



Research Article

PHARMACOLOGICAL SCREENING OF ANTI DIABETIC ACTIVITY OF AQUEOUS EXTRACT OF LEAVES AND FLOWERS OF *BUTEA MONOSPERMA*

Kondalreddy J. ^{1*}, Nukala Aparna ², U. Preethi ³, D. Sri Sruthi ³

^{1*} Assistant Professor, Vision College of Pharmaceutical Sciences & Research, Boduppal, Hyderabad 500092, Telangana, INDIA.

² Doctor in Pharmacy, C L Baid Mehta College of Pharmacy, Chennai, INDIA.

³ B. Pharmacy, Vision College of Pharmaceutical Sciences & Research, Hyderabad, Telangana, INDIA.

Received on: 22-01-2019; Revised and Accepted on: 13-03-2019

ABSTRACT

Butea Monosperma is an Indian traditional plant used in treating diabetes. *Butea Monosperma* possesses strong antioxidant, α -glucosidase inhibitory activity. And the aqueous extract of the flower also has the chemical constituents which are responsible for having free radical scavenging nature. The Phytochemical screening of the aqueous extract of flower of *Butea Monosperma* showed the positive results for Flavonoids, Glycosides, Carbohydrates and Amino Acids. From the Phytochemical study we found that the extract has the antioxidant properties. By this result we extended our work by administering in diabetes induced SD Rats to check whether it has any antidiabetic activity or not. So, after treating the diabetic rats for 14 days with our extract we found that the blood glucose levels got decreased when we compared it with the first day of glucose levels. Hence, we confirmed that the aqueous extract of the flower of *Butea Monosperma* has the antidiabetic activity.

KEYWORDS: Diabetic, *Butea Monosperma*, Pharmacological Activity, Alloxan, Aqueous Extract.

INTRODUCTION

Diabetes mellitus is reaching as epidemic proportion across the globe, a metabolic disorder characterized by hyperglycemia resulting from defect in insulin secretion, insulin action or both (ADA, 2009). Currently, there are 40 million people with diabetes in India estimated to rise to almost 70 million by 2025 (IDF, 2006). It is estimated that 25% of the world population is affected by this disease. Hyperglycemia is the most important factor in the onset and progress of diabetic complications mainly by producing oxidative stress (Giugliano *et al.*, 1996). Altered cellular metabolism caused by hyperglycemia has been suggested to play an important role in increasing the risk of cardiovascular, renal, ophthalmic and neurological complications of diabetes mellitus (Brownlee and Cerami, 1981). Diabetes is a disease condition in which blood glucose level concentrations are elevated i.e. hyperglycemia due to loss of insulin secretion by pancreatic β -cells called type-I diabetes or loss of insulin responsiveness in targeted tissues referred as type-II diabetes or both (Schwarz, P.E *et al.*, 2009). It is one of the most common

metabolic syndromes, since there are 200 million diabetic individuals in the world it is needed to understand the etiology and factors influencing onset of the disease (WHO, 1985). Pathogenic factors that are involved in the development of disease are auto-immune destruction of the β -cells of pancreas with consequent insulin deficiency to abnormalities results in insulin resistance. Deficient action of insulin on target tissues and hyperglycemia are the basis of the abnormalities in fat, carbohydrate, and protein metabolism, causing diabetic characteristic clinical features, increased risk of cardiovascular disease, micro and macro vascular complications (WHO, 1985). Diabetes mellitus is a metabolic disorder characterized by hyperglycemia, Polyuria, glycosuria. Reasons for this rise include sedentary lifestyle, obesity, consumption of energy rich diet, higher life span. It is important to remember that diabetes can occur without any symptoms. Diabetes is a complex disease and not everyone has the same symptoms. In general, excess sugar in blood is removed by the kidneys, using up large amounts of water in the process: sooner or later the most common symptoms is that you may pass more urine and drink a lot more in order to compensate for the loss of liquid.

Insulin is the principal signal that controls the conversion of glucose to glycogen for internal storage in liver and muscle cells. Reduction in the insulin release from the beta cells and reverse conversion of glycogen to glucose when fall in glucose level results in lowering glucose levels that is controlled by the glucagon hormone which counteracts the action of insulin (Rama Rao Naidu B.V.A, 2005). Glucose that is produced from the internal liver cells stores as glycogen which re-enters the blood stream; when muscle cells lack the

***Corresponding author:**

Kondalreddy J.

Assistant Professor,
Vision College of Pharmaceutical Sciences & Research,
Boduppal, Hyderabad 500092, Telangana, INDIA.

