



Evaluation of Anti-diabetic Activity of *Bambusa Bambos* using Streptozocin Diabetic Model Induced

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Abstract:

Diabetes mellitus, commonly known as diabetes, is a group of metabolic disorders characterized by a high blood sugar level over a prolonged period of time. If left untreated, diabetes can cause many health complications. Acute complications can include diabetic ketoacidosis, hyperosmolar hyperglycemic state, or death. Serious long-term complications include cardiovascular disease, stroke, chronic kidney disease, foot ulcers, damage to the nerves, damage to the eyes and cognitive impairment. Herbal medicine is a drug or preparation made from plants or parts of plant (leaves, root, bark, seeds and flowers) valued for its medicinal, aromatic or savoury qualities to treat the symptoms of wide range of problems from depression to cold and flu. Herbal medicine has been widely used for thousands of years. One of them is *Bambusa bambos* which is very well known for its ethno pharmacological activities, also known as Bans in hindi and vansa in sanskrit. leaves were washed and dried was extracted with hydroalcohol. The present study represents the assessment of anti-hyperglycemic effects of the hydro alcoholic extract from *bambusa bambos* in streptozotocin (STZ) induced diabetic rats. The *bambusa bambos* hydroalcoholic extracts at the tested doses of 100, 200 and 400 mg/kg showed comparable activity with the standard. Metformin decreases hyperglycaemia primarily by suppressing glucose production by the liver. The streptozotocin induced diabetes is characterized by a severe loss in body weight. The decrease in the body weight of diabetic rats in this study was due to the loss or degradation of structural proteins. Insulin plays an important role in the regulation of protein synthesis and proteolysis in skeletal muscle. In the present study, diabetic control rats showed marked reduction in their body weight when compared to normal rats. The ability of the extract to protect body weight loss in diabetic rats seems to be the result of their ability to reduce hyperglycemia. During the study, it was found that the extract significantly ($p < 0.001$) controlled the blood glucose level in STZ-induced diabetic rats. The *bambusa bambos* hydroalcoholic extracts showed reduction in blood glucose level in STZ-induced diabetic rats when compared to the diabetic control group at a dose of 400 mg/kg,

Keywords: *Bambusa bambos*, Hydroalcoholic extracts, Metformin, Streptozotocin.

1. INTRODUCTION

Diabetes mellitus, commonly known as diabetes, is a group of metabolic disorders characterized by a high blood sugar level over a prolonged period of time. Symptoms often include frequent urination, increased thirst and increased appetite. If left untreated, diabetes can cause many health complications. Acute complications can include diabetic ketoacidosis, hyperosmolar hyperglycemic state, or death. Serious long-term complications include cardiovascular disease, stroke, chronic kidney disease, foot ulcers, damage to the nerves, damage to the eyes and cognitive impairment¹. Diabetes is due to either the pancreas not producing enough insulin, or the cells of the body not responding properly to the insulin produced. Insulin is a hormone which is responsible for helping glucose from food get into cells to be used for energy. There are three main types of diabetes mellitus: Type 1 diabetes results from failure of the pancreas to produce enough insulin due to loss of beta cells. This form was previously referred to as "insulin-dependent diabetes mellitus" or "juvenile diabetes". Type 2 diabetes begins with insulin resistance, a condition in which cells fail to respond to insulin properly. Type 2 diabetes is more common in older adults, but there is a great increase in the number of children of obesity which led to more cases of type 2 diabetes in younger people². The most common cause is a combination of excessive body weight and insufficient exercise. Gestational diabetes is the third main form, and occurs when pregnant women without a previous history of diabetes develop high blood sugar levels. In women with gestational diabetes, blood sugar usually returns to normal soon after delivery. However, there is a higher risk of suffering from type 2 diabetes if you have had gestational diabetes. As of 2019, an estimated 463 million people had diabetes worldwide (8.8% of the adult population), with type 2 diabetes making up about 90% of the cases. Rates are similar in women and

