



PHARMACOLOGICAL SCREENING EVALUATION OF ANTI DIABETIC ACTIVITY ON ETHANOLIC EXTRACT OF ACACIA ARABICA LEAVES

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ABSTRACT

In India, the number of people suffering from diabetes is believed to be rising steadily and the current antidiabetic therapies are frequently reported to have adverse side effects. Ethno medicinal plant use has shown promise for the development of cheaper, cost-effective antidiabetic agents with fewer side effects. The aim of this study was to investigate the antidiabetic activity and mechanism of action of Ethanolic extract prepared from Samanea saman. Since this claim has not been investigated scientifically, the aim of this study was to evaluate the antidiabetic effect and phytochemical screening of Streptozotocin -induced diabetic Rats. The leaves of Samanea saman have been used in traditional health systems to treat diabetes mellitus. However, the antidiabetic activity of this medicinal plant is not scientifically validated and authenticated. The present study aimed to investigate the in vitro and in vivo anti-diabetic activity of flower crude extract and solvent fractions of Samanea saman. The in vitro α -amylase inhibition of the crude extract and solvent fractions of Samanea saman. Blood glucose lowering activity of 80% Ethanolic crude extract and solvent fraction was studied in animal models: Hypoglycemic rats model, oral glucose loaded rat model, dose-treated Streptozotocin -induced diabetic Rat model. The effect of the crude extract on diabetic lipid profile was studied. The acute toxicity study of Samanea saman leaves extract did not show mortality in the animals at the limit dose during the observation period. The result of α -amylase enzyme inhibition activity was found in a dose-dependent manner, the strongest activity was shown by Crude extract fraction (89.60 % inhibition at 1000 μ g/mL) compared to the standard acarbose having 97.19% inhibition at 1000 μ g/mL. The crude extract of Samanea saman showed significant blood glucose-lowering effect on hypoglycemic rats and oral glucose loaded rats. In Streptozotocin -induced diabetic rats model, the crude extract fraction significantly decreased the fasting blood glucose level after 14 days of treatment. The result demonstrated the beneficial biochemical effects of Samanea saman extract by inhibiting α -amylase improving serum lipid profile levels. The leaves crude extract are effective in lowering blood glucose levels in diabetic and hypoglycemic rats. The claimed traditional use as antidiabetic has scientific ground.

Key words: Diabetes mellitus, Herbal medicine, Samanea saman, Streptozotocin, Anti diabetic activity.

INTRODUCTION

The most important genus in the Leguminosae family is Acacia, which Linnaeus originally described in 1773. About two-thirds of the about 1380 species of Acacia that are thought to exist globally are native to Australia, with the other species dispersed across tropical and subtropical parts of the world ^{1, 2}. In his 'Flora of Madras Presidency,' Gamble (1918) listed around 40 species of this genus in India.

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In India, acacia species, often referred to as "babool," have long been utilized in ethnomedicine to address issues with the skin, sex, stomach, and teeth. *Acacia arabica* (Lam.) Willd. (Mimosaceae) and *Acacia nilotica* (L.) Del. syn. Arabic gum trees, sometimes called babul, kikar, or Indian gum trees, are well acknowledged for their many uses. It is abundantly found throughout the world's arid and semi-arid regions. Malaria, toothache (bark), and sore throat (aerial section) have all been successfully treated with *Acacia arabica*. The anti-fertility properties of *A. arabica* nuts and pods have been investigated in tests ³⁻⁸.

A. arabica pod methanolic extracts have been shown to be effective against HIV-PR ^{9,10}. The antiplasmodial activity of *A. nilotica* ethyl acetate extract has currently been evaluated by one team of researchers against several strains of *Plasmodium falciparum* ¹¹ that are susceptible and resistant to chloroquine. According to reports, this species' fresh plant parts are the most effective against Hepatitis C virus ¹². It is a significant multifunctional tree that has been widely used to cure a number of illnesses, including leucoderma ¹³, biliousness, bronchitis, diarrhea, dysentery, colds, and bleeding piles.

Only a small number of research have been done to evaluate *Acacia arabica*'s effects on insulin secretion and action, despite the fact that it is thought to have glucose-lowering properties ¹⁴. In order to determine the ethanol extract of *Acacia arabica* (EEAA) bark's mechanism of action in the treatment of Type 2 diabetes, the current experiment was conducted to examine the antidiabetic effects of the bark both in vitro and in vivo.

About 400 million individuals worldwide suffer from diabetes mellitus (DM), a serious public health condition. ¹⁵. This category of common metabolic illnesses is caused by a number of pathogenic processes, all of which lead to elevated blood glucose levels. Globally, the population is growing at a very quick pace. Its pathophysiology, which includes reduced insulin secretion, impaired insulin sensitivity, increased glucose synthesis, and abnormalities in fat and protein metabolism, is caused by both hereditary and environmental causes. Acute symptoms and metabolic problems might result from hyperglycemia. ¹⁶ However, chronic consequences such as retinopathy, neuropathy, nephropathy, and cardiovascular disease that result from sustained hyperglycemia have led to an increase in diabetes morbidity rates. Maintaining blood glucose control and treating comorbid conditions like dyslipidemia and hypertension might help many individuals avoid these long-term problems. ¹⁷ The medical system still finds it difficult to manage diabetes without any negative side effects, which fuels a growing hunt for better antidiabetic medications. The WHO has suggested that this subject merits investigation because few of the plant remedies used in traditional medicine for diabetes have undergone scientific evaluation. ¹⁸

MATERIALS

Acacia Arabica was procured from Tirumala hills, Tirupati, Andhra Pradesh, India, Glibenclamide was procured from Sanofi, Streptozotocin was procured Loba Chemie, Ethanol was procured Honeywell.

