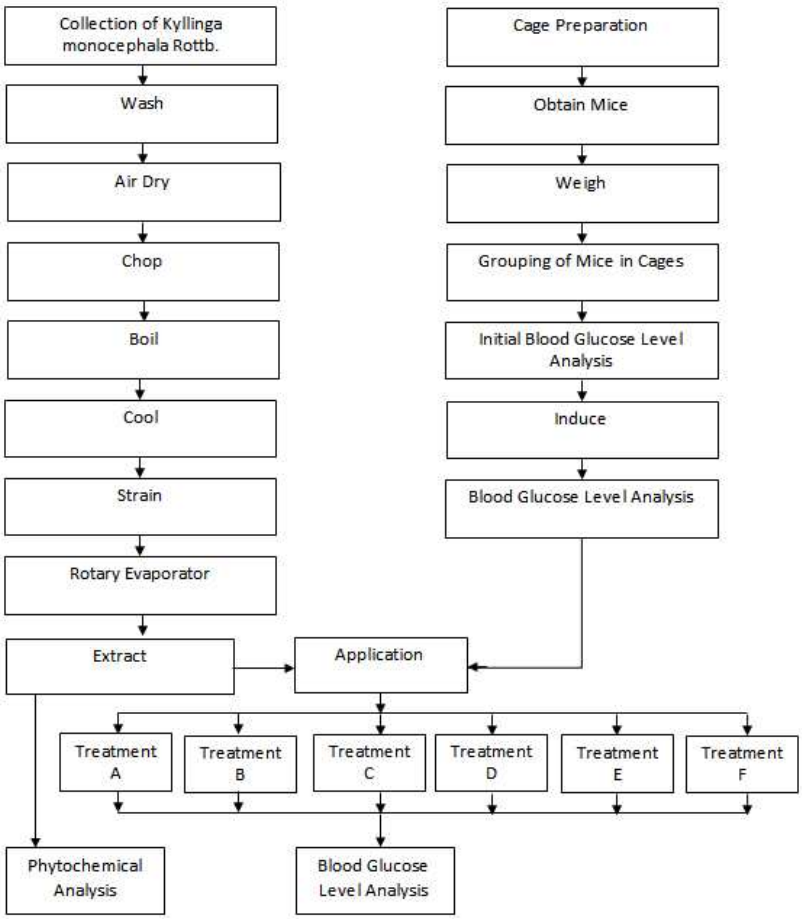
Treatment B – 75%=22.5 milliliters of anuang extract + 7.5 milliliters distilled water

Treatment C – 50%=15 milliliters of anuang extract + 15 milliliters distilled water

Treatment D – 25%=7.5 milliliters of anuang extract + 22.5 milliliters distilled water

Treatment E – Positive Control – 30 milliliters of metformin hydrochloride

Treatment F – Negative Control – 30 milliliters distilled water



[Fig. 2] The Flow Process

[Fig. 2] shows the flow of processes conducted in this experiment

2.3 Gathering of Data

To determine the blood glucose level counts of mice (milligrams/deciliter) at initial, the induced, and anuang Kyllinga monocephala Rottb. plant extract supplemental feeding, the extracted blood samples were subjected to Glucose Visual Reagent Test, and measured by glucometer.

On the other hand, the plant extract was then subjected to rotary evaporator and phytochemical analysis to identification of the active compounds present.

The following tests were done:

Test for Alkaloids. One (1) milliliter of anuang plant extract was transferred to a clean test tube. Three (3) drops of Dragendorff's Reagent was added on the test tube. Brick-red precipitation indicates the presence of alkaloids.

Test for Flavonoids. Three (3) milliliters of anuang plant extract was transferred to a clean test tube. Three (3)v pieces of magnesium turnings and three drops concentrated hydrochloric acid (HCI) was added in the test tube and heated on a boiling water bath for a few minutes. Orange tom pink discoloration indicates the presence of flavonoids.

Test for Tanins. One (1) milliliter of anuang plant extract was transferred to a clean test tube. Three (3) drops of five (5) percent iron chloride (FeCI) solution was added in the test tube. Green to black precipitation indicates the presence of tannins.

Test for Terpenoids. One (1) milliliter of anuang plant extract was transferred to a clean test tube. Three(3) drops of concentrated sulfuric acid was added in the test tube. Dark brown to black precipitation indicates the presence of terpenoids.