men. Trends suggest that rates will continue to rise. Diabetes at least doubles a person's risk of early death. In 2019, diabetes resulted in approximately 4.2 million deaths. It is the 7th leading cause of death globally. The global economic cost of diabetes-related health expenditure in 2017 was estimated at US\$727 billion. In the United States, diabetes cost nearly US\$327 billion in 2017. Average medical expenditures among people with diabetes are about 2.3 times higher. Damage to the kidneys, known as diabetic nephropathy, can lead to tissue scarring, urine protein loss, and eventually chronic kidney disease, sometimes requiring dialysis or kidney transplantation³. Damage to the nerves of the body, known as diabetic neuropathy, is the most common complication of diabetes. The symptoms can include numbness, tingling, sudomotor dysfunction, pain, and altered pain sensation, which can lead to damage to the skin^{1,2}. Diabetes-related foot problems (such as diabetic foot ulcers) may occur, and can be difficult to treat, occasionally requiring amputation. Additionally, proximal diabetic neuropathy causes painful muscle atrophy and weakness. Herbal medicine is a drug or preparation made from plants or parts of plant (leaves, root, bark, seeds and flowers) valued for its medicinal, aromatic or savoury qualities to treat the symptoms of wide range of problems from depression to cold and flu. Herbal medicine has been widely used for thousands of years. One of them is *Bambusa bambos* which is very well known for its ethno pharmacological activities, also known as Bans in hindi and vansa in sanskrit^{4,5}.

2. MATERIALS AND METHODS

2.1 Plant material :

The fresh flowers of *Bambusa bambos* was collected in the month of April from local area of Dausa city (Rajasthan). The plant authentication number is RUBL-211710 and BSI/AZRC/1.1202/Tech./2022-23(PI.-Id.)/35

2.2 Preparation of extract

leaves were washed and dried under shade at room temperature for one month. The dried leaves were powdered and stored in air tight container. 3 kg powder was extracted with hydro-alcohol in a ratio of 30:70 by soxhlet extraction at temperature of 65-75°C until the siphoning tube liquid become colourless. The remaining solvent was removed at 40-50°C in rotatory evaporator under reduced pressure to give solid extract which was then weighed to calculate percentage yield. Percentage yield of leaves extract was 12.7%. The dried extract was stored in air tight container at 4-8°C for further investigation⁶⁻⁸.

2.3 Experimental Animals

Swiss albino rats of either sex weighing 150-200 g were used for the study. They were maintained in the animal house of Bilwal medchem and research laboratory Pvt.Ltd. for experimental purpose. The animals were maintained under controlled conditions of temperature (23 ± 3°C), humidity (50 ± 5%). All the animals were acclimatized for seven days before the study. The animals were randomized into experimental and control groups and housed individually in sanitized polypropylene cages containing sterile paddy husk as bedding. They had free access to standard pellets as basal diet and water ad libitum. Animals were habituated to laboratory conditions for 48 hours prior to experimental protocol to minimize if any of non-specific stress. All the studies conducted were approved by the Institutional Animal Ethical Committee (IAEC) of Bilwal medchem and research laboratory Pvt.Ltd. (REG.No.-2005/PO/RcBT/S/18/CPCSEA), according to prescribed guidelines of Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Government of India.

2.4 Preparation of diabetic rats

Diabetes was induced by intraperitoneal of STZ in a single dose of 50 mg/kg body weight. STZ was dissolved in a freshly prepared 0.01 M citrate buffer (pH 4.5)⁸

2.5 Determination of acute toxicity (LD50):-

The acute toxicity Study for Hydro alcoholic extract of *bambusa bambos* was performed on

swiss albino rats of either sex weighing between 150-200 gm as per OECD Guidelines for duration of 72 hrs. The animals were fasted overnight prior to the experiment. The Up and Down method was adopted for acute toxicity studies^{6,9}.

2.6 Statistical analysis:-

The result were expressed as mean + SEM. The significance of difference between mean values for various treatment was tested using the impaired student "t" test. p<0.01 was considered as statistically significant. The data were subject to one-way analysis of variance (ANOVA).

3. RESULT

3.1 Acute Toxicity Study:

The acute toxicity Study for Hydro alcoholic extract of *bambusa bambos* was performed on swiss albino rats of either sex weighing between 150-200 gm as per OECD Guidelines for duration of 72 hrs. The animals were fasted overnight prior to the experiment. The Up and Down method was adopted for acute toxicity studies. After administration of Hydro alcoholic extract of *bambusa bambos*. No sign, symptoms and mortality of animals were observed during Acute Toxicity Studies. so, The maximum non lethal dose was observed 2000mg/kg animal body weight. Hence 1/5th, 1/10th and 1/20th of the dose was taken as effective dose (200 mg/kg and 100mg/kg body weight) for the extract to evaluate antidiabetic activities^{6,10,11}.

