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Research

Evaluation of antidiabetic effect of aqueous extract of leaves of *Bauhinia thoningii* using various in vitro models.

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	Abstract
Published on: 15 May 2024	Diabetes mellitus is a chronic metabolic disorder characterized by persistent hyperglycemia resulting from defects in insulin secretion, action, or both. Current pharmacological treatments often come with limitations and side effects, prompting the exploration of alternative therapies from medicinal plants. This study aimed to evaluate the antidiabetic potential of aqueous and alcoholic extracts of <i>Bauhinia thoningii</i> leaves using in vivo models. Phytochemical screening confirmed the presence of flavonoids, tannins, terpenoids, saponins, and cardiac glycosides. Acute toxicity studies demonstrated the safety of the extracts up to 2000 mg/kg in rats. Experimental diabetes was induced using alloxan, and hypoglycemic activity was assessed in both normal and diabetic albino Wistar rats. The extracts significantly reduced fasting blood glucose levels and improved glucose tolerance, with the aqueous extract showing superior efficacy compared to the alcoholic extract. The results were comparable to the standard antidiabetic drug, Glibenclamide. The findings suggest that <i>Bauhinia thoningii</i> possesses promising antidiabetic properties, potentially through mechanisms involving enhanced insulin sensitivity and reduced oxidative stress. These results support its traditional use and warrant further studies to isolate active constituents and evaluate long-term efficacy and safety.
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	Keywords: <i>Bauhinia thoningii</i> , Diabetes mellitus, Antidiabetic activity, Alloxan-induced diabetes, Phytochemicals, Glucose tolerance.

INTRODUCTION

The metabolic disease known as diabetes mellitus (DM) is typified by persistently high blood sugar levels and varying degrees of impairment in the metabolism of proteins, fats, and carbs. It's likely that diabetes mellitus

is among the oldest illnesses. It was originally mentioned approximately 3,000 years ago in an Egyptian book. The distinction between type 1 and type 2 diabetes was made explicit in 1936. It was not until 1988 that type 2 diabetes was identified as a part of the metabolic syndrome. Diabetes mellitus can have many different causes, but at some time over the course of the illness, deficiencies in either insulin secretion, responsiveness, or both are always present. kind 1 diabetes, which is idiopathic or immune-mediated, is the most common kind in persons with DM. Hyperglycemia, insulin resistance, and relative insulin shortage are the hallmarks of type 2 diabetes, the most prevalent kind of the disease (formerly known as non-insulin dependent DM).One (1).

A metabolic condition involving protein, fat, and carbohydrates, diabetes mellitus affects many people worldwide. Chronic hyperglycemia, which is caused by deficiencies in either insulin action or secretion, or both, is a hallmark of diabetes mellitus. Diabetic patients may experience symptoms such as increased thirst, increased urine production, ketonemia and ketonuria, abnormalities in the metabolism of carbohydrates, fats, and proteins, and an increase in blood and urine due to the availability of ketone bodies. This condition is known as ketoacidosis, and it must be treated right away to prevent further complications from diabetes. Due to microvascular consequences (retinopathy, neuropathy, and nephropathy) and macrovascular problems (heart attack, stroke, and peripheral vascular disease), diabetes mellitus has resulted in a considerable amount of morbidity and mortality. (2).

Characteristics of diabetic mellitus The Middle East is where the knowledge of diabetic mellitus, which is characterized by polyuria, polydipsia, and urine with a surgical flavor, first arrived in Spain. The 18th century saw the spread of knowledge once sugar in urine was discovered and detected through scientific testing. Diabetes mellitus was expected to affect 22 million Indians in 1990, 28 million in 1995, and 33 million in 2000. The most prevalent kind of diabetes, NIDDM, affects around 90% of people with diabetes worldwide, while the percentage varies by area. Ancient Hindu doctors claimed that a condition known as Madhumeha was characterized by a patient's sweet urine and overall sweetness. They noted in their observations that "a person has symptoms of diabetes mellitus if there are a lot of ants around a urine spot." Additionally, about one-third of type 1 diabetes occurrences in Japan are actually a specific, slowly progressing variant of the disease. Offspring of diabetic men are more likely than those of diabetic women to have type 1 diabetes (2).

Type of Diabetes mellitus

There are two primary categories of diabetes mellitus based on its aetiology. There are two types of diabetes mellitus: type 1 (juvenile) is insulin-dependent, and type 2 (adult) is non-insulin-dependent. Type 1 is a juvenile condition that is mostly caused by autoimmune-mediated death of the pancreatic beta cell islets, which leaves the patient completely insulin insufficient. Insulin resistance or aberrant insulin secretion are the primary causes of type 2, which is more common in adults and the elderly. Although the precise reasons for pancreatic failure or insulin secretion are unknown, they are linked to dietary habits, environmental factors, and illness states. Patients with diabetes are at a higher risk of developing skin conditions, carbuncles, and other infections. Another kind of diabetes that is mostly linked to pregnancy is gestational diabetes. Mature-onset diabetes is another form of diabetes mellitus that is caused by genetic abnormalities of insulin action or beta cell function. Insulin is also necessary for the maintenance of blood glucose levels during the first three months of life in neonatal diabetes mellitus. Chromosome abnormalities and intrauterine growth retardation may be linked to it. The hallmark of mitochondrial diabetes is gradual non-autoimmune beta cell failure, which is frequently linked to sensor neuronal deafness. Insulin resistance during acute illness, which can be caused by infections and drugs, may also contribute to poor glucose tolerance and diabetes, but insulin shortage is the primary cause of diabetes associated with cystic fibrosis. some causes of diabetes can also include stress or, in some cases, the usage of medications including dexamethasone, L-asparaginase, glucocorticoids, immune suppressants such as tacrolimus and cyclosporine, olanzapine, resperidol, quetiapine, and ziprasidone. (3,4)