METHODOLOGY

a) Plant collection

The aerial part of *Acacia arabica* was collected from Tirumala hills, Tirupati, Andhra Pradesh, India. It was identified and authenticated by Prof. Madhava Chetty, K., Taxonomist, S.V. University, Tirupati, Andhra Pradesh, India. A voucher specimen has been kept in our laboratory for future reference.

The leaves of *Acacia arabica* were collected and authenticated by Department of Botany. After shade-dried (Temp<40°C.), plant material was grounded into a moderately coarse powder. The extract was made by maceration and the ethanolic extract was made by using Soxhlet apparatus. The extract was allowed to dry. Both the extracts were preserved in the refrigerator till further use.

Invitro antidiabetic activity of *Acacia arabica* leaves extracts

Alpha-amylase inhibition assay

The a-amylase inhibition assay was performed using the 3,5-dinitrosalicylic acid (DNSA) method.⁵⁰ The crude and solvent fractions of *Acacia arabica* were dissolved in buffer ((Na₂HPO₄/ NaH₂PO₄ (0.02 M), NaCl (0.006 M) at pH 6.9) to give concentrations ranging from 50 to 1000 mg/mL. A volume of 200 mL of a-amylase solution (Molychem) (2 units/mL) was mixed with 200 mL of the extract and was incubated for 10 minutes at 30 C. Thereafter, 200 mL of the starch solution (1% in water w/v) was added to each tube and incubated for 3 minutes. The reaction was terminated by the addition of 200 mL DNSA reagent (12 g of sodium potassium tartrate tetrahydrate in 8.0 mL of 2 M NaOH and 20 mL of 96 mM 3,5-DNSA solution) and was boiled for 10 minutes in a water bath at 85°C. The mixture was cooled to ambient temperature and was diluted with 5 mL of distilled water, and the absorbance was measured at 540 nm using a UV-visible spectrophotometer (Agilent Technologies). The blank with 100% enzyme activity was prepared by replacing the plant extract with 200 mL of the buffer. A blank reaction was similarly prepared using the plant extract at each concentration in the absence of the enzyme solution. A positive control sample was prepared using acarbose (Bayer) and the reaction was performed similarly to the reaction with plant extract as mentioned above. The inhibition of a-amylase was expressed as percentage of inhibition and was calculated by the following equation: Inhibition (%) $\frac{1}{4} [(Ac - Acb) (As Asb) / (Ac - Acb)] \times 100$, where Ac is the absorbance of control; Acb is the absorbance of control blank; As is the absorbance of sample; and Asb is the absorbance of sample blank. The % a-amylase inhibition was plotted against the extract concentration and the IC₅₀ values were obtained from the graph.

Preliminary phytochemical screening of Ethanolic leaves extract of Acacia arabica

The Ethanolic leaves extract of Acacia arabica was used for testing preliminary phytochemical screening in order to detect major chemical groups.

Test for carbohydrates

Molisch's test: Dissolved small quantity of 300mg alcoholic and dried leaves extract powder of Pimenta dioica separately in 4ml distilled water and filtered. The filtrate was subjected to Molisch's test.

Fehling's test: Dissolve a small portion of extract in water and treat with Fehling's solution.

Phenols test: The extract was spotted on a filter paper. A drop of phosphomolybdic acid reagent was added to the spot and was exposed to ammonia vapours.

Test for flavanoids

Shinoda test: To 2 to 3ml of extract, a piece of magnesium ribbon and 1ml of concentrated HCl was added.

Lead acetate test: To 5ml of extract 1ml of lead acetate solution was added.

Test for tannins

Braemer's test: To a 2 to 3ml of extract, 10% alcoholic ferric chloride solution was added.

Test for steroid/terpenoid

Liebermann-Burchardt test: To 1ml of extract, 1ml of chloroform, 2 to 3ml of acetic anhydride and 1 to 2 drops of concentrated Sulphuric acid are added.

Test for alkaloids

Draggendorf's test: A drop of extract was spotted on a small piece of precoated TLC plate and the plate was sprayed with modified Draggendorf's reagent.

Hager's test: The extract was treated with few ml of Hager's reagent.

Wagner's test: The extract was treated with few ml of Wagner's reagent.

Tests for Glycosides

Legal's test: Dissolved the extract [0.1g] in pyridine [2ml], added sodium nitroprusside solution [2ml] and made alkaline with Sodium hydroxide solution.

Test for Saponins

Foam test: 1ml of extract was dilute with 20ml of distilled water and shaken with a graduated cylinder for 15 minutes.

Test for Anthraquinones

Borntrager's test: About 50 mg of powdered extract was heated with 10% ferric chloride solution and 1ml of concentrated HCl. The extract was cooled, filtered and the filtrate was shaken with diethyl ether. The ether extract was further extracted with strong ammonia.

Test for Amino acids

Ninhydrin test: Dissolved a small quantity of the extract in few ml of water and added 1ml of ninhydrin reagent.

Test for fixed oils and fats

Press small quantity of the petroleum ether extract between two filter paper.

Note: the results for the above experiments can be noted as follows.