3.2 Experimental design for screening model streptozocine:^{8,12}

Group 1 Normal control group

Group 2 STZ induced group(50 mg/kg body weight)

Group 3 STZ 50 mg/kg body weight + HAEBB extract (100 mg/kg b.wt.)

Group 4 STZ 50 mg/kg body weight + HAEBB extract (200 mg/kg b.wt.)

Group 5 STZ 50 mg/kg body weight + HAEBB extract (400 mg/kg b.wt.)

Group 6 STZ 50 mg/kg body weight + Metformin (100 mg/kg b.wt.)

3.3 Experimental Protocol¹³⁻¹⁶

In the diabetic rats, 5 days after the induction of diabetes, animal were divided into six groups each having six rats. Non diabetic animals are

grouped for control. Total of 6 groups of animal of six each were used for study

Group I: Received 2% w/v Gum acacia 1 ml per/kg orally served as control group (non diabetic control).

Group II: Served as STZ induced diabetic control received 2% w/v gum acacia 1 ml/kg orally for 20 days.

Group III: STZ induced diabetic animal received hydro alcoholic extract of *bambusa bambos*

leaves 100 mg/per kg body weight once daily orally for 20 days.

Group IV : STZ induced diabetic animal received hydro alcoholic extract of *bambusa bambos*

leaves 200 mg/per kg body weight once daily orally for 20 days..

Group V : STZ induced diabetic animal received hydro alcoholic extract of *bambusa bambos*

leaves 400 mg/per kg body weight once daily orally for 20 days..

Group VI : STZ induced diabetic animal received the standard drug Metformin 100 mg/kg

body weight once daily orally for 20 days..

3.4 Streptozotocin induced diabetic rats

Administration of STZ leads to elevation in blood glucose level in rats. In STZ induced diabetic rats, the fasting blood glucose level was found to increase from 298 to 309 mg/dl. Oral administration of HAEBB 100 mg/kg showed a significant ($P<0.01$) decrease in blood glucose level from 283 ± 2.3 to 278.2 ± 3.5 as compared with diabetic control. The daily treatment of rats with HAEBB leads to dose dependent fall in blood glucose level . HAEBB at all doses showed significant ($P<0.01$) decrease in blood glucose level but effect at 400 mg/kg was superior. In the standard drug treated group, blood glucose level was found to decrease throughout the study.

Table 1: Effect of the Bambusa leaves HAEBB on blood glucose level of Streptozocine rats after treatment.

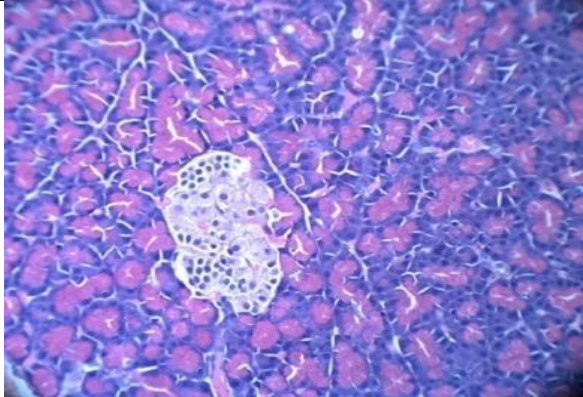
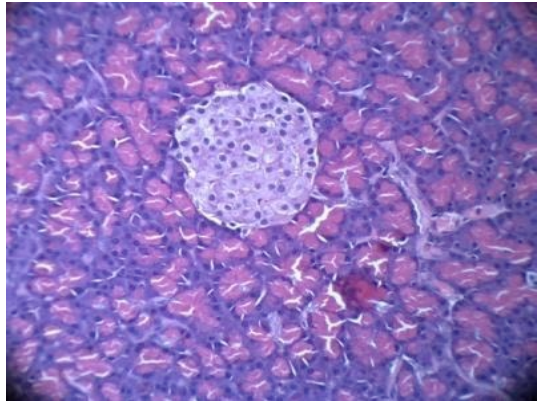
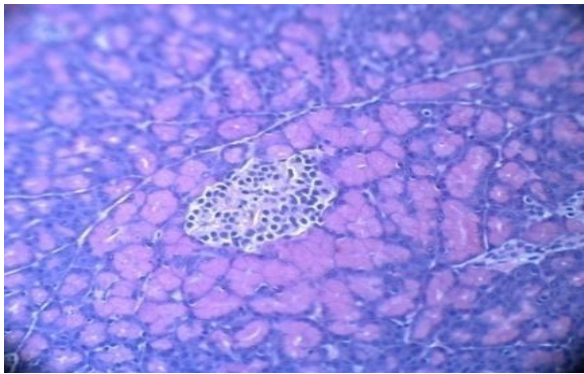
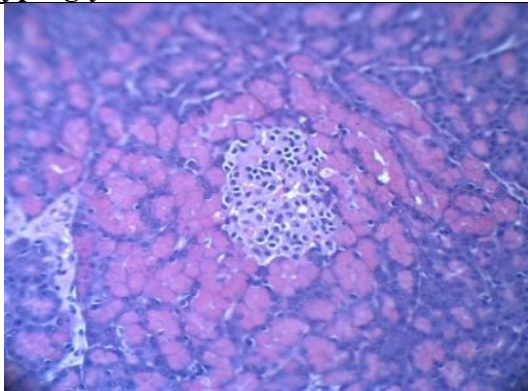
Treatment	Group	Blood glucose mg/dl			
		0 min	30 min	60 min	90 min
Group I	Normal Control	75±1.6	77±3.6	80±1.5	83±1.9
Group II	STZ(50mg/kg b.wt)	160±3.4	158.2±2.6	155.2±1.6	152.2±2.6
Group III	STZ+HEBB (100 mg/kg)	145±2.3	147.6±2.8*	148.2±3.1*	149.2±3.5*
Group IV	STZ+HEBB (200 mg/kg)	135±2.7	137.6±2.6**	138±2.9**	139±2.8**
Group V	STZ+HEBB (400 mg/kg)	125±2.3	126±2.9***	128±2.2***	130±2.5***
Group VI	STZ+metformin(100 mg/kg b.wt)	105±3.1	103.2±2.2***	101.2±1.4***	100.2±2.4***