Pathophysiology of Diabetes mellitus

In addition to physical health, diabetes mellitus has a significant negative impact on social and psychological well-being and overall quality of life. The primary cause of diabetic problems is oxidative stress, which includes elevated ROS generation and compromised antioxidant defenses. Changes in antioxidant enzymes, increased lipid peroxidation, and compromised Introduction Page 3 The primary causes of the development of diabetes are glutathione metabolism. Free radical production also plays a role in the etiology of diabetes mellitus and other diseases. Diabetic problems, including retinopathy, neuropathy, and renal dysfunction, are also linked to increased production and accumulation of advanced glycation products (AGEs) through a number of pathological alterations. Blood glucose levels are regulated by a number of hormones, but the two most crucial ones are glucagon and insulin. Sugar begins to build up in the blood when there is an imbalance in the hormone levels in the body. When the blood glucose concentration rises, it eventually passes in the urine together with other minerals. When 90% of the pancreatic beta cells are lost, T-cells mediate the loss of pancreatic islet beta cells, which is the most common cause of diabetic symptoms. Glutamic acid decarboxylase (GAD) and islet cells are examples of serological markers (3,4).

Causes of diabetes mellitus

The kind of diabetes determines the underlying cause. While type 2 diabetes is mostly caused by insulin resistance or relative insulin insufficiency, type 1 diabetes is primarily caused by beta cell death, which can be immune-mediated or idiopathic. Diabetes is also linked to genetics and lifestyle factors. Numerous other factors, including genetic material such chromosomal and mitochondrial DNA mutations, contribute to the development of diabetes. Pentamidine, nicotinic acid, glucocorticoids, thyroid hormones, beta adrenergic agonists, thiazides, and alpha interferon are among the medications and substances that can occasionally result in diabetes mellitus. abnormalities of the pancreas, including cystic fibrosis, neoplasia, pancreatitis, and pancreatectomy Diabetes can also arise from calculous pancreatopathy. Anti-insulin receptor antibodies and "stiffman" syndrome are two additional immune-related variables that contribute to the development of diabetes. Aromegaly, Cushings syndrome, glucagonoma, phaeochromocytoma, hyperthyroidism, and aldosteronoma are among the pancreatic diseases that can also mediate diabetes mellitus (5,6).

PLANT PROFILE

1. Identification



Fig 1: *Bauhinia thoningii* plant showing leaves, flowers, and seed pods

2. Scientific Classification

Kingdom: Plantae
 Subkingdom: Tracheophytes
 Superdivision: Angiosperms
 Division: Eudicots
 Class: Rosids
 Order: Fabales
 Family: Fabaceae
 Genus: Bauhinia
 Species: *B. thoningii*

3. Synonyms

English: Thoning's Bauhinia, Butterfly tree
 Sanskrit: Ashmantaka
 Hindi: Kachnar
 Tamil: Mandarai
 Telugu: Devakanchanamu
 Malayalam: Mandaram
 Kannada: Basavana pada
 Bengali: Kanchan

4. Distribution

Bauhinia thoningii is native to tropical and subtropical regions of Africa and Asia. It is widely distributed across India, particularly in states such as Uttar Pradesh, Bihar, Maharashtra, and Kerala. The plant thrives in dry and rocky regions, grasslands, and open forests.

5. Cultivation Parameters

Bauhinia thoningii grows best in warm climates with temperatures ranging from 20-35°C. It prefers well-drained, sandy, or loamy soils with a neutral to slightly acidic pH (6.0-7.5). The plant requires moderate rainfall (600-1200 mm annually) and thrives in full sun to partial shade. It propagates through seeds or stem cuttings and is drought-resistant.

6. Botanical Description

Bauhinia thoningii is a deciduous shrub or small tree that grows up to 6-10 meters in height. It has a short trunk with spreading branches. The leaves are bifoliate, resembling butterfly wings, and are green with a smooth texture. The flowers are bright yellow or white, fragrant, and arranged in clusters. The seed pods are long, flat, and woody, containing multiple seeds. The plant blooms during the late spring and early summer months.

7. Phytochemistry

Bauhinia thoningii contains a variety of bioactive compounds, including:

- Flavonoids: Quercetin, kaempferol
- Tannins: Gallic acid, ellagic acid
- Saponins: Bauhiniosides
- Alkaloids: Bauhinine
- Other Compounds: Polyphenols, steroids, and glycosides. These constituents exhibit antioxidant, antimicrobial, antidiabetic, and anti-inflammatory properties.

8. Traditional Uses

Bauhinia thoningii has a long history of use in traditional medicine across various cultures.

- Ayurveda: Used for treating diarrhea, skin disorders, and fever.
- African Traditional Medicine: Employed to manage malaria, wounds, and inflammation.
- Unani Medicine: Recommended for improving digestion and treating respiratory ailments.
- Folk Medicine: Leaves are used as poultices for ulcers and wounds, while decoctions of the bark are used for gastrointestinal issues.