- If the response to the test is high it can be noted as +++ which indicates that the particular group is present as the major class.
- If the response is average then note it as ++ indicates the presence in moderate quantity.
- If the response is very small then note it as + indicating the presence of only in traces.
- If no response is then negative.

g) Acute oral toxicity study

In a research study when a drug is administered to a biological system there will be some interactions may happen. In most case these are desired and useful, but many effects are not advantageous. Acute, sub acute and chronic toxicity studies are performed by the manufacturers in the investigation of a new drug. Acute toxicity is involved in estimation of LD50 (It is the lethal dose (causing death) to 50% of tested group animals).

LD50 (median lethal oral dose)

LD 50 (median lethal oral dose) is a statistically derived oral dose of a substance that can be expected to cause death in percent of animals when administered by the oral route. The LD50 value is expressed in terms of weight of test substance per unit weight of animal (mg/kg).

In this study acute oral toxicity study was carried out in Rats. The procedure was followed by using OECD 423(Acute toxic class method). The Rats are fasted overnight, prior to dosing. The three dose levels are administered by orally the help of oral feeding needle over the prior of 24 hours. After the drug has been administered, food may be withheld for a further 3-4 hours in Rats. The purpose of sighting study is to allow selection of the appropriate starting dose for main study.

The test substance is administered to a single animal in a sequential manner following from the fixed dose levels of 5, 50, 300 and 2000mg/kg. The interval between dosing of each level is determined by the mortality/onset, duration and severity of toxic signs over the period of 24 hours, special attention given during the first 4 hours.

Four hours after the drug administration, provide the food and water for 14 days and daily observed some parameters such as food intake, water intake, mortality, onset, Duration and severity of toxic signs. The animal weight is recorded on weekly once. On the day fourteen all the animals are sacrificed, to isolate the organs and observe the histopathological changes. Based on the mortality result of sighting is decided and carried out with five animals per dose level (5 or 50 or 300 or 2000mg/kg).Based on the mortality result on 14th day of observation, the doses for in vivo study are selected.

In vivo antidiabetic activity of Acacia arabica leaves extract in Streptozotocin induced diabetic Rats.

Prior to the experiment the rats were housed in a clean polypropylene cages (6 Rats / cages) for a period of 7 days under standard temperature (25 - 30°C), relative humidity (45 – 55%), dark / light cycle (12 /12 hrs). The studies were performed with the approval of Organizational Animal Ethics Committee (OAEC) (DAEC/TNA/965/345/16). The animals were put in overnight fasting were deprived of food for 16 hrs but allowed free access of water.

Hypoglycemic Test Groupings were done as follows:

Group I served as control – Carboxy Methyl Cellulose (CMC) 0.5% (0.3ml\100g Rats)

Group II served as Positive control – Glibenclamide (2mg /kg)

Group III served as ethanolic extract of Acacia arabica – (200mg/kg)

Group IV served as ethanolic extract of Acacia arabica – (400mg/kg).

Blood samples were collected by the tail nipping method and glucose level checked by glucometer. After drug Administration blood samples have been collected different time intervals at 30, 60 and 120.

Oral Glucose Tolerance Test Groupings were done as follows:

Group I served as control – Carboxy Methyl Cellulose (CMC) 0.5% (0.3ml\100g Rats)

Group II served as Positive control – Glibenclamide (2 mg /kg)

Group III served as Ethanolic extract of Acacia arabica – (200mg/kg)

Group IV served as Ethanolic extract of Acacia arabica – (400mg/kg).

All the groups of animals were fasted for 24h and blood samples were collected before drug or solvent treatment. The drug, extract and solvent, have been administered to different groups and 30mins later all the groups of Rats were treated with glucose orally at dose 10gm/kg body weight by using oral feeding needle. Blood samples were collected by the tail nipping method and glucose level checked by glucometer. After drug Administration blood samples have been collected different time intervals at 30, 60 and 120.

Induction of diabetes to animals.

A single dose (100 mg/kg b.w., i.p.) of Streptozotocin dissolved in sodium citrate buffer was used for the induction of diabetes in Rats after overnight fasting. After 1 hr of Streptozotocin administration, the animals were given feed and libitum and 5% dextrose solution was also given in feeding bottle for a day to overcome early hypoglycaemic phase. The animals were stabilized for a week and animals showing blood glucose level more than 200 mg/dl were selected for the study.

Experimental design

Five groups of Rats six in each groups received the following treatment schedule for 14 days.

GROUP I - Normal control (normal saline 10 ml /kg, P.O)

GROUP II - Streptozotocin treated control (100 mg/kg, I.P)

GROUP III - Streptozotocin (100 mg/kg, I.P) + Standard drug Glibenclamide (2 mg/kg, P.O).

GROUP IV - Streptozotocin (100 mg/kg, i.p.) + EEAA (200 mg/kg, P.O)

GROUP V - Streptozotocin (100 mg/kg, i.p.) + EEAA (400 mg/kg, P.O)

Plant leaves extract, standard drug and normal saline were administered with the help of oral feeding needle. Group I serve as normal control which received normal saline for 14 days. Group II to Group V were diabetic control Rats. Group IV and Group V (which previously received Streptozotocin 100mg/kg) were given fixed doses of ethanol leaves extract (200 mg/kg, P.O, 400 mg/kg, P.O) of Acacia arabica and group III received standard drug Glibenclamide (2 mg/kg,P.O) for 14 consecutive days. (EEAA- Ethanolic extract of Acacia arabica Leaves).

Collection of blood samples

Fasting blood samples were drawn from retro orbital puncture of Rats at weekly intervals till the end of the study 1, 7, and 14 days.

Estimation of biochemical parameters Serum blood glucose

On 1, 7, and 14 days fasting blood samples were collected and analyzed the blood glucose.

