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EVALUATION OF ANTIDIABETIC ACTIVITY OF *SAMANEA SAMAN* (JACQ.) MERR

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ABSTRACT

In the present study methanolic extract of *Samaneasaman*(Jacq.) Merr. was evaluated for its potential anti diabetic activity by *in-vitro* α - amylase inhibition and *in-vivo* epinephrine induced diabetic rats. Several drugs such as biguanides and sulfonylureas are presently available to reduce hyperglycemia in diabetes mellitus. These drugs have side effects and thus searching for a new class of phytochemical. Phytochemical screening of methanolic extract of *Samaneasaman*(Jacq.) Merr. Revealed the presence of flavonoids, carbohydrates, glycosides, saponins and gums and mucilage. Methanolic extract of *Samaneasaman*(Jacq.) Merr. at the doses of 250 mg/kg p.o and 500 mg/kg p.o significantly reduces the increased blood glucose level as compared to the disease control group ($p < 0.001$) at 1 and 2, ($p < 0.05$) 1/2 hours respectively in epinephrine induced diabetic rats. Also shows significant α -amylase inhibition in concentrations such as 50 μ g/ml < 100 μ g/ml < 150 μ g/ml < 200 μ g/ml < 250 μ g/ml. Evaluation of active compounds from the methanolic extract of *Samaneasaman*(Jacq.) Merr. for their antidiabetic activities may pave the way for the identification of a new class of phytochemical for the treatment of diabetes mellitus.

KEYWORDS

Samaneasaman(SS), α - amylase, Epinephrine, Methanolic extract and Phytochemical.

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INTRODUCTION

Diabetes mellitus (DM) is a group of metabolic disorders characterized by hyperglycemia and abnormalities in carbohydrate, fat and protein metabolism. It results from defects in insulin secretion and insulin sensitivity¹. Diabetes mellitus is found to be one of the five leading cause of death in the world. The number of diabetic people is expected to rise to 366 million in 2030. Management of diabetes without any side effects is still a challenge to the medical community². About 130

pure chemical substances extracted from some 100 species of higher plants are used in medicines throughout the world. According to United Nations Development Project (UNDP) report, the annual value of medicinal plants derived from developing countries is about \$ 32 billion (Rs. 100,000 crore). Theoretically, there is the possibility of discovering 328 modern drugs lying hidden in nearly 325,000 species found in tropical rain forests. There are 47 major modern plant based drugs on the world market and the predicted 328 more potential drugs have an estimated value of \$ 147 billion³. Globally various extracts of plant parts are screened for its antidiabetic activity. World Health Organization (WHO) has recommended the traditional plant treatment for diabetes warrant further evaluation.

Samaneasaman(Jacq) Merr. belonging to the family *Mimosaceae* is a large tropical tree growing as much as 60 m tall, with rough wrinkled bark and developing a symmetrical broad umbrella shaped crown about 80 m wide⁴.

Samaneasaman (Jacq) Merr. is a herbal drug, which has long been used by tribes and native medical practitioners in the treatment for many kind of diseases such as rheumatism, constipation, leprosy, diabetes, microbial infection, inflammation and spasms. The root decoction is used in hot baths for stomach cancer in Venezuela. Rain Tree is a traditional remedy for colds, diarrhea, headache, intestinal ailments and stomachache⁵. The leaf infusion is used as a laxative. In West Indies, seeds are chewed for sore throat. The alcoholic extract of the leaves inhibits *Mycobacterium tuberculosis*. In Colombia, the fruit decoction is used as a sedative⁶. *Samaneasaman*(Jacq) Merr. was traditionally used for the treatment of diabetes mellitus in India⁷, however there is no scientific proof regarding its use as anti diabetic agent.

MATERIALS AND METHOD

PLANT MATERIAL

The leaves were collected from Moosaari, a place near Karungal, Kanyakumari District, Tamil Nadu, India in September 2012 and was identified and authenticated by Taxonomist Dr. V. Ganesan.

Professor and Head, Dept. of Botany, Ayyanadar Janakiammal College of Arts and Science, Virudhunagar Dist, Sivakasi, Tamil Nadu. The plant specimen was certified as *Samaneasaman*(Jacq.) Merr. of family *Mimosaceae*. The leaves are shade dried and size reduced for further extraction.

Preparation of Extract

About 85 gm leaf of *Samaneasaman*(Jacq.) Merr. air dried powdered material was taken in 1000ml Soxhlet apparatus and continues extraction was conducted with hexane, chloroform, ethyl acetate and methanol⁸.

Phytochemical Screening

All the four extracts were screened for preliminary phytochemical analysis⁹⁻¹¹.