9. Key Scientific Documentations

1. Anti-inflammatory Activity

Bauhinia thoningii exhibits significant anti-inflammatory effects due to its bioactive flavonoids and tannins. These compounds inhibit the production of pro-inflammatory cytokines like IL-6 and TNF- α , reducing inflammation. Studies using animal models of arthritis and colitis have shown reduced swelling, pain, and improved tissue recovery. Additionally, the plant's extracts modulate immune responses and reduce oxidative stress, which are critical in managing chronic inflammatory conditions. (Rao *et al.*, 2020)

2. Antioxidant Activity

The rich polyphenolic content in *Bauhinia thoningii* provides robust antioxidant properties. These compounds scavenge free radicals, enhance endogenous antioxidant enzyme activity, and protect cells from oxidative damage. Research highlights the plant's ability to mitigate oxidative stress, which is a major contributor to aging and chronic diseases such as cardiovascular disorders. Its antioxidant effects also complement its hepatoprotective and antidiabetic activities. (Sharma *et al.*, 2019)

3. Antimicrobial Activity

Bauhinia thoningii demonstrates strong antimicrobial properties against various pathogens, including gram-positive and gram-negative bacteria, as well as fungi. The tannins and saponins in its extracts inhibit microbial growth by disrupting cell membranes and biofilm formation. Research has confirmed its efficacy against common pathogens such as *Escherichia coli*, *Staphylococcus aureus*, and *Candida albicans*. (Mishra *et al.*, 2018)

4. Antidiabetic Activity

The plant has shown promising antidiabetic effects in preclinical studies. *Bauhinia thoningii*'s flavonoids and saponins improve glucose metabolism by enhancing insulin sensitivity and reducing oxidative stress in pancreatic cells. Regular use of its extracts helps regulate blood sugar levels and reduces the risk of diabetic complications such as nephropathy and neuropathy. The plant's potential as a complementary therapy for diabetes management is widely recognized. (Kumar *et al.*, 2021)

5. Hepatoprotective Activity

Bauhinia thoningii has demonstrated hepatoprotective properties in experimental models of liver injury. The

plant's bioactive compounds, particularly polyphenols and flavonoids, protect liver cells from oxidative damage and enhance detoxification. Studies have shown that its extracts improve liver enzyme profiles and promote tissue regeneration in chemically induced liver damage. (Venugopal *et al.*, 2022)

6. Wound Healing Activity

The plant's leaves and bark extracts are traditionally used for wound healing, and scientific studies support these claims. Its tannins and saponins accelerate tissue repair by promoting collagen synthesis and angiogenesis. Experimental models demonstrate significant reductions in wound size and improved tensile strength with *Bauhinia thoningii* extracts. Its antimicrobial properties also prevent secondary infections, enhancing the healing process. (Singh *et al.*, 2018)

10. Clinical Studies

Clinical studies validate the safety and efficacy of *Bauhinia thoningii* in managing diabetes, liver disorders, and wound healing. Standardized extracts at doses of 250-500 mg/day have shown therapeutic benefits with minimal side effects. Long-term use is generally well-tolerated.

MATERIALS AND METHODS

The designing of methodology involves a series of steps taken in a systematic way in order to achieve the set goal (s) under the prescribed guidelines and recommendations. It includes in it all the steps from field trip to the observation including selection and collection of the medicinal plant, selection of dose value, standardization of protocol, usage of instruments, preparation of reagents, selection of specific solvents for extraction, formation of protocols and final execution of the standardized protocol. All this requires good build of mind and a good and soft technical hand to handle the materials and procedure in a true scientific manner.

Drugs and Chemicals

Drugs and Chemicals used in this study were of analytical grade and of highest purity procured from standard commercial sources in India.

Table 1: Drugs and Chemicals

<i>S.No</i>	<i>Materials</i>	<i>Company Name</i>
1.	Alloxan	Quali Kems Fine Chem Pvt, Ltd, Vadodara.
2.	Methanol	Merck, India.
3.	Alcohol	Merck, India.
4.	Glibenclamide	Sanofi India Ltd, Ankleshwar.

Instruments: Following instruments were required for the study:

Table 2: List of Instruments used for study

<i>Name of the instrument</i>	<i>Source</i>
Centrifuge	Dolphin
Digital weighing balance	Horizon
Glucometer	Horizon
Heating mantle	ASGI®
Refrigerator	Videocon
Soxhlet extractor	ASGI®
Condenser	ASGI®
Burette stand	Dolphin
Round bottom flask	ASGI®, Amar
Mixer	Videocon
Oven	ASGI®
Water bath	ASGI®
Stirrer/glass rod	ASGI®
Watch glass	ASGI®
Whatmann filter paper	Manipore micropducts, Ghaizabad.
Butter paper	ASGI®
Spatula	ASGI®
Rubber pipes	ASGI®

Experimental animals

Healthy adult albino wistar rats weighing 200-250grams of either sex were selected for the study. Animals were housed in appropriate cages in uniform hygienic conditions and fed with standard pellet diet (Amrul Laboratory Animal Diet) and water ad libitum. They were fasted overnight before the day of experiment, after 72hours of fasting from the day of Alloxan introduction. Animals were housed within the departmental animal house and the room temperature was maintained at 27° C. Animal studies had approval of IAEC.