Blood glucose level

The blood glucose level test measures the amount of glucose in the blood sample obtained from the animals. The test is usually performed to check for elevated blood glucose levels which can be an indication of diabetes or insulin inhibition.

Statistical analysis

Statistical analysis was done by using GRAPHPAD PRISM 5.0.All the values of Biochemical parameters and body weight were expressed as Mean \pm Standard Error Mean (SEM). The values were analyzed for statistical

significance using one- way analysis of variance (ANOVA), comparison was done by using Dunnett's t test. P values < 0.05 were considered as significant, P values < 0.01 were considered as very significant, P values < 0.001 were considered as highly significant and ns were considered as not significant.

RESULTS AND DISCUSSION

Appearance and percentage yield of EEAA (Ethanolic Extract of *Acacia arabica* Leaves)

Table.1 a-Amylase Inhibitory Activities of the Crude Extract and Solvent Fractions.

Percentage inhibition					
Concentration (mg/mL)	Chloroform fraction	Ethyl acetate fraction	Aqueous fraction	Crude extract	Acarbose
50	4.52 ± 0.2	17.25 ± 0.95	26.03 ± 1.03	31.20 ± 0.36	57.65 ± 0.79
100	13.51 ± 0.10	23.65 ± 0.51	32.11 ± 0.11	43.01 ± 1.42	68.10 ± 0.46
200	26.26 ± 0.03	28.99 ± 2.58	40.65 ± 0.56	60.12 ± 0.98	76.93 ± 1.53
400	31.31 ± 0.12	44.98 ± 0.20	57.92 ± 0.19	73.84 ± 0.76	88.51 ± 0.17
600	40.09 ± 0.35	52.15 ± 1.47	70.82 ± 0.56	80.19 ± 0.24	93.06 ± 0.26
800	47.26 ± 0.64	66.09 ± 0.12	77.60 ± 0.74	83.55 ± 0.19	96.27 ± 0.17
1000	55.39 ± 0.33	72.69 ± 0.41	84.21 ± 0.22	88.98 ± 0.74	97.19 ± 0.92
IC50	33.25 ± 0.19	23.56 ± 0.82	15.36 ± 0.69	9.69 ± 0.91	3.34 ± 0.14

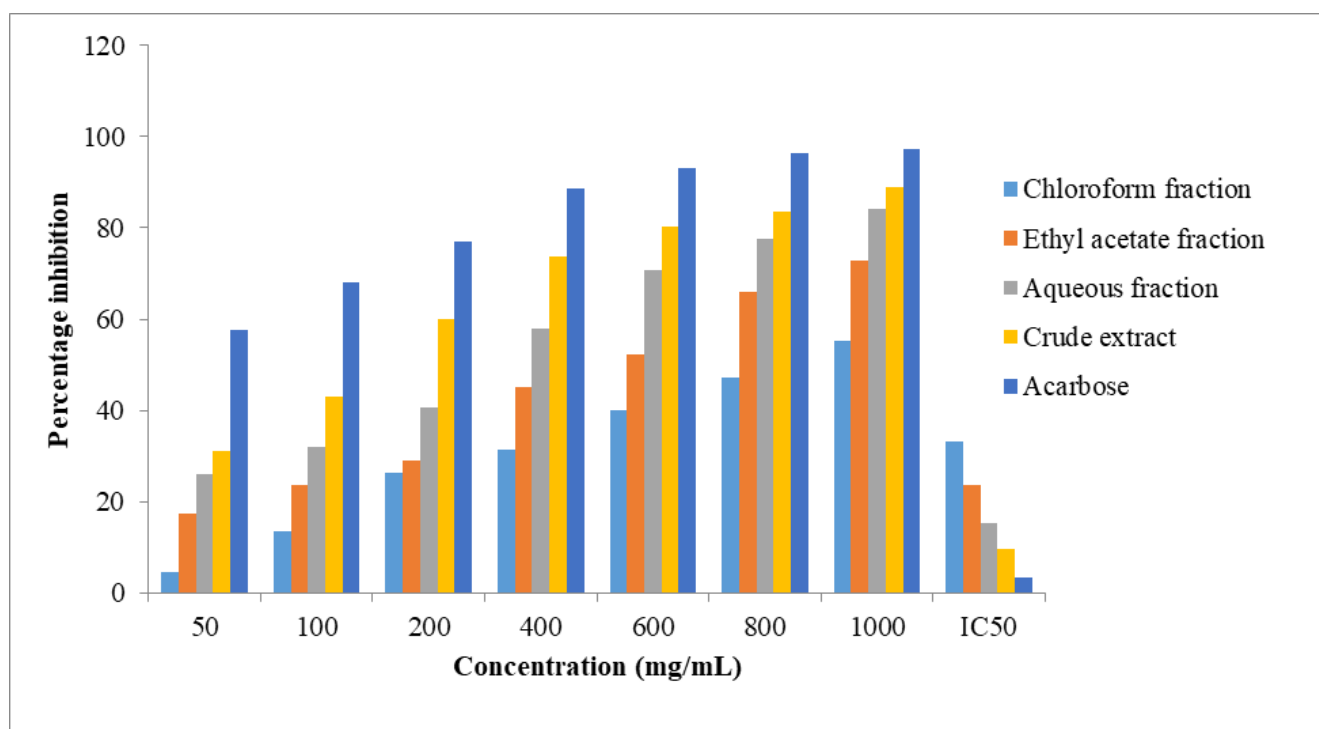


Figure.1 a-Amylase inhibitory activity of the ripe crude extract and solvent fractions of *Acacia Arabica*