Test for Saponins. One (1) milliliter of anuang plant extract was transferred to a clean test tube. Three-fourths (3/4) of the test tube was filled with water and sealed with a cork stopper. It was shaken vigorously to froth and was let stood for a few minutes. Continued frothing indicates the presence of saponins.

3. Results and Discussion

3.1 Findings

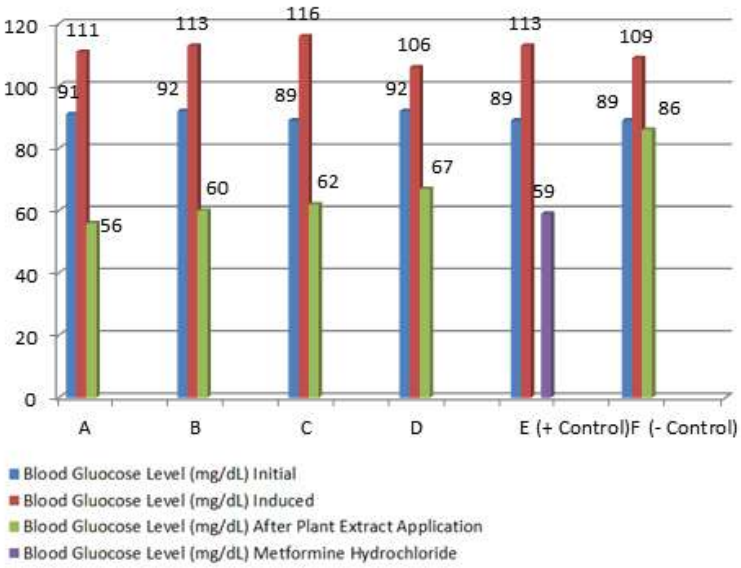
The result of the experimentation on the efficacy of Kyllinga monocephala Rottb. plant extract in lowering the blood glucose level is presented in [Table 1].

[Table 1] Average Data of Blood Glucose Levels

Treatment	Blood Gluocose Level (mg/dL)				Glucose Dropping Difference	Percentage of Dropping Difference (%)
	Initial	Induced	After Plant Extract Application	Metformine Hydrochloride		
A	91	111	56		55	49.54
B	92	113	60		53	46.90
C	89	116	62		54	46.55
D	92	106	67		39	36.79
E (+ Control)	89	113		59	54	47.79
F (- Control)	89	109	86		23	21.11

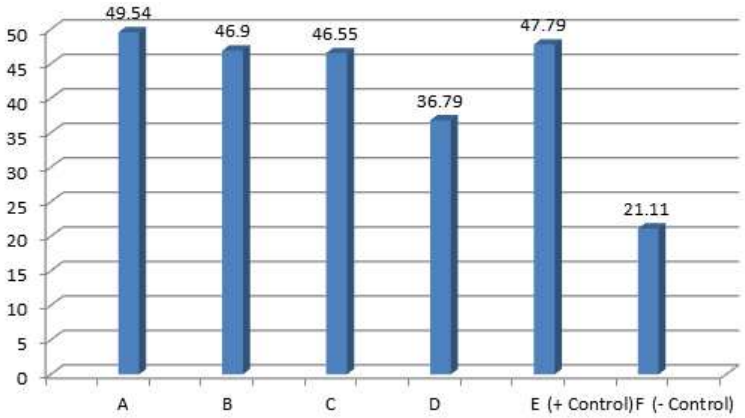
[Table 1] shows the average data of blood glucose levels of mice before fasting and before treatment; after treatment application; change in blood glucose level and percentage blood glucose level change which depicts the efficacy of Kyllinga monocephala Rottb. plant extract in lowering blood glucose level.

Results of the tests in [Table 1] show a drop in the glucose level counts (in milligrams/deciliter) for all mice. The negative control group (Treatment F) with no plant extract lowering less at 21.11 percent. The experimental groups (Treatments A,B,C and D) treated orally with varying formulation of plant extract lowering greatly at 46.54, 46.90, 46.55 and 36.79 percent, respectively. The results further revealed that the higher the treatment dose of plant extract, the larger the reduction effect in blood glucose level. The results also show that plant extract has helped in lowering high blood glucose level on mice comparable to the commercial antihyperglycemic drug metformin hydrochloride with a changing percentage of 47.79 percent (Treatment E).