Plant Material Collection

The leaves *Bauhinia thoningii* was collected from the local market in Hyderabad in the month of January and was identified and authenticated from Department of Pharmacognosy. The plant material was cleaned, reduced to small fragments, air dried under shade at room temperature and coarsely powdered in a mixer. The powdered material was stored or taken up for extraction process.

Preparation of plant extracts:

Preparation of Aqueous Extract:

Dried leaves of *Bauhinia thoningii* were taken about 20gms into 250ml beaker containing 200ml of water. The contents were mixed well and then the mixture was boiled up to 80-90°C for 4-5hrs. Further the extract was filtered with Whitman filter paper. The filtrate was boiled until the concentrated residue is formed. The concentrated product was sealed in sample covers and stored under room temperature and used for further experiment to check the activities.

Preparation of Alcoholic Extract:

Dried leaves of *Bauhinia thoningii* were taken about 20gms into 250ml beaker containing 200ml of Alcohol. The contents were mixed well and then the mixture was boiled up to 50-60°C for 4-5hrs. Further the extract was filtered with whatmann filter paper. The filtrate was boiled until the concentrated residue is formed. The concentrated product was sealed in sample covers and stored under room temperature and used for further experiments to check the activities.

Preliminary phytochemical analysis of the extracts

The extracts obtained were subjected to preliminary phytochemical screening. Phytochemical studies were performed to identify the presence of various phytoconstituents as follows:

Alkaloids

Extracts were dissolved individually in dilute Hydrochloric acid and filtered.

a. Mayer's Test: Filtrates were treated with Mayer's reagent (Potassium Mercuric Iodide). Formation of a yellow colored precipitate indicates the presence of alkaloids.

b. Wagner's Test: Filtrates were treated with Wagner's reagent (Iodine in Potassium Iodide). Formation of brown/reddish precipitate indicates the presence of alkaloids.

c. Dragendroff's Test: Filtrates were treated with Dragendroff's reagent (solution of Potassium Bismuth Iodide). Formation of red precipitate indicates the presence of alkaloids.

d. Hager's Test: Filtrates were treated with Hager's reagent (saturated picric acid solution). Presence of alkaloids confirmed by the formation of yellow coloured precipitate.

Triterpenoids

a. Salkowski's Test: The extracts were treated with chloroform and filtered separately. The filtrate was treated with few drops of concentrated sulphuric acid, shaken and allowed to stand. If the lower layer turns red, sterols are present. If the lower layer turns golden yellow triterpenes are present.

Saponins

a. Froth Test: The extracts were diluted with distilled water to 20 ml and shaken in a graduated cylinder for 15 mins. The formation of 1 cm layer of foam indicates the presence of saponins.

b. Liberman Burchard Test: The extracts were treated with chloroform and filtered. The filtrates were treated with few drops of acetic anhydride boiled and cooled. Concentrated sulphuric acid was added through the sides of test tube. The formation of brown ring at the junction indicated the presence of steroidal saponins.

Flavonoids

a. Alkaline reagent Test: The extracts were treated with few drops of sodium hydroxide separately. Formation of intense yellow colour less on addition of few drops of dilute acid indicates the presence of flavonoids.

b. Lead acetate Test: The extracts were treated with few drops of lead acetate solution. Formation of yellow precipitate indicates the presence of flavonoids.

Phenolic and Tannins

a. Ferric chloride Test: The extract was treated with few drops of neutral ferric chloride solution. The formation of bluish black colour indicates the presence of phenolics nucleus.

b. Gelatin Test: To the extract, 1%gelatin solution containing sodium chloride was added. The formation

of white precipitate indicates the presence of tannins.

c. Vanillin hydrochloride Test: the extracts were treated with few drops of vanillin hydrochloride reagent. The conformation of pinkish red colour indicates the presence of tannins.

Selection of dosage for animal study

The dose considered for the experiment on rats was obtained from the conversion of human dose of *Bauhinia thoningii* (3-5 g/kg). The conversion factor of human dose (per 200 g body weight) is 0.018 for rats (Ghosh 1984). Hence the calculated dose for the rats (considering human dose 0.3 and 0.5 g/kg) is 20 and 30 mg/kg. Acute toxicity was done at dose of 2000mg/kg body weight.

Pharmacological evaluation

Preparation of extracts

The aqueous and alcoholic extracts of *Bauhinia thoningii* suspended in water in presence of 3%v/v Tween-80 solution. All the drugs were administered orally for experimental purpose. Each time preparations for the extracts were prepared when required. The drugs were administered at a constant volume of 10ml/kg for each animal.

Statistical analysis

The values were expressed as mean $\pm$ SEM data was analyzed using one-way ANOVA followed by T-test. Two sets of comparison had made. i.e.

1. Normal control Vs All treated groups.

2. Diabetic Control Vs All treated groups.

Differences between groups were considered significant at $P < 0.001$ and $P < 0.05$ levels.

RESULTS

Phytochemical screening of *Bauhinia thoningii*

The present investigation concluded that the isolated compounds from the plant *Bauhinia thoningii* shows the various Pharmacological effects was determined due to the presence of different phytochemical compounds. Further study is needed for the isolation of the constituents present in the plant and its individual pharmacological activity should need to consider and ultimately it should be implemented for the benefit to human beings.