Discussion: In Vitro a-Amylase Inhibition Activity of Crude Extract and Solvent Fractions In vitro a-amylase inhibitory study evaluating the percent of a-amylase inhibition as a function of extract concentrations and the IC50 values were calculated (Figure). Concentration dependent inhibitions were observed for various concentrations of the tested extracts and the standard. Among the extracts, the crude extract exhibited the lowest IC50 of 67.21 ± 0.91 mg/mL and the IC50 values of water fraction, ethyl acetate fraction, and the chloroform fraction were 15.36 ± 0.69, 23.56 ± 0.82, and 33.25 ± 0.19mg/mL, respectively. The standard positive control acarbose showed an IC50 of 3.34 ± 0.14mg/mL (Table). Minimum % Inhibition was found *Acacia arabica* leaves which resemblance to %Inhibition of positive control, So Ethanolic extract of *Acacia arabica* contain active constituents of antidiabetic.

Phytochemical studies

Table No.2 Results of Ethanolic extract of *Acacia arabica* leave

Class of compounds	Tests performed	Results
Carbohydrates	Molisch's test	-
	Fehling's test	
Phenols	Phosphomolybdic acid test	+
Flavonoids	Shinoda test	+
	Lead acetate test	+
Tannins	Braemer's test	-
Alkaloids	Wagner's	+
	Mayer's	+
	Draggendorf's test	+
Glycosides	Legal's test	+
	Brontranger's test	+
Saponins	Foam test	+
Sterols	Salkowski's test	-
Amino acids	Ninhydrin test	-
Terpenoids	Lieberman Burchardt test	+

+Present in moderate amount

-Absence

Discussion: The phytochemical studies results revealed that the Molisch's test no characteristic observation indicated the absence of carbohydrates, by phosphomolybdic acid test Blue coloration of the spot indicated the presence of phenols. Shinoda test and Lead acetate test gave pink or red coloration of the solution indicated the presence of flavonoids Flocculent white precipitate also indicated the same. There is no dark blue or greenish grey coloration of the solution indicated the absence of tannins in the drug. No characteristic observation for steroids and dark pink or red coloration of the solution indicated the presence of Terpenoids. Orange coloration of the spot indicated the presence of alkaloids. Yellow or reddish brown precipitation indicated the presence of alkaloids. Pink to red colour solution indicates the presence of glycosides. No layer of foam formation indicates the absence of Saponins. If the response to the test is indicated table-1high it can be noted or which indicates that the particular group is present as the major class. If the response is average then note it as indicates the presence in moderate quantity and note it as indicating the presence of only in traces. If no response is then negative.

Table No.3 Hypoglycemic Test

TREATMENT	DOSE mg/kg	BLOOD GLUCOSE LEVEL (mg/dl)		
		0 min	30min	1hr
CONTROL Carboxyme Thyl Cellulose (CMC)	0.5%	70.25±1.361	69.15±4.320	72.21±1.261
Positive Control Glibenclamide	2	69.36±3.209	51.62±1.492**	31.96±2.415***
Ethanolic Extract of <i>Acacia arabica</i>	200	68.10±1.251	58.20±3.482*	56.14±1.111*
Ethanolic Extract of <i>Acacia arabica</i>	400	67.08±3.420	51.35±3.281**	35.2±2.810***

Discussion: The glucose levels were analyzed by using glucometer and each value is the mean \pm standard error (n= each group consist of 6 animals)(p<0.05)*, (p<0.001)**& (p<0.0001)*** as compared to control & positive control group evaluated by one way, ANOVA followed by Dunnet 't' test.

