

Hypoglycemic Effect of *Andrographis paniculata* Nees and *Lagerstroemia speciosa* extract combination

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ABSTRACT

The aim of this present study is to determine the hypoglycemic effect of combined extract consists of alcoholic extract from herbs of *Andrographis paniculata* Nees and leaves of *Lagerstroemia speciosa* in alloxan-induced diabetic mice. Hypoglycemic effects were tested by treating mice alloxan-induced Balb/C in five groups of five animals for each group using extract from herbs of *Andrographis paniculata* Nees, extract from leaves of *Lagerstroemia speciosa* and combined extract from herbs of *Andrographis paniculata* Nees and leaves of *Lagerstroemia speciosa* at doses of 28 mg/20 g bodyweight. Obtained results were statistically compared with the results from group treated with CMC-Na 0.5% (control) using one way ANOVA followed by Tukey test to determine the significant changes in blood glucose level. Orally administration of 28 mg/20 g body wt. of *Andrographis paniculata* Nees extract exhibited blood glucose level reduction significantly on 7th days of treatment, on the other hand mice treated with *Lagerstroemia speciosa* extract and the combination from extract of *Andrographis paniculata* Nees and *Lagerstroemia speciosa* with all ratio for 7 days did not significantly reduce blood glucose level in alloxan-induced diabetic mice.

Key words: Hypoglycemic effect, *Andrographis paniculata* Nees, *Lagerstroemia speciosa*,. extract combination.

INTRODUCTIONS

Diabetes mellitus is a group of metabolic disorders of fat, carbohydrate, and protein metabolism that results from defects in insulin secretion, insulin action (sensitivity), or both. The two major classifications of DM are type 1 (insulin deficient) and type 2 (combined insulin resistance and relative deficiency in insulin secretion). They differ in clinical presentation, onset, etiology, and progression of disease. Both are associated with microvascular and macrovascular disease complications (Dipiro *et al.*, 2008).

The number of people with diabetes is increasing due to population growth, aging, urbanization, and Increasing prevalence of obesity and physical inactivity (Wild *et al.*, 2004).

Regardless of the type of diabetes, patients are required to control their blood glucose with medications and/or by adhering to an exercise program and a dietary plan. These treatments have their own drawbacks, ranging from the developing of resistance and adverse effects to lack of responsiveness in large segment of patients population.

The limitations of currently-available oral antidiabetic agents either in terms of efficacy/safety coupled with the emergence of the disease into a global epidemic have encouraged a concerted effort to discover drugs that can manage type 2 diabetes more efficiently. Also, with increasing incidence of diabetes mellitus in rural population throughout the world and due to adverse effects of synthetic medicine, there is a clear need for development of indigenous, inexpensive

botanical sources for anti-diabetic crude or purified drugs. (Jarald *et al.*, 2008)

Andrographis paniculata is a plant that has been effectively used in traditional Asian medicines for centuries. the aerial part of the plant, used medicinally, contains a large number of chemical constituents, mainly lactones, diterpenoids, diterpene glycosides, flavonoids, and flavonoid glycosides (Akbar, 2011).

The hot water (0.8 g/kg b.w.) and ethanol extracts (2 g/kg b.w.) of *A. paniculata* reduced the elevated glucose level by 41.51 and 41.82%, respectively in glucose-loaded rats as compared to the respective diabetic control rats. On the other hand, administration of hot water and ethanol extracts of *A. paniculata* decreased the blood sugar level by 46.21 and 45.13%, respectively in alloxan-induced diabetic rats, when compared with that of diabetic control rats. (Hossain *et al.*, 2007)

Lagerstroemia speciosa (L.) Pers. known as "banaba" is traditionally used as a herbal medicine in the Philippines. *Lagerstroemia speciosa* has been shown to produce hypoglycemic effects in some mice models of diabetes. It have been reported that ethanol extract of *L. speciosa* leaves posses the hypoglycemic activity (Mishra *et al.*, 1990).

Thus, The aim of this research is to determine the hypoglycemic effect of combined extract consists of alcoholic extract from herbs of *Andrographis paniculata* Nees and leaves of *Lagerstroemia speciosa*, even though it has been proved by previous research about the hypoglycemic effect of each extract.

MATERIAL AND METHODS

Animals. Adult male mice with body weights ranging from 20–40 g were obtained from Animal Laboratory, Department of Pharmacognosy and Phytochemistry, Airlangga University.

Chemicals. Alloxan monohydrate (Sigma®), glibenclamide (Kimia Farma®), CMC-sodium, ethanol 96%, Glucometer (EasyTouch® GCU Meter) and Blood Gluco-strip (EasyTouch® GCU Meter).