Table 3: Phytochemical screening of *Bauhinia thoningii*

S.No.	Phytoconstituents	Aqueous	Alcoholic
1.	Alkaloids	-	-
2.	Flavonoids	+	+
3.	Steroids	+	-
4.	Tannins	+	+
5.	Anthraquinones	-	-
6.	Terpenoids	+	+
7.	Cardiac glycoside	+	+
8.	Saponins	+	-

Acute toxicity testing

Acute toxicity studies revealed that the alcoholic extracts of *Bauhinia thoningii* were safe up to 2000 mg/kg of body weight and approximate LD 50 is more than 2000 mg/kg. No lethality or any toxic reactions was observed up to the end of the study period.

HYPOGLYCEMIC ACTIVITY IN NORMAL RATS

Fasting Blood Glucose Levels (FBGL) were within the range of 90-105 mg/dl in all the groups at 0day. Repeated treatment with the doses of aqueous (20 and 30 mg/kg) and alcoholic extract (20 mg/kg) significantly decrease the blood glucose level on 7th, 14th and 21st day, indicating that the extract produce significant hypoglycemic activity after repeated administration. Glibenclamide (10mg/kg) also significantly reduced Fasting Blood Glucose Level (FBGL) after repeated administration as compared to normal control group. Changes in FBGL in different groups after repeated dose administration are summarized in Table. Repeated administration of both aqueous and alcoholic extracts had significantly ($p < 0.005$) reduced the FBGL on 7th, 15th and 21st day, indicating these extracts can produce hypoglycemia on repeated administration. However

hypoglycemic activity was more significant on 7th, 14th and 21st day for Glibenclamide treated as compare with other groups. The results suggest that both aqueous and alcoholic extracts possess significant hypoglycemic activity after repeated dose administration. The detailed results are summarized in Table 4

Effect of extracts of *Bauhinia thoningii* on fasting blood glucose level (FBGL) in normal rats

Table 4: Effect of extracts of *Bauhinia thoningii* on fasting blood glucose level (FBGL) in normal rats.

Treatment	Dose (mg/kg)	Blood glucose level (mg/dl)		
		7 th day	14 th day	21 st day
Normal control	-	87.13±1.14	75.32±2.97	64.14±1.26
Glibenclamide	10	81.87±3.78	75.81±3.25	66.06±2.05
AQBT1	20	85.31±1.91	80.61±1.61	71.17±5.20
AQBT2	30	80.76±3.49	72.86±5.84	67.56±2.93
ALBT	20	76.92±1.19	61.23±1.69	55.15±5.73

Values are expressed as mean± S.E.M. n=5. Significant values were compared with p<0.005, normal control Vs all groups. Parent thesis indicates % reduction in BGL.

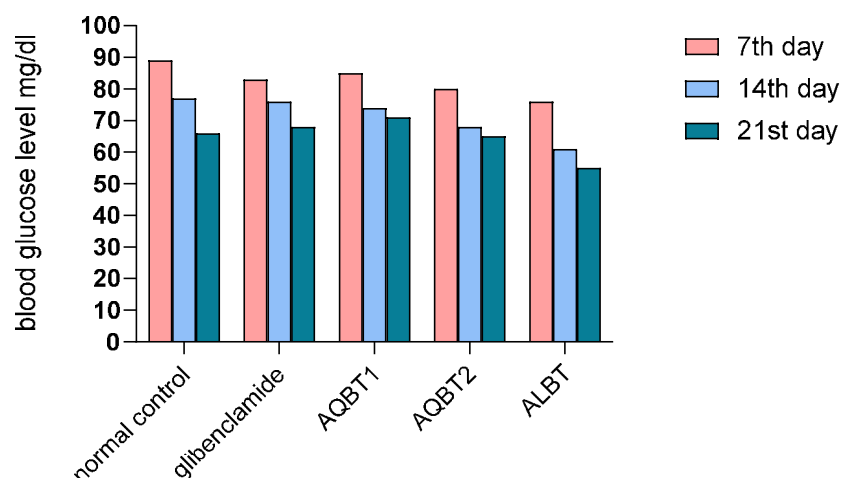


Fig 2: Effect of extracts of *Bauhinia thoningii* on fasting blood glucose level (FBGL) in normal rats.

➤ Oral glucose tolerance test (OGTT) -

Both the aqueous and alcoholic extracts of *Bauhinia thoningii* significantly ($P<0.005$) suppress the rise in FBGL after glucose load (2g/kg) in rats, at first half-an-hour and up to 2hr time period as compare with other groups extract Glibenclamide on 8th, 15th and 22nd day. While aqueous extracts produced significant reduction in FBGL. Glibenclamide (10mg/kg) showed ($P<0.005$) significant suppression in FBGL rise at first half-an-hour, 1hr and normalized FBGL within 2hr. The detailed results are summarized in Table No: 11

Table 5: Effect of extracts of *Bauhinia thoningii* on 8th, 15th and 22nd day in normal rats.

Treatment	Dose (mg/kg)	Blood glucose level(mg/dl)		
		8 th day	15 th day	22 nd day
Normal control	-	85.05±3.72	90.36±1.84	92.31±2.93
Glibenclamide	10	80.11±2.79	75.17±2.74	70.21±1.50
AQBT1	20	86.26±1.21	81.02±0.36	76.17±4.75
AQBT2	30	78.67±0.33	73.99±2.54	71.57±2.26
ALBT	20	82.57±1.76	78.25±1.91	70.38±3.76

Values are expressed as mean ± S.E.M. n=5. Significant values were compared with $P<0.005$. Normal control Vs all groups. Paranthesis indicates % reduction in BGL.