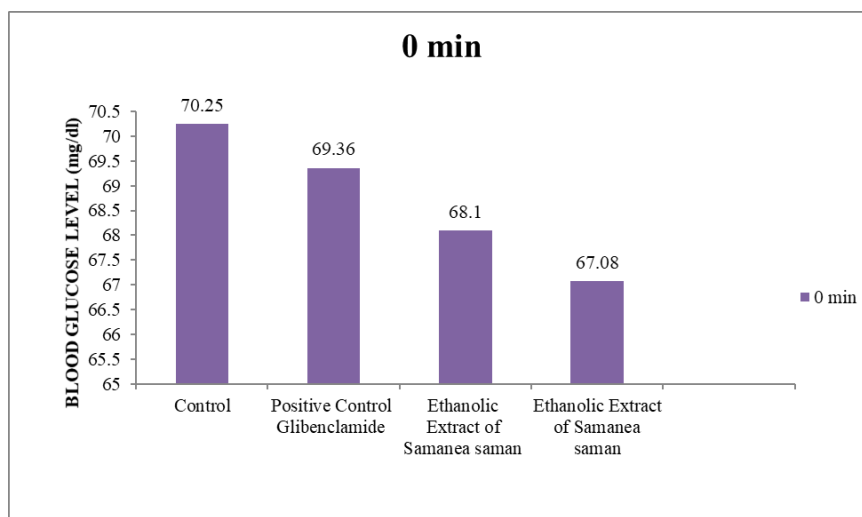


Figure No.2 Blood glucose level 0min

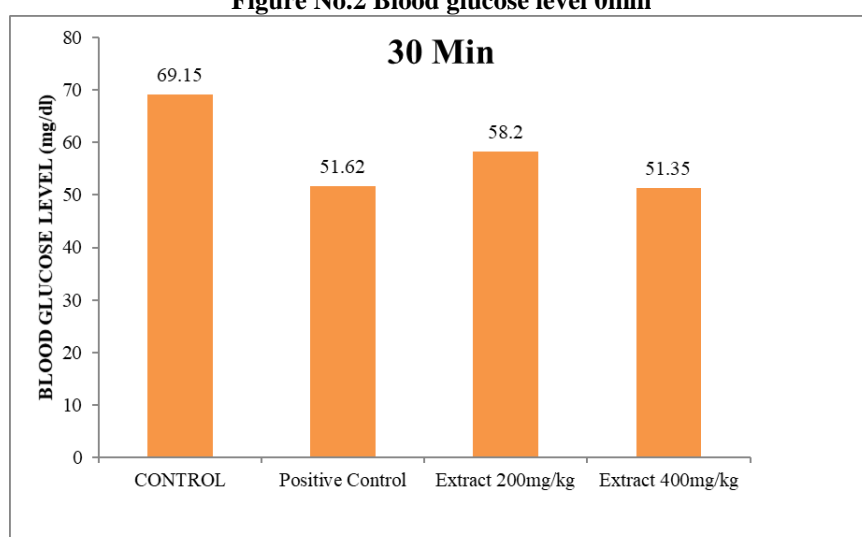


Figure No.3 Blood glucose level 30min

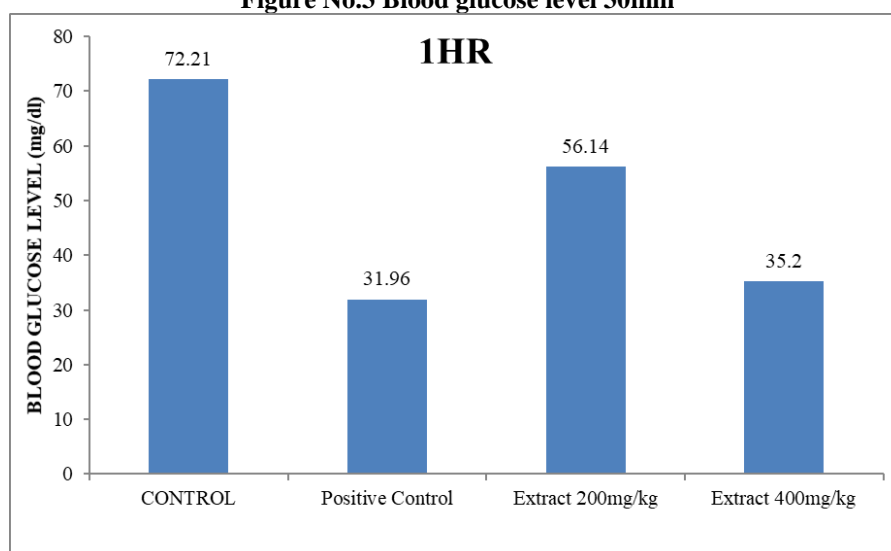


Figure No.4 Blood glucose level 1hr

Discussion: The hypoglycemic test results have shown Table No: which indicated ethanolic extract of *Acacia arabica* treated animals 200 & 400, significantly decreased in blood glucose level when compared to control and positive control.

Invivo antidiabetic study

Table No.4 Results of the effects of Ethanolic extract on blood Glucose levels

TREATMENT	BLOOD GLUCOSE LEVEL (mg/dl)		
	0 min	30min	1hr
Normal control 10 ml/kg P.O	78.14±1.125	72.0±1.105	71.6± 1.182
Negative control	263.2±1.51	260.1±2.062	265.1±2.0
Positive control (Glibenclamide 2mg/kg) P.O	253.25±2.136	132.41±1.6***	110±2.3***
EEAA 200 mg/kg P.O	256±2.1	241.2±1.154**	236.1±2.120**
EEAA 400 mg/kg P.O	261±2.05	168.0±2.68***	150.0±1.3***

(The values were expressed as Mean ± S.E.M. (n=6 animals in each group).

Discussion: The experimental results have indicated on Table the negative control group glucose levels were significantly increased when compared to each other groups. All the groups of animals were affected in diabetes, which indicated blood glucose levels were slight changes in the blood glucose level for normal control group at 7th and 14th days. On day 7th glucose levels were significantly decreased Glibenclamide 2mg/kg treated group when compared with control group at 7th and 14th days. The Ethanolic leaves extract of *Acacia arabica* treated groups 200 & 400 mg/kg were dose dependent manner decreased when compared with control group but positive control have more anti diabetic activity at 7th day.