* E-Mail: kondal.maddy@gmail.com

DOI:

necessary export mechanism. Normally when the insulin levels are low liver cells do this which normally correlates with low levels of blood glucose (Yoganarasimhan, S.N, 2005).

Higher insulin levels increase in anabolic process, such as protein synthesis, fat storage, growth and duplication. Presence or lack of insulin is the principal signal in converting many of the bidirectional process of metabolism i.e from catabolism to anabolism and *vice versa*. Particularly low insulin level is the trigger for entering or leaving ketosis which is the fat burning metabolic phase. If there is shortage of insulin availability, cells respond poorly to the effects of insulin called insulin sensitivity or resistance. Glucose will not have its usual effect when there is defect in insulin, so there will be no proper absorption by the body cells that require it, nor stored appropriately in the liver and muscles. The net effect is persistent high glucose levels in blood, other metabolic derangements, such as acidosis and poor protein synthesis.

MATERIALS & METHODS

Experimental Animals:

Healthy Wister rats, weighing between 150-200gm of either sex are used for the study. Animals procured from the animal house of Sainath Agencies and were housed individually in clean polypropylene cages in air-conditioned room where the temperature was $22\pm 2^{\circ}\text{C}$ with $50\%\pm 10\%$ relative moisture with a 12 h light and dark cycle all over the study, animals were maintained at normal laboratory circumstances and were given commercial laboratory animal feed and water with libitum. Prior to conducting experiments, ethical permission was taken from Institute's Animal Ethics Committee (IAEC) of CPCSEA Reg.no.930/PO/a/2018/CPCSEA.

Chemicals:

Alloxan (Sigma Pvt. Ltd. India). Glibenclamide (10mg/kg) reagent kits for alkaline SGOT, SGPT, TG's, (EX-cel life science Pvt.Ltd), Sodium citrate, HCl, CMC and all other

chemicals worn in the study are of analytical grades procured from respective manufacturers.

Plant material collection and authentication:

The fresh whole plants of *Butea Monosperma* were obtained from Boduppal, Hyderabad, Telangana in the month of February and they were authenticated by Prof. Dr. H Ramakrishna, Botanist, Osmania University.

Extraction procedure:

The whole plants were shade dried, powdered, and sieved (mesh no. 40) to get coarse powder. Then this powdered coarse plant material was packed into soxhlet apparatus subjected to soxhlation using aqueous as solvent for 72 hours at a temperature of 40°C . After filtration, it was evaporated using water bath at a temperature not exceeding 40°C until the crude extract was obtained as a semi solid and air dried to get solid mass.

Selection of dose:

Plant extract was prepared according to ED₅₀ of individual herbs. ED₅₀ of individual plants was found to be as, *Butea Monosperma* (200 mg/kg). Plant extract at a dose of 200mg/kg and 400mg/kg were used for the present study according to ED₅₀ values from previous studies.

Experimental Design:

The healthy female animals were randomly selected into five groups, each group containing 6 animals were treated for 28 days, as given in following treatment schedule Aqueous extract of *Butea Monosperma* was freshly suspended in distilled water and administered to animals by oral feeding needle. The standard drug Glibenclamide (5mg/kg, p.o) was prepared by suspending in a 1% CMC using mortar and pestle, made as a suspension and administered immediately.

Table No. 1: Treatment schedule for assessing anti antidiabetic activity of Aqueous Extract of Butea Monosperma

S.No	GROUPS	TREATMENT	PURPOSE
I	Normal	Normal saline	Serves as normal
II	Negative control	Alloxan(40mg/kg i.p)	Serves as disease control
III	Standard	Alloxan + Glibenclamide(5mg/kg p.o)	To study the effect of Glibenclamide in disease condition
IV	Test-I	Alloxan + Low dose of Plant extract	To study the effect of <i>Butea Monosperma</i> at low dose in disease condition
V	Test-II	Alloxan + High dose of Plant extract	To study the effect of <i>Butea Monosperma</i> at high dose in disease condition

Acute toxicity studies:

Acute toxicity studies were performed to test the lethal or toxic dose of the test drug on animals to avoid any complications while administering the plant extract. The acute oral toxicity study was carried out according to the OECD 423 guidelines protocol. In this study female rats (n=3) weighing about 180-220gm were used in the testing dose of 2000mg/kg body weight. Then they were observed for signs of toxicity and mortality for 14 days from the initial day of dosing.