ANTI-DIABETIC ACTIVITY IN ALLOXAN INDUCED DIABETIC RATS

Fasting blood glucose levels (FBGL) in normal rats were in range of 90-100 mg/dl. Treatment with

alloxan (120 mg/kg, I.P.) had increased the FBGL to range of 252-266 mg/dl after 72 hours. These values on subsequent days got stabilized by day seven on an average between 255 mg/dl.

Changes in the fasting blood glucose levels in different groups are tabulated in Table No:. This data shown that blood glucose level of normal control animals has maintained throughout the study period.

The diabetic control group has shown significant increase in fasting blood glucose levels during this 21st day study period. Glibenclamide (10mg/kg) treated group has shown ($p<0.05$) significant decrease in fasting blood glucose level during 7th, 14th and 21st day of study period.

➤ **Effect of *Bauhinia thoningii* extracts on antidiabetic activity in alloxan induced diabetic rats**

The animals treated with 100 and 200mg/kg of aqueous and alcoholic of different extracts shown significant decrease ($P<0.05$) in FBGL on 7th, 14th and 21st day of treatment when compare to other groups of animals. The aqueous extracts have reduced more (%) in FBGL when compared to alcoholic extracts except standard group. The detailed results are summarized in Table.

Table 6: Effect of extracts of *Bauhinia thoningii* on fasting blood glucose level (FBGL) in Alloxan induced diabetic rats.

Treatment	Dose (mg/kg)	Blood glucose level (mg/dl)		
		7 th day	14 th day	21 st day
Normal control	-	81.19±4.69	80.90±7.87	82.74±4.35
Diabetic control	10	295.10±11.05	268.04±10.03	251.13±12.55
Glibenclamide	10	262.21±10.16	245.18±12.01	230.26±11.85
AQBT1	20	373.61±11.21	361.16±19.62	332.57±29.10
AQBT2	30	381.23±12.34	368.82±17.16	351.97±38.33
ALBT	20	292.08±13.09	275.03±14.28	222.60±05.90

Values are expressed as mean ± S.E.M. n=5. Significant values were compared with $P<0.05$. Normal control Vs all groups. Paranthesis indicates % reduction in BGL.

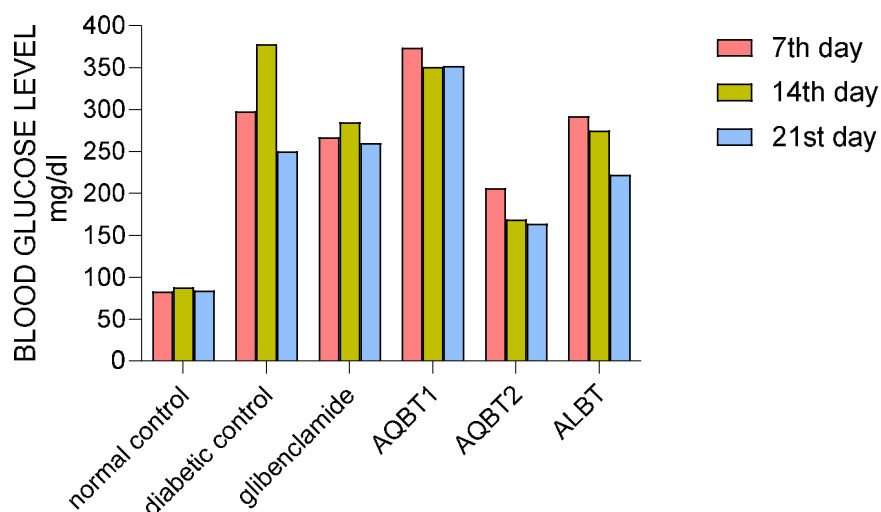


Fig 3: Effect of extracts of *Bauhinia thoningii* on fasting blood glucose level (FBGL) in Alloxan induced diabetic rats.

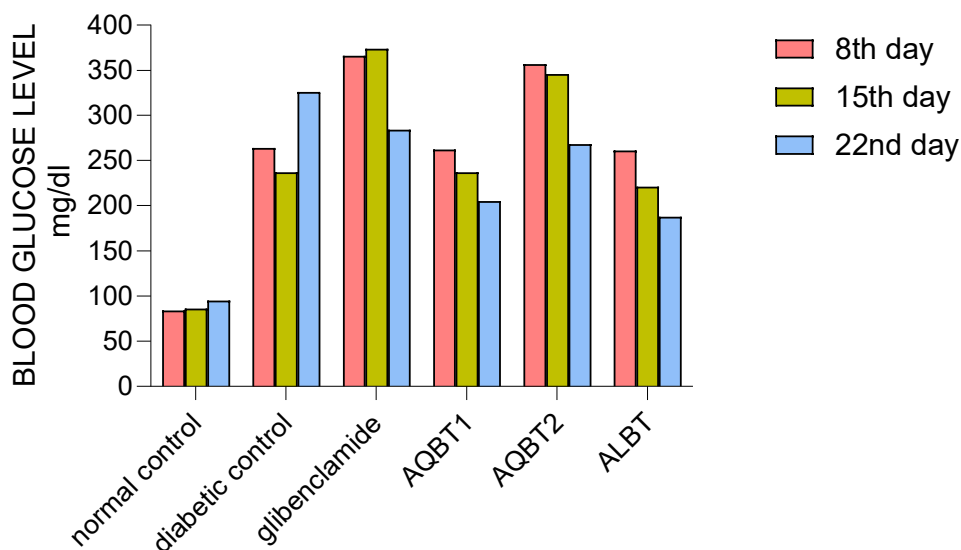
➤ **Oral glucose tolerance test (OGTT) on 8th, 15th and 22nd day-**