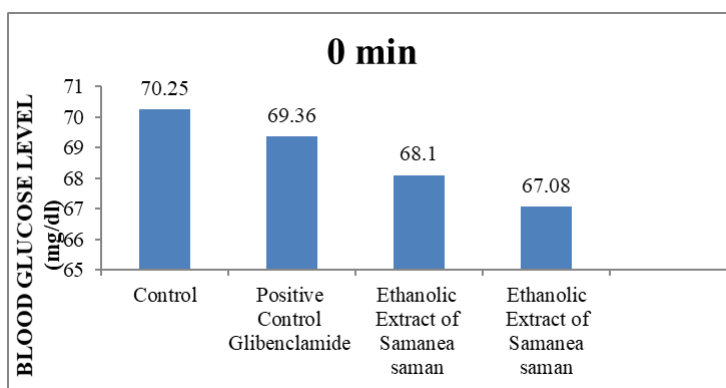


Figure No.5 Blood glucose level 0 min

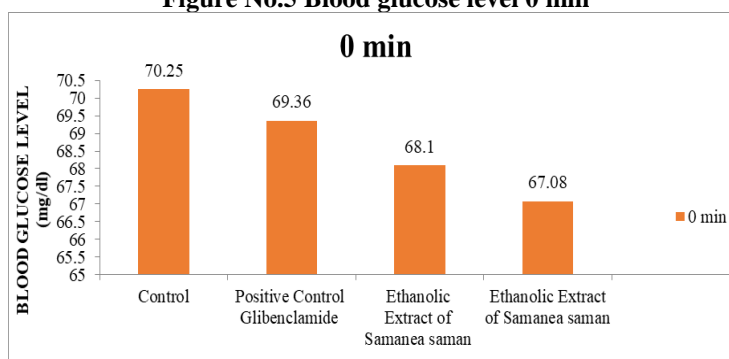


Figure No.6 Blood glucose level 30 min

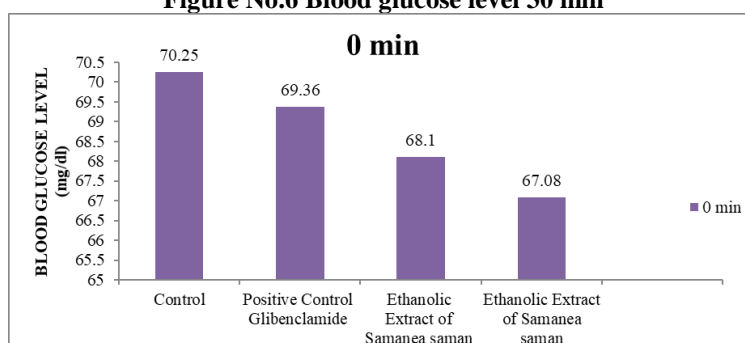


Figure No.7 Blood glucose level 1 hr

Discussion: The Ethanolic leaves extract of *Acacia arabica* at the dose level 400mg/kg have equipotent activity when compared with positive control at 7th day. The Ethanolic leaves extract of *Acacia arabica* 200 & 400 mg/kg have been expressed dose dependent anti diabetic action when compared to control and positive control. On day 14th, Ethanolic leaves extract of *Acacia arabica* treated animals 200 & 400 mg/kg significantly decreased and maintain the blood glucose level when compared to control and positive control.

Table No.5 Oral Glucose Tolerance Test

Treatment	DOSE mg/kg	Blood Glucose Level (mg/dl)						
		0 min	0.5hr	1hr	1.5hr	2hr	2.5hr	3hr
Control (CMC)	0.5%	68.05 ±1.141	143.2 ±1.325	184.3 ±1.120	173.2 ±10.42	153.2 ±4.121	150.0 ±3.142	128.3 ±9.36
Positive Control Glibenclamide	2	70.24 ±0.21	104.1 ±3.154**	112.1 ±3.24***	93.10 ±1.121***	80.14 ±3.011**	74.03 ±1.201***	70.50 ±3.512***
EEAA	200	65.10 ±1.109	123.5 ±1.001	142.3 ±1.121*	131.5 ±0.162*	121.3 ±0.101*	110.12 ±0.20**	103.0 ±2.106**
EEAA	400	67.01 ±2.141	111.2 ±0.156**	120 ±4.116**	100.0 ±2.211***a	90.12 ±3.251*** a	83.01 ±1.02***a	79.03 ±201***a