Table No 1: Data Analysis: Statistical interpretation was calculated from normal animal group evaluated with test control animal. All the values are expressed as mean ±SEM n = 6 in each group. Values are significantly different from control . ANOVA was calculated. $P\leq 0.05$ it was statically significant. The results were expressed as mean ± SD (n = 6)

Table 2: Effect of the Bambusa leaves HAEBB on blood glucose level of Streptozocine rats after treatment.

Treatment	Group	Blood glucose mg/dl			
		0 min	5 th days	10 th days	20 th days
Group I	Normal Control	73±1.6	76±3.6	79±1.5	82±1.9
Group II	STZ(50mg/kg b.wt)	298±3.4	300.2±2.6	305.2±1.6	309.2±2.6
Group III	STZ+HEBB (100 mg/kg)	283±2.3	280.6±2.8*	279.2±3.1*	278.2±3.5*
Group IV	STZ+HEBB (200 mg/kg)	281±2.7	265.6±2.6**	267±2.9**	270±2.8**
Group V	STZ+HEBB (400 mg/kg)	279±2.3	255±2.9***	257±2.2***	258±2.5***
Group VI	STZ+metformin(100 mg/kg b.wt)	260±3.1	240.2±2.2***	244.2±1.4***	247.2±2.4***

Table 2: Data Analysis: Statistical interpretation was calculated from normal animal group evaluated with test control animal. All the values are expressed as mean ±SEM n = 6 in each group. Values are significantly different from control . ANOVA was calculated. P≤0.05 it was statically significant. The results were expressed as mean ± SD (n = 6)

	
<p>Figure 1: Group 1- Photomicrograph of normoglycaemic rats</p>	<p>Figure 2: Group 2- Photomicrograph of hyperglycaemic rats</p>
	
<p>Figure 3: Group 3- Photomicrograph of HAEBB extract(100 mg/kg) treated rats</p>	<p>Figure 4: Group 4- Photomicrograph of HAEBB extract(200 mg/kg) treated rats</p>