[Fig. 3] Blood Glucose Levels between Each Treatment

[Fig. 3] shows the graphical trend between treatments. This is a graphical representation of the data from [Table 1] which depicts the initial blood glucose levels, the induced blood glucose level, and the levels after treatment of the plant extract (A,B,C,D,F) and the metformin hydrochloride (E).



[Fig. 4] Percentage Blood Glucose Level Drop

[Fig. 4] shows the graphical representation of the data from [Table 1] regarding the percentage of blood glucose level drop. Results show that there is a bigger blood glucose dropping percentage on

treatments with higher concentration of the extract.

The results of the phytochemical analysis of the active compounds present in extracted plant exhibiting blood glucose level reduction on mice in [Table 2].

[Table 2] Summary of the Active Compounds Present in Kyllinga monocephala Rottb. through Phytochemical Analysis

Active Compounds	Color Reaction	Observed Results
Flavonoids	Orange	+
Alkaloids	Brick Red	+++
Tannins	Dark Green	++
Saponins	N/A(Froth)	+++

Legend: +++= Substantial; ++= Moderate; += Trace

The presence of saponins, and alkaloids in the plant extract as shown in [Table 2] would probably explain the drop of the blood glucose levels on the subjects. Based on the above components present contributes to its efficacy as an antihyperglycemic cure. Thus, in effect, lowers the blood glucose level. Kyllinga monocephala Rottb. contains essential oils such as α-humolene, α-cyperone and β selinene.

3.2 Analysis of Data

The data obtained in this study were analyzed and interpreted by the researcher as presented in [Table 3].

[Table 3] Analysis of Variance (ANOVA) Results of the Test of Varying Doses of K. monocephala Rottb. Plant Extract on the Blood Glucose Level.

Source of Variation	Degree of Freedom	Sum of Squares	Mean Squares	Computed F	Tabulated	
					0.05	0.01
Treatment	5	1802.94	360.59	58.44**	3.11	5.06
Error	12	74	6.17			
Corrected Total	17	1876.94				

**Highly Significant
*Significant
nsNot Significant

Analysis of Variance (ANOVA) results of the tests in [Table 3] showed that K. monocephala Rottb. plant extract at varying concentrations does significantly drop or lower the high blood glucose level of mice. The results further revealed a drop in the blood glucose level counts on all mice with negative

control group dropping less compared to the experimental and positive control groups of mice. This means that the plant extract as anti-hyperglycemic agent is comparable to commercially prepared anti-hyperglycemic drug, metformin hydrochloride.

The results revealed significant differences among treatments K. monocephala Rottb. plant extract and the positive control (metformin hydrochloride) on the blood glucose level counts of mice. The comparison revealed that the plant extract at varying doses has highly significant effect on blood glucose level of mice. The results further show that there is no significant difference in the blood glucose level counts between experimental treatments (Treatments A,B,C, and D) and the positive control treatment (Treatment E). This means that the plant extract can be developed into commercial anti-hyperglycemic drug.

4. Conclusion and Recommendation

4.1 Conclusion

Based on the results of the study, K. monocephala Rottb. plant extract at varying concentrations, even in the very diluted concentration lowered the high levels of blood glucose on mice comparable to commercial anti-hyperglycemic drug, metformine hydrochloride. Hence it can be developed into a commercial anti-hyperglycemic drug. Moreover results of the phytochemical analysis of the plant extract indicate the presence of active compounds alkaloids, saponins, flavonoids, tannins, and essential oils, respectively. The hypoglycemic activity of the plant extract on the high levels of blood glucose on mice may be possibly due to the presence of active compounds saponins, and alkaloids. These active compounds enhanced in lowering the high levels of blood glucose in mice. Henceforth, the plant extract can be an economical alternative to complement the use of synthetic drugs as anti-hyperglycemic herbal medicine, because of its effectiveness in lowering the blood glucose of mice. The higher the treatment dose, the better the reduction effect.

4.2 Recommendations

In review of results and conclusion of the study, the researcher recommends that further researches should be conducted on a specific mechanism and comparative study with other indigenous plant materials to confirm the potential hypoglycemic effect of K. monocephala Rottb. plant extract as low cost and economical raw materials for antihyperglycemic drug. It is recommended further, that the K. monocephala Rottb. plant extract should be tested on higher vertebrates. The researcher also recommends that there must be further studies regarding the hypoglycemic effect of the plant parts such as stem, leaves and roots.

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