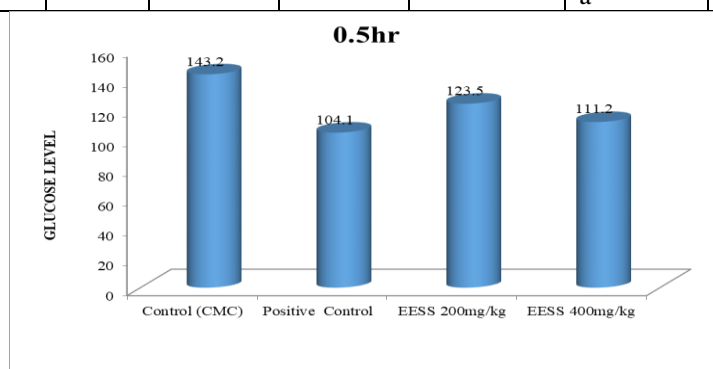


Figure No.8 Blood Glucose Level (mg/dl) 0.5hr

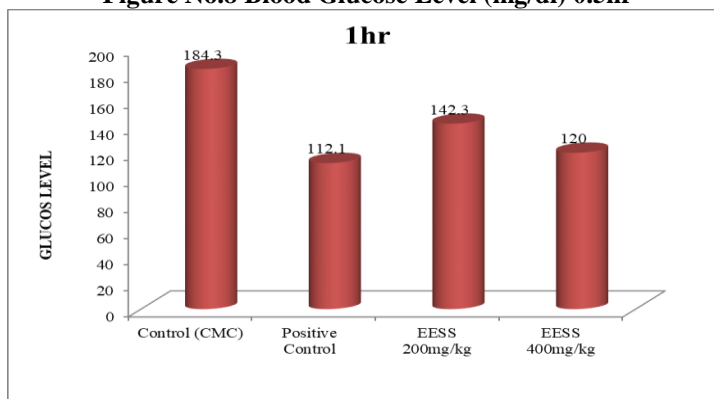


Figure No.9 Blood Glucose Level (mg/dl) 1hr

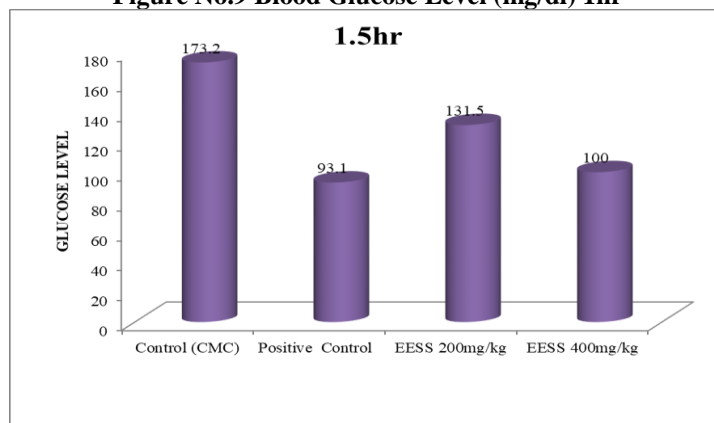


Figure No.10 Blood Glucose Level (mg/dl) 1.5hr

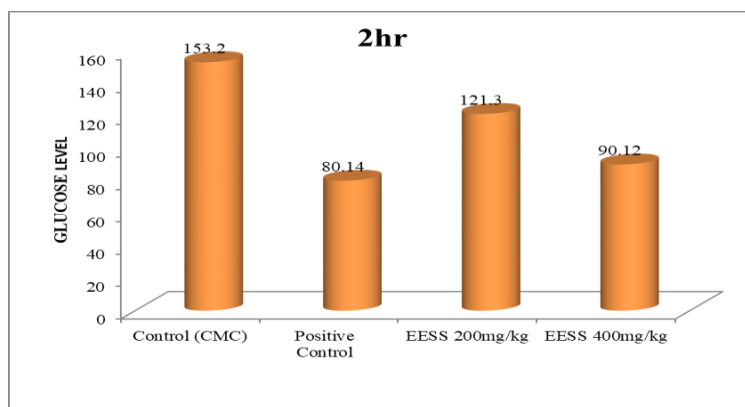


Figure No.11 Blood Glucose Level (mg/dl) 2hr

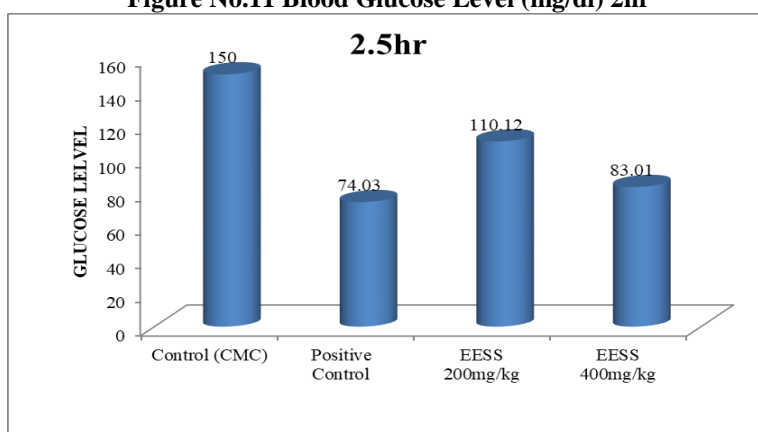


Figure No.12 Blood Glucose Level (mg/dl) 2.5hr

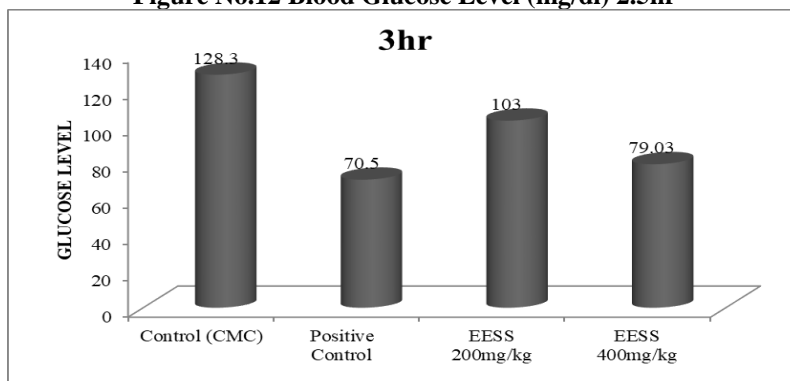


Figure No.13 Blood Glucose Level (mg/dl) 3hr

Discussion: Oral Glucose Tolerance Test (OGTT) results have been expressed on Table. Half hour after the glucose treatment, all the groups of animal blood glucose levels were significantly increased. The blood glucose levels were significantly decreased for, Ethanolic extract of *Acacia arabica* 200 & 400 mg/kg when compared to control and positive control at 1hour and each and every ½ hour blood glucose levels (200 mg/kg were changes in the dose dependent manner extract treated group of animals compared to control and positive control but 400mg/kg produce the equipotent activity).

CONCLUSION

This study revealed that the crude extract and solvent fractions of *Samanea saman* have showed significant lowering of blood glucose level on diabetic, Hypoglycemic and oral glucose loaded Rats and not permitted bodyweight loss of diabetic. The results also verified that inhibition of intestinal α -amylase by the extracts may contribute to the antihyperglycemic activity. The results give scientific support for the use of the plant in folk medicine for the management of diabetes and its associated complications. *Samanea saman* would be promising for further clinical studies in the management of DM. Further studies to find out the mechanism of this plant for its antidiabetogenic effect and there is a need for bioactivity guided investigation to isolate the lead compound responsible for the antidiabetic activity.

The present study suggested that the isolation of active constituents from Ethanolic extract of *Samanea saman* leaf and characterize the compounds by using preliminary phytochemical studies.

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