It was observed that there were no signs of toxicity and mortality for 14 days at a testing dose of 2000mg/kg body weight. Therefore one-tenth of the maximum no mortality dose

of extract were selected low dose 200mg/kg and high dose 400mg/kg.

RESULTS & DISCUSSION

In-Vivo studies:

The in-vivo testing was performed on albino Wistar rats of either sex (150-220gm) obtained from Sai nath agencies animal house which were housed under standard laboratory conditions and fed with commercial rat feed (Lipton India Ltd., Mumbai, India) and boiled water, ad libitum. All the animal experiments were carried out according to NIH guidelines, after getting the approval from the Institute's Animal Ethics Committee (IAEC) of CPCSEA Reg.no.930/PO/a/2018/CPCSEA.

Acute toxicity studies:

Acute toxicity studies were performed to test the lethal or toxic dose of the test drug on animals to avoid any complications while administering the plant extract. The acute oral toxicity study was carried out according to the OECD 423 guidelines protocol. In this study female rats (n=3) weighing about 180-220gm were used in the testing dose of 2000mg/kg

body weight. Then they were observed for signs of toxicity and mortality for 14 days from the initial day of dosing.

It was observed that there were no signs of toxicity and mortality for 14 days at a testing dose of 2000mg/kg body weight. Therefore one-tenth of the maximum no mortality dose of extract were selected low dose 200mg/kg and high dose 400mg/kg.

Table No. 2: Results of Acute toxicity studies

S.no	Initial weight	Final weight	Signs of toxicity			Mortality
			Behavioral	Somatic	Neurological	
1.	185	210gm	No signs	No signs	No signs	Nil
2.	190	204gm	No signs	No signs	No signs	Nil
3.	199	214gm	No signs	No signs	No signs	Nil

Oral Glucose Tolerance Test (OGTT):

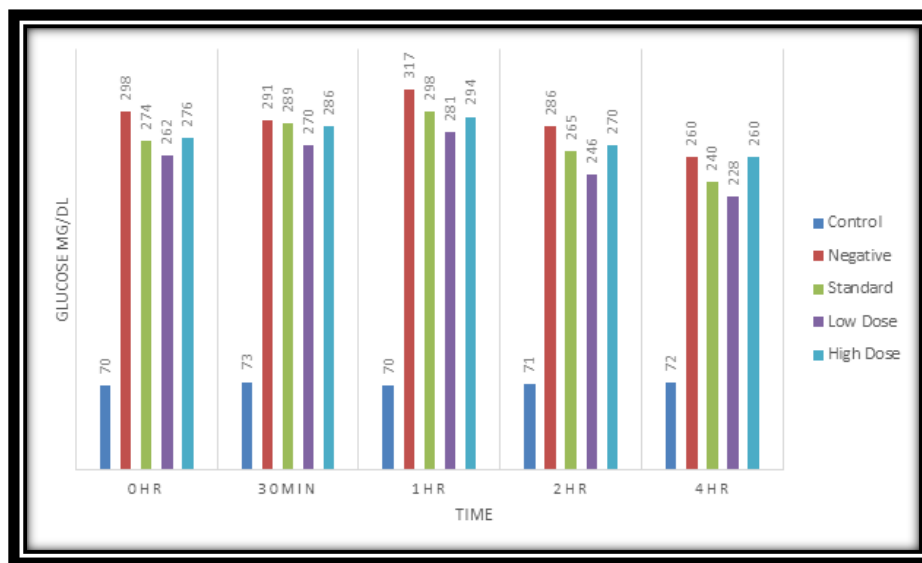
OGTT was carried out to confirm the induction of type 2 diabetes. Rats, after 12 hr of fasting were given orally, a glucose challenge of 2g/Kg body weight. Blood glucose was

determined by the above mentioned method at 0, 30, 60, 120 and 180 min after glucose challenge. A plot of Blood glucose level versus time obtained was analyzed for impairment in glucose tolerance, to confirm the induction/extent of diabetes.

Table No. 3: Results of Blood Glucose Levels 7th Day

CONDITION	0hr	30min	1hr	2hr	4hr
Control	70±2.1	73±2.22	70±1.9	71±1.1	72±2.29
Negative control	298±2.5	291±3.1	317±2.6	286±1.5	260±2.53
Standard	274±1.3	289±2.3	298±1.6	265±2.1	240±2.51
Low dose	262±1.2	270±2.9	281±2.29	246±2.9***	228±1.78**
High dose	276±1.9	286±2.4	294±3.4	270±3.03	260±2.23

* Values are significant as the P values are <0.0001, were determined by using one-way ANOVA test with DUNNET'S multiple comparison test.