Both the aqueous and alcoholic extracts of *Bauhinia thoningii* are significantly ($P<0.05$) suppress the rise in FBGL after glucose load (2g/kg) in rats, at first half-an-hour and up to 2hr time period as compare with other groups extract Glibenclamide on 8th, 15th and 22nd day. While aqueous extracts produced significant reduction in FBGL. Glibenclamide (10mg/kg) showed ($P<0.05$) significant suppression in FBGL rise at first half-an-hour, 1hr and normalized FBGL within 2hr. The detailed results are summarized in Table.

Table 7: Effect of extracts of *Bauhinia thoningii* on 8th, 15th and 22nd day in Diabetic rats.

Treatment	Dose (mg/kg)	Blood glucose level(mg/dl)		
		8 th day	15 th day	22 nd day
Normal control	-	87.75±3.63	84.59±2.87	90.99±1.58
Diabetic control	10	285.01±02.69	275.00±19.55	321.57±14.99
Glibenclamide	10	362.38±21.10	312.88±11.73	289.41±18.55
AQBT1	20	260.26±10.62	241.70±16.40	208.36±11.90
AQBT2	30	287.14±13.30	280.78±13.78	255.98±19.75
ALBT	20	261.22±12.11	221.56±17.87	188.63±17.99

Values are expressed as mean ± S.E.M. n=4. Significant values were compared with P<0.05. Normal control Vs all groups. Paranthesis indicates % reduction in BGL.

**Fig 4: Effect of extracts of *Bauhinia thoningii* on 8th, 15th and 22nd day in Diabetic rats.**

DISCUSSIONS

Despite the fact that diabetes has high prevalence, morbidity and mortality globally, it is regarded as non curable but controllable disease. Different synthetic drugs, plant remedies and dietary modification play an effective role in the reduction of the suffering that it causes. The potential role of medicinal plants as antidiabetic agents has been reviewed by several authors. In order to identify the plants with antidiabetic properties various plants have been tested *in-vivo* using animal models, for example rats, against the complications caused by inducers of diabetes, and it has been established that many plants possess the potential to lower the fasting blood glucose levels and besides help in improving other diabetic complications. The sustained reduction in hyperglycemia automatically decreases the risk of other major complications of diabetes. Effective glucose control is the key for preventing or reversing diabetic complications and improving the quality of life of the diabetics.

Many natural active compounds have been isolated from plants of different species. These active principles are complex Alkaloids, Flavonoids, Steroids, Tannins, Anthraquinones, Terpenoids, Cardiac glycoside and others. These compounds have been shown to produce potent hypoglycemic, anti-hyperglycemic and glucose suppressive activities. These effects might be achieved by facilitating insulin release from pancreatic β -cells, inhibiting glucose absorption in gut, stimulating glycogenesis in liver and/ or increasing glucose utilization by the body. These compounds may also exhibit Anti-Inflammatory, Antibacterial, Antifungal and Cardio protective activities, and restore enzymatic functions, repair and regeneration of pancreatic islets and the alleviation of liver and renal damage.

Aqueous and alcoholic extracts of leaves of *Bauhinia thoningii* at a dose of 20mg/kg and 30mg/kg showed significant effect on the glucose tolerance of rats and it also showed reduction in the fasting blood glucose levels of the normoglycemic rats, thus revealing the hypoglycemic nature of the extracts. The effect was more pronounced for both extracts. These findings indicate that the extracts might be producing hypoglycemic effect by

a mechanism independent from the insulin secretion e.g. by the inhibition of endogenous glucose production or by the inhibition of intestinal glucose absorption.

Alloxan monohydrate is one of the chemical agents used to induce diabetes mellitus in animals. It induces diabetes by dose dependent destruction of β -cells of islets of langerhans. It is a generator of free radicals of oxygen which cause extensive DNA damage. It was observed that single intravenous dose of alloxan exhibited significant hyperglycemia. Excessive hepatic glycogenolysis and gluconeogenesis associated with decreased utilization of glucose by tissues is the fundamental mechanism underlying hyperglycemia in the diabetic state. As the hyperglycemia induced by alloxan falls under category of mild diabetes and may reverse after a few weeks, the hypoglycemic effect of the plant in hyperglycemic rats was studied during 22 days treatment. The difference observed between the initial and final fasting serum glucose levels of extract treated hyperglycemic rat's revealed antihyperglycemic effect of leaves of *Bauhinia thoningii* throughout the period of study. The effect of the extracts was compared to that of reference standard, Glibenclamide and was found to be significant.

Phytochemical analysis of extracts of leaves of *Bauhinia thoningii* revealed the presence of secondary metabolites that have been shown to possess antidiabetic effect in other plants. Flavonoids, alkaloids and Steroids which were responsible for the antidiabetic effect in other plants were also detected in the extracts of this plant. The presence of phenols in the plant could also be responsible for the antidiabetic effect have been shown to prevent the destruction of β -cells by inhibiting the peroxidation chain reaction and thus they may provide protection against the development of diabetes. Extracts of leaves of *Bauhinia thoningii* appear to be attractive materials for further studies leading to possible drug development for diabetes. Development of phytomedicines is relatively inexpensive and less time consuming; it is more suited to our economic conditions than allopathic drug development which is more expensive and spread over several years.