Animals

Animal experimentation part was performed strictly adhering to Indian regulations and approved by Institutional Animal Ethical Committee (Reference No: SBCEP/ 2012-2013/ CPCSEA/ IAEC-III/ 01). Adult wister albino rats weighing 150-200 gms were used for the study. Animals were maintained under 12 hr light-dark cycles with *ad libitum* access to standard rat pellet diet and water.

EXPERIMENTAL DESIGN

Epinephrine Induced Diabetes

Adult Wister albino rats are fasted overnight and are randomly divided into five groups of 5 each after determining their fasting blood glucose (FBG). All animals in control group will receive epinephrine hydrochloride (0.8mg/kg) intraperitoneally and blood glucose will be estimated at 0, 1/2, 1, 2 h later by using glucometer. The standard and extract groups first will receive their respective drug and plant extracts (250 and 500 mg/kg) p.o. Two hours later, epinephrine hydrochloride (0.8mg/kg) will be given intraperitoneally. The blood glucose will be determined in the same manner as that of control¹².

GROUP-1

Normal control (saline)

GROUP-2

Diabetic control, Epinephrine (0.8 mg/kg) i.p

GROUP-3

Diabetic rat + Glibenclamide (600µg/kg) p.o

GROUP-4

Diabetic rat + SS Methanolic extract (250mg/kg) p.o

GROUP-5

Diabetic rat + SS Methanolic extract (500mg/kg) p.o.

α-amylase inhibition

α-amylase inhibitory properties of the extract was determined by the method of Bernfield with slight modifications as described below. The working enzyme solution was prepared by dissolving 1 mg of α-amylase enzyme in 10 ml of phosphate buffer (pH 6.9). In brief 100 µl of the extract was allowed to react with 200 µl of α-amylase enzyme (Euro Diagnostic Systems Pvt. Ltd) and 100 µl of 200 mM phosphate buffer (pH-6.9). After 20 min of incubation 100 µl of 1% starch was added. The same was performed for the control where 200 µl of enzyme was replaced by the buffer. After incubation for 5 minutes, 500 µl of DNS was added to both the control and test. The tubes were kept in a boiling water bath for 10 minutes. The absorbance was recorded at 540 nm using a spectrophotometer and the percentage of α-amylase inhibition was calculated using the formula

$$\text{Inhibition (\%)} = 100 \left(\frac{\text{Absorbance of Control} - \text{Absorbance of Test}}{\text{Absorbance of Control}} \right)^{13, 14}$$

Statistical Analysis

Values are expressed as mean ± SEM. Data collected from the studies are subjected for statistical analysis using two way ANOVA followed by Tukey’s multiple comparison tests. The difference between each groups are considered statistically significant at P<0.05. All statistical analysis are performed using Graph Pad prism statistical software (version 5.03).

RESULTS AND DISCUSSION

The leaves of *Samaneasaman*(Jacq.) Merr. was subjected to different solvent extractions and the extracts were dried and finally percentage yield also calculated. Methanolic extract showed the presence of maximum phytoconstituents. Thus methanolic extract was used for further study. Phytochemical screening of methanolic extract of *Samaneasaman*(Jacq.) Merr. revealed the presence of flavanoids, carbohydrates, glycosides, saponins and gums and mucilage. The activity of methanolic extract of *Samaneasaman* (Jacq.) Merr.at the doses of 250mg/kg p.o and 500mg/kg p.o, on epinephrine induced diabetes were shown in the Table No.1, 2 and Figure No.1, 2.

Table No.1: Effect of methanolic leaf extract of *Samaneasaman* (Jacq.) Merr.in epinephrine induced diabetic rats

G. No	Drug and Treatment	Normal Blood Glucose mg/dl	Blood Glucose Level in (hour) mg/dl			
			0	1/2	1	2
I	Normal control (saline)	121.67±11.98	125.33±10.73	126.67±11.22	128.67±11.05	129.52±9.87
II	Diabetic control	102.67±3.71	126.34±9.42	176.67±10.29	221.67±11.05**	252.24±9.87*
III	Diabetic treated + Glibenclamide (600µg/kg) p.o	126.48±8.02	139.23±13.97	101.67±23.08##	82.71±10.83#	73.67±12.69#
IV	Diabetic treated + SS 250mg/kg p.o	104.05±5.69	123.67±10.99	102.33±28.70##	94.33±28.90#	86.36±10.09#
V	Diabetic treated + SS 500mg/kg p.o	121.67±5.90	137.67±16.19	107.67±32.42##	98.26±33.31#	88.67±19.81#

Values were expressed as mean blood sugar level (mg/dl) ± SEM (n=5). Statistical analysis was performed using two way ANOVA followed by Tukey’s multiple comparison tests using Graph Pad prism 5.03.*p<0.001, **p<0.01 diabetic control Vs normal control and #p<0.001, ##p<0.05 diabetic control Vs treatment groups.