**Fig. 1: Results of Blood Glucose Levels 7th Day****Table No. 4: Results of Blood Glucose Levels 14th Day**

CONDITION	0hr	30min	1hr	2hr	4hr
Control	71±1.87	72±4.1	70±1.2	71±1.11	72±1.01
Negative control	270±2.76	280±3.22	292±2.1	282±2.59	269±2.09
Standard	268±1.9	270±3.67	286±1.89	241±1.84***	212±1.47**
Low dose	246±3.2	260±1.98	270±3.1	232±1.91	226±1.83
High dose	226±3.2	240±1.98	260±3.1	252±1.91	236±1.83

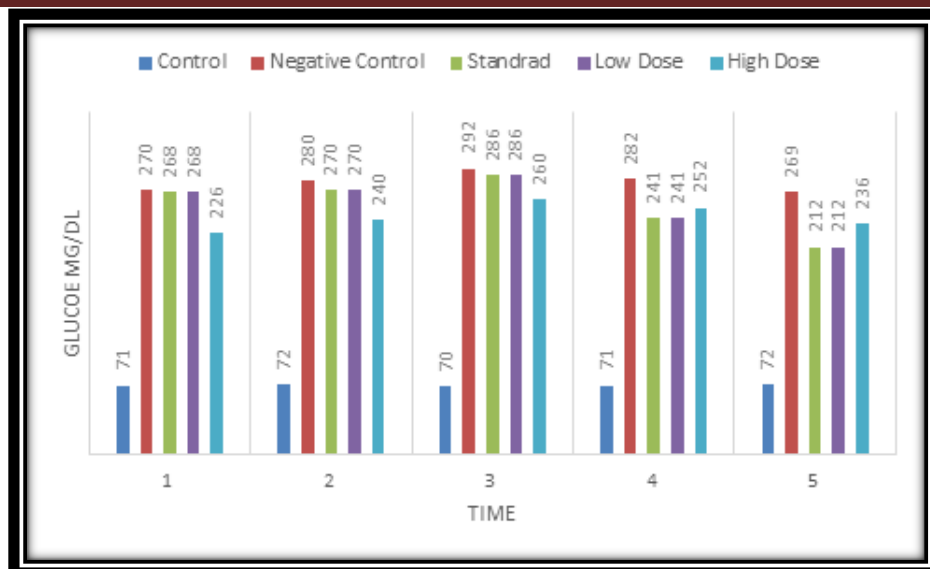


Fig. 2: Results of Blood Glucose Levels 14th Day

Values are expressed in mean $\pm$ SEM, (n=6), when compared with control, * $P < 0.01$, ** $P < 0.001$, one-way ANOVA followed by Dunnett's t - Test. ALLOXAN (40mg/kg) was injected to all groups except control. Alloxan induced diabetic group vs normal group, *Extract treated group vs ALLOXAN induced diabetic group.

DISCUSSION

Diabetes mellitus is a metabolic disorder characterized by hyperglycemia and alterations in carbohydrate, fat and protein metabolism, associated with the continuous destruction of β -cell leads to the disturbance in glucose homeostasis. Liver and pancreas are the two main organs which plays important role in maintain the level by regulating alternative pathways between glucose uptake and gluconeogenesis (Ferre *et al.*, 1996). Diabetes is induced by Alloxan (ALLOXAN) in animal model.

Currently available drugs for the treatment of Diabetes mellitus have a number of limitations, such as adverse effects and a high rate of secondary failure. Although there is a growing trend towards using natural remedies adjunct to conventional therapy, traditionally used plants might provide a useful source of new hypoglycemic compounds. Although *Butea Monosperma* may be described as a medicinal plant used for various purposes, no scientific reports exist on its antihyperglycaemic effect.

ALLOXAN is known to induce diabetes in animal models along with hyperlipidemia/atherosclerosis, diabetic nephropathy, retinopathy, and neuropathy. ALLOXAN mediated toxicity is might be due to their proposed site of action at nuclear DNA. The decomposition of ALLOXAN leads to formation of highly reactive carbonium ions, which cause DNA bases alkylation and also damage pancreatic-cell membrane and break the DNA strand which guide to the activation of poly (ADPribose) synthetase and NAD depletion, which ultimately leads to cell death (Portha *et al.*, 1989). It has been previously reported that ALLOXAN selectively destroy the pancreatic cells that secrete insulin and results in substantial hyperglycemia as well as also result in weight loss, ketosis and a high rate of mortality (Arulmozhi *et al.*, 2004, Szkudelski, 2001).