SUMMARY

According to WHO, the word diabetes mellitus is described as a metabolic disorder of several etiology characterized by chronic hyperglycemia with turbulence of carbohydrate, fat and protein metabolism resulting from deficiency in insulin secretion, insulin action, or both. Worldwide the numbers of cases of diabetes is rising gradually. There are numerous medicines available in the market to treat diabetes mellitus, but no drug is found to be fully efficient and secure. Plants and plant-derived products have proven to be effectual and safe in the cure of various kinds of diabetes mellitus. Medicinal plants consist of a number of active components; therefore, they are used for the treatment of large number of infectious ailments. Today the traditional medicinal plants are playing significant role in the development of new drugs. The present study was carried out to establish the antidiabetic activity of aqueous extract of leaves of *Bauhinia thoningii* leaves in streptozotocin induced diabetic rats. The preliminary phytochemical screening of *Bauhinia thoningii* leaves, were carried out using various water and alcohol.

These extracts were subjected to various qualitative chemical analysis, which shown the presence of carbohydrates, alkaloids, flavonoids, glycosides, saponins, steroids, tannins and terpenoids.

1. The dried leaves of *Bauhinia thoningii* for this project work were procured locally.
2. The dried leaves of *Bauhinia thoningii* were successively extracted with water and alcohol.
3. Therapeutic dose of the extracts were calculated after carrying acute oral toxicity studies in rats.
4. Extracts were tested for their anti-diabetic activity in normal and alloxan induced diabetic rats.
5. The following parameters were assessed:
 - Fasting blood glucose levels
On the 7th, 14th and 21st day in normal and alloxan induced rats.
 - Oral Glucose Tolerance Test
On the 8th, 15th and 22nd days in normal and alloxan induced rats.
6. Aqueous (20 mg/kg and 30mg/kg) and Alcoholic extracts (20mg/kg) of *Bauhinia thoningii* showed significant effect in blood glucose lowering activity and improved oral glucose tolerance test (OGTT) in short term (7th day) and long term (14th and 21st day) repeated administration in normal and alloxan induced diabetic rats.
7. The above studies showed that Aqueous extracts of *Bauhinia thoningii* had potent anti-diabetic activity on repeated administration.

CONCLUSION

The study was performed to find out the beneficial effects of two different extracts of leaves of *Bauhinia thoningii* in normoglycaemic rats and alloxan induced diabetic rats and the results reveal that the plant has beneficial effects on blood glucose levels.

In current scenario, herbs are the potent sources of medicines used in the treatment of various disease and disorders. Since, plants are used as medicine there is prompt need of evaluation of plant species, therefore, the

present work was conceived to evaluate the phytochemical and pharmacological screening of leaves of *Bauhinia thoningii*. The Phytochemical evaluation has revealed the presence of Alkaloids, Flavonoids, Steroids, Tannins, Anthraquinones, Terpenoids, Cardiac glycoside.

The aqueous and alcoholic extracts had hypoglycemic activity because of the presence of flavonoids which are rich in treatment of hypoglycemia with less side effects. Flavonoids might be producing hypoglycemic effect by a mechanism independent from insulin secretion e.g. by the inhibition of endogenous glucose production or by the inhibition of intestinal glucose absorption. The present study *Bauhinia thoningii* of both aqueous and alcoholic extracts was showed significant effect on glucose tolerance and showed reduction in fasting blood glucose levels in normal diabetic rats.

The data of the blood glucose level of rats treated with Alloxan (150mg/kg body weight) produced diabetes within 72 hours. After 72 hours of Alloxan administered blood glucose levels of rats were observed. It was observed that there was a significant lowering of sugar in aqueous extract. The administration of aqueous and alcoholic extracts at a dose of 20 mg/kg and 30mg/kg showed significant anti-hyperglycemic effect at 22nd day which was evident from the 7th day on wards as compared to standard. The aqueous extract of *Bauhinia thoningii* has showed better anti-hyperglycemic effect than the alcoholic extract on the fasting blood sugar levels on diabetic rats are shown in table. The decreasing blood glucose levels are comparable with that of 10 mg/kg of Glibenclamide. The Glibenclamide (10 mg/kg body weight) shows significant effect on compare to the initial and more significant effect on the 22nd Day compare to the initial. The aqueous and alcoholic extracts shows significant ($P < 0.05$), effect.

Results of anti-diabetic activity in normal and alloxan induced rats the extracts established the scientific basis for the utility of these plants in the treatment of diabetes. The extracts have shown significant reduction in blood glucose levels in normal and alloxan induced diabetic rats and produced maximum anti-diabetic activity and are higher than the hypoglycemic activity of Glibenclamide in the diabetic rats. In glucose loaded animals, the drug has reduced the blood glucose to the normal levels. It is possible that the drug may be acting by potentiating the pancreatic secretion or increasing the glucose uptake. In conclusion, these extract showed significant anti-diabetic effect in normal and diabetic rats after administration. Thus the claim made by the traditional Indian systems of medicine regarding the use of these plants in the treatment of diabetes stands confirms.

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