Table No.2: α - Amylase Inhibition

S.No	Sample	Concentration ($\mu\text{g/ml}$)	% inhibition
1	Methanolic extract of SS	50	21.670
		100	30.742
		150	41.713
		200	54.742
		250	97.865

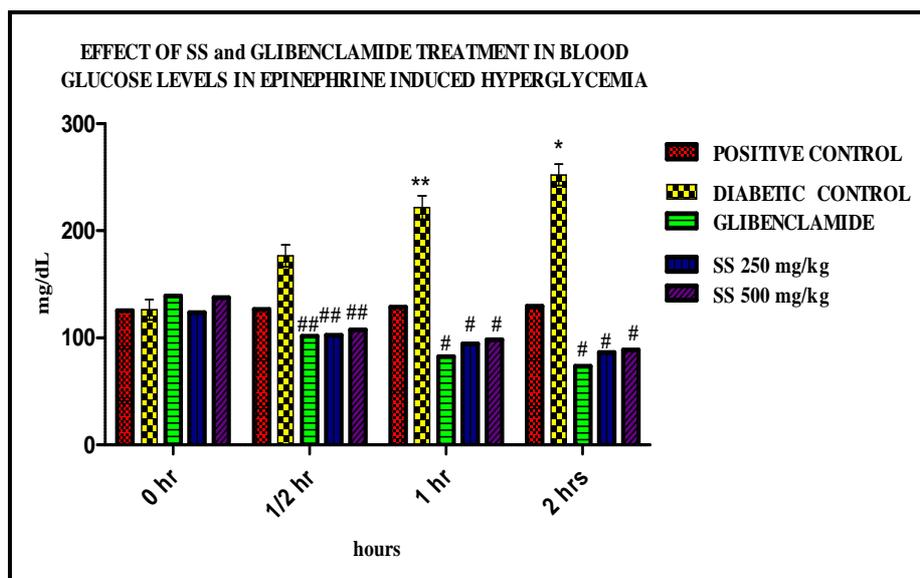


Figure No.1: Effect of SS and Glibenclamide treatment in Blood glucose levels in Epinephrine induced Hyper Glycemia

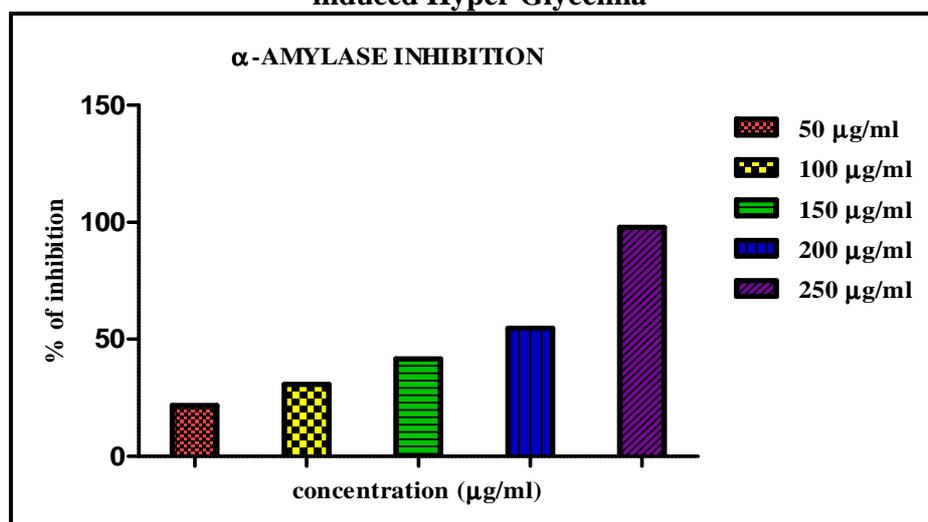


Figure No.2: α - Amylase Inhibition

CONCLUSION

Finding of the present study provide evidence that, methanolic extract of *Samaneasaman* (Jacq.) Merr. at the doses of 250 and 500 mg/kg p.o respectively have potential anti diabetic effect in epinephrine induced diabetic rats. It also shows high *invitro* alpha amylase inhibition in various concentrations. Therefore, the plant *Samaneasaman* (Jacq.) Merr. contains alkaloids, flavanoids and glycosides was considered as powerful anti diabetic agent could offer useful support to the anti diabetic therapy. Isolation and evaluation of active compounds from the methanolic extract of *Samaneasaman* (Jacq.) Merr. for their antidiabetic activities may paw the way for the identification of a new class of phytochemical for the treatment of diabetes mellitus.

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