The present work was designed to investigate the antihyperglycaemic effects of Aqueous extract of *Butea Monosperma* (AEBM) extract in Alloxan diabetic albino rats. *Butea monosperma* is having the antioxidant properties it might be responsible for the antidiabetic activity.

CONCLUSION

Butea monosperma possess various pharmacological activities such as Diabetes Mellitus, Tonic, Jaundice, Diuretic, Scabies, Gonorrhea, and Antioxidant.

In the present study, aqueous extract of *Butea monosperma* showed a promising anti diabetic effect in Alloxan and high fat diet induced diabetes in rats. Hence oral administration of Aqueous extract of *Butea monosperma* has the potential of antidiabetic activity. Further the active principles of *Butea monosperma* responsible for this property is too isolated Phytochemically and studies with these purified constitutions are waiting to understand the complete mechanism of antidiabetic activity.

REFERENCES:

1. American Diabetes Association (ADA). Diagnosis and classification of diabetes mellitus. *Diabetes Care* **2009**; 32:62-7.
2. Awai M, Narasaki M, Yamonoi Y, Seno S. Induction of diabetes in animals by parenteral administration of ferric nitrilotriacetate: A model of experimental hemochromatosis. *Am J Pathol* **1979**; (95)3:663-673.
3. Baeyens L, De BS, Lardon J, Mfopou JK, Rooman I, Bouwens L. In vitro generation of insulin-producing beta cells from adult exocrine pancreatic cells. *Diabetologia* **2005**; 48:49-57.
4. Bailey CJ, Day C. Traditional plant medicines as treatments for diabetes. *Diabetes Care* **1989**; 12:553-564.

5. Beretta A. Campanha de prevencao e diagnostic do diabetes realizada pela UNIARARAS e prefeitura municipal na cidade de Araras. Laes and Haes **2001**;22 (131):188-200.
6. Bonnevie-Nielsen V, Steffes MW, Lernmark A. A major loss in islet mass and B-cell function precedes hyperglycemia in mice given multiple low doses of streptozotocin. *Diabetes* **1981**;30:424-429.
7. Brioukhanov AL, Netrusov AI. Catalase and superoxide dismutase: distribution, properties, and physiological role in cells of strict anaerobes. *Biochem* **2004**;69:949-962.
8. Brownlee M, Cerami A. The biochemistry of the complications of diabetes mellitus. *Annu Rev Biochem* **1981**;50:385-432.
9. Card JW and Magnuson BA. A review of the efficacy and safety of Nanoparticle-Based Oral Insulin delivery system. *Am Phys Soc* **2011**.
10. Chen H, Zheng C, Zhang X, Li J, Li J, Zheng L et al. Apelin alleviates diabetes-associated endoplasmic reticulum stress in the pancreas of Akita mice. *Peptides* **2011**;32: 1634-1639.
11. Choi SB, Park CH, Choi MK, Jun DW, Park S. Improvement of insulin resistance and insulin secretion by water extracts of *Cordiceps militaris*, *phellinus linteus* and *paecilomyces tenuipes* in 90% pancreatectomized rats. *J Biotech Biochem* **2004**;68: 2257-2264.
12. Colagiuri RN, Colagiuri S, Yach D and Pramming S. *Am J Public Health* **2006**;96:1562-1569.
13. Deeds MC, Anderson JM, Armstrong AS, Gastineau DA, Hiddinga HJ, Jahangir A, et al. Single dose streptozotocin-induced diabetes: considerations for study design in islet transplantation models. *Lab Anim* **2011**;45:131-140.
14. Dekel Y, Glucksam Y, Elron-Gross I, Margalit R. Insights into modeling streptozotocin-induced diabetes in ICR mice. *Lab Anim (NY)* **2009**;38:55-60.
15. Drel VR, Pacher P, Stavniichuk R, Xu W, Zhang J, Kuchmerovska TM et al. Poly (ADP-ribose) polymerase inhibition counteracts renal hypertrophy and multiple manifestations of peripheral neuropathy in diabetic Akita mice. *Int J Mol Med* **2011**;28:629-635.
16. Eman GEH, MHA. Hasan, AM. Mustafa and A. Al-Kamel. Effect of Aloe vera extract on some physiological parameters in diabetic albino rats. *Egy J Hosp Med* **2003**;12:53-61.
17. Ferre T, Pujol A, Riu E, Bosch F, Valera A. Correction of diabetic alterations by glucokinase. *Proc Natl Acad