Plant material. The herbs of *Andrographis paniculata* Nees and leaves of *Lagerstroemia speciosa* were collected from the local area on east java in month of March and identified by Purwodadi Botanical Garden, Pasuruan, Indonesia.

Preparation of polar polyherbal formulation. The herbs of *Andrographis paniculata* Nees and leaves of *Lagerstroemia speciosa* were extracted individually with ethanol 96% using simple maceration process and evaporated to concentrated the extract. The residual extracts were mixed in the three ratios (1:1); (1:2) and (2:1).

The extract combination was formulated into suspension form by using 0.5% CMC-Sodium.

Experimental induction of diabetes. The mice were injected with alloxan monohydrate (Sigma®) dissolved in sterile normal saline at a dose of 150 mg/kg body wt. intra peritoneally. Before the injection, all mice were fasted for 18 hours. After three days, mice with blood glucose of 200-600 mg/dl were used for the experiment.

Experimental design. The mice were divided into eight groups after the induction of alloxan diabetes.

Group 1. Normal (Non-Diabetes) mice., Group 2. Diabetic mice given with CMC-Sodium 0,5%., Group 3. Diabetic mice given with glibenclamide orally (0,013 mg/20 g body wt)., Group 4. Diabetic mice given with ethanol 96% extract of *Andrographis paniculata* Nees herbs (28 mg/20 g body weight)., Group 5. Diabetic mice given with ethanol 96% extract of *Lagerstroemia speciosa* leaves (28 mg/20 g body weight)., Group 6. Diabetic mice given with *A. paniculata* : *L. speciosa* extract with 1:1 ratio (28 mg/20 g body weight)., Group 7. Diabetic mice given with *A. paniculata* : *L. speciosa* extract with 1:2 ratio (28 mg/20 g body weight)., Group 8. Diabetic mice given with *A. paniculata* : *L. speciosa* extract with 2:1 ratio (28 mg/20 g body weight).

The Glibenclamide tablet in powdered form was Glibenklamid (Kimia Farma®, Indonesia). The blood samples were collected through the tail vein puncturing with a needle.

Statistical analysis. All values were expressed as the mean \pm S.E.M obtained from a number of experiments (n). Data from all the tables of normal, diabetic control, reference drug treated and extract combination treated animals were compared by ANOVA followed by Tukey test.

RESULTS AND DISCUSSION

Hypoglycemic activity of ethanol 96% combined extracts from The herbs of *Andrographis paniculata* Nees and leaves of *Lagerstroemia speciosa* in alloxan induced diabetic mice, evaluated for seven days. For seven days the mice treated with the test substance suspension in accordance with their respective groups. The observational data from the blood glucose levels of mice presented by mean \pm S.E.M. Effect of treatments given in blood glucose level were shown in Figure 1.

Within seven days, glibenclamide at dosage 0.013 mg/20 g body wt reduced blood glucose levels by 76.4% (313.0 ± 40.3 to 73.8 ± 4.6). treatment with extract of *Andrographis paniculata* Nees herbs (28 mg/20 g body weight) can reduce blood glucose levels in mice by 232.20 mg/dl (45.33%). Extract of *Lagerstroemia speciosa* (28 mg/20 g body wt), can reduce blood glucose levels in mice by 123.40 mg / dl (25.62%). The extract combinations with 1:1 ratio can reduce blood glucose levels in mice by 104.40 mg / dl (20.68%). The extract combinations with 1:2 ratio can reduce blood glucose levels in mice by 154.60 mg / dl (29.39%). Meanwhile, The extract combinations with 2:1 ratio can reduce blood glucose levels in mice by 49.80 mg / dl (10.15%).

The only one that produced significant ($P < 0.05$) reductions in the blood glucose levels of diabetic mice after 7 days of treatment when compared with the diabetic control mice were extract of *Andrographis paniculata* Nees herbs (28 mg/20 g body weight).

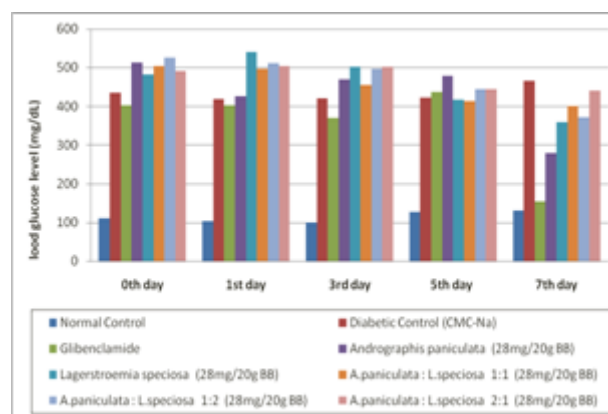


Fig 1. Effect of substances treated to alloxan induced diabetic mices's blood glucose level after 7 days