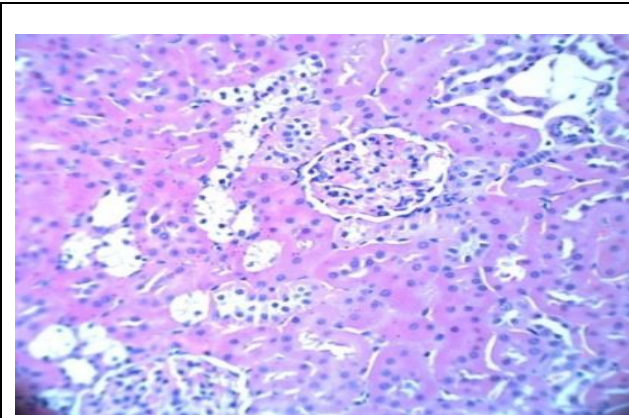


Figure 5: Group 5- Photomicrograph of HAEBB extract(400 mg/kg) treated rats

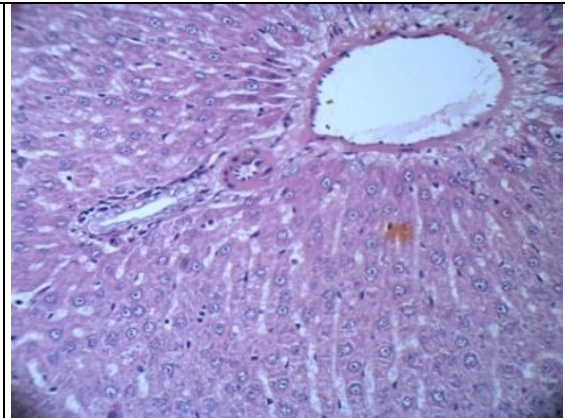


Figure 6: Group 6- Photomicrograph of metformin (100 mg/kg) treated rats

4. DISCUSSION

Recently diabetes is one of the leading diseases around the globe. Management of diabetes is being a tough task with the synthetic medicines as they have many side effects. The interest has been shifted towards the medicinal plants used as a remedy for reducing the risk of Medicinal plants, the potential sources of bioactive agents are gaining adequacy worldwide²⁰. In the present scenario, the scientists have emphasized the potential herbal extracts and initiated extensive research to observe their effective and protective role in the diseased animal models²¹.

The present study represents the assessment of anti-hyperglycemic effects of the hydroalcoholic extract from *bambusa bambos* in streptozotocin induced diabetic rats. Pancreas is the primary organ involved in sensing the organism's dietary and energetic states via glucose concentration in the blood and in response to elevated blood glucose, insulin will be secreted.

However, streptozotocin is a naturally occurring nitrosourea product and it is widely used to induce diabetes in experimental animals. Usually, the intraperitoneal injection of a single dose (50 mg/kg, b. wt) exerts damage of pancreatic β -cells resulting in necrosis within 48-72 h and causes a permanent hyperglycemia. When there are not enough available β -cells to

supply sufficient insulin to meet the needs of the body, insulin-dependent diabetes results^{8,22-24}

The *bambusa bambos* hydroalcoholic extracts at the tested doses of 100, 200 and 400 mg/kg showed comparable activity with the standard. Metformin decreases hyperglycaemia primarily by suppressing glucose production by the liver. The streptozotocin induced diabetes is characterized by a severe loss in body weight. The decrease in the body weight of diabetic rats in this study was due to the loss or degradation of structural proteins. Insulin plays an important role in the regulation of protein synthesis and proteolysis in skeletal muscle. In insulin resistance or deficiency state, muscle wasting and weight loss in diabetic rats results from the excessive catabolism of protein which provides amino acids for gluconeogenesis²⁴.

In the present study, diabetic control rats showed marked reduction in their body weight when compared to normal rats. Histopathology also shows improvement. Mainly in high dose which was equivalent to metformin. The weight loss was reverted by administration of extract to the diabetic rats for a period of 20 days. The ability of the extract to protect body weight loss in diabetic rats seems to be the result of their ability to reduce hyperglycemia. During the study, it was found that the extract significantly ($p < 0.001$) controlled the blood glucose level in STZ-induced diabetic rats. The *bambusa*

bambos hydroalcoholic extracts showed reduction in blood glucose level in STZ-induced diabetic rats when compared to the diabetic control group at a dose of 400 mg/kg,

5. CONCLUSION:

Based on the results obtained in the present study, it concluded that the hydroalcoholic extract of *bambusa bambos* possesses anti hyperglycaemic properties due to the presence of much more quantity of carbohydrate. Therefore, these medicinal plants could be considered as a potential and alternative approach for the treatment of diabetes. However, further investigations are needed to identify the lead molecule and to elucidate absolute mechanism of action for antidiabetic effect. In this study, investigation has also opened avenues for further research to the development of potent phytomedicine for diabetes mellitus from the *bambusa bambose*.

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