Table 1. Effect of oral administration from herbs of *Andrographis paniculata* Nees and leaves of *Lagerstroemia speciosa* extract formulation on the blood glucose level in alloxan-induced diabetic mice

Group	Blood Glucose Level (mg/dl)			
	0 th – 1 st Day	0 th – 3 rd Day	0 th – 5 th Day	0 th – 7 th Day
Normal (non-diabetes)	7.20 ± 3.53	10.67 ± 3.47	-16.20 ± -0.97	-20.80 ± 3.50
Diabetic control (CMC-Na 0,5%)	16.20 ± 21.17	15.60 ± 20.67	13.80 ± 18.65	-30.40 ± 16.35
Glibenclamide 0.013 mg/20 g BB	0.20 ± 40.27	32.40 ± 36.42	-34.80 ± 35.77	248.0 ± 34.10*
Extract of <i>Andrographis paniculata</i> Nees 28 mg/20 g BB	86.00 ± 9.69	43.40 ± 9.64	33.20 ± 20.45	232.20 ± 43.62*
Extract of <i>Lagerstroemia speciosa</i> 28 mg/20 g BB	-58.80 ± 34.10	-20.20 ± 35.01	64.20 ± 55.07	123.40 ± 80.57
<i>A.paniculata</i> : <i>L.speciosa</i> extract with 1:1 ratio 28 mg/20 g BB	7.80 ± 41.49	49.80 ± 27.50	90.60 ± 39.34	104.40 ± 33.21
<i>A.paniculata</i> : <i>L.speciosa</i> extract with 1:2 ratio 28 mg/20 g BB	15.40 ± 42.13	29.60 ± 12.55	82.60 ± 21.39	154.60 ± 71.51
<i>A.paniculata</i> : <i>L.speciosa</i> extract with 2:1 ratio 28 mg/20 g BB	-14.00 ± 21.41	-11.20 ± 6.19	45.80 ± 19.92	49.80 ± 24.60

The values are expressed as mean + SEM. n = 6 animals in each group. Statistical significant test for comparison was done by ANOVA, followed by Tukey's test. The blood glucose values of each groups are compared with negative control animals, values *P<0.05

Table 2. Glucose Blood Reduction.

Group (28 mg/20 g body wt)	7 th Day	
	Blood Glucose Reduction	% Reduction
<i>Andrographis paniculata</i> Nees	232.20 mg/dl	45.33%
<i>Lagerstroemia speciosa</i>	123.40 mg/dl	25.62 %
<i>A.paniculata</i> : <i>L.speciosa</i> extract with 1:1 ratio	104.40 mg/dl	20.68 %
<i>A.paniculata</i> : <i>L.speciosa</i> extract with 1:2 ratio	154.60 mg/dl	29.39 %
<i>A.paniculata</i> : <i>L.speciosa</i> extract with 2:1 ratio	49.80 mg/dl	10.15 %

From the data mentioned above (Table 2), it can be seen that treatment which gives a largest reduction in blood glucose levels is *Andrographis paniculata* Nees extract with 28 mg/20 g body weight dose by 232.20 mg/dl or 45.33 % within 7 days.

Androdrographolide the primary active component of *A. paniculata*, lowers plasma glucose in STZ-diabetic rats by increasing glucose utilization. The db/db diabetic mice progressively develop insulinopenia with age, a feature commonly observed in late stages of human type 2 diabetes when blood glucose levels are not sufficiently controlled. When an Andro analog was administered orally to db/db mice at a dose of 100 mg/kg daily for 6 days, the blood glucose level decreased by 64%, and plasma triglyceride level by 54%. These data showed that *A. paniculata* and Andro had significant activity for diabetes. (Zhang *et al.*, 2010).

Gallotanin compounds, especially β-PGG (Penta-O--D-Glukopiranoose Galloil), has a major role in the hypoglycemic activity of the leaf extract of *Lagerstroemia speciosa*. Compounds contained in the leaves of *Lagerstroemia speciosa* effectively lowers blood glucose levels by increasing the rate of glucose transport, induces glucose transport (insulin-like) and reduce the release of glycerol (Liu *et al.*, 2001; Klein *et al.*, 2007). β-PGG (Penta-O--D-Glukopiranoose Galloil) has a halftime about 2 hours (Chai *et al.*, 2010).

The data showed that the combination from both extracts did not gave significant effect on blood sugar level, and could be happen related to mechanism of action of active substance and other factor that needed investigated with further study

Conclusion

The extracts combination with all ratios did not gave significant effect on blood glucose levels reduction in alloxan-induced diabetic mice compared to diabetic control.

However *Andrographis paniculata* extract gives the biggest reduction in blood glucose levels by 232.20 mg/dl or 45.33 % within 7 days.

Further study needed to find out the factor that causes the reduction in hypoglycemic activity when both extract were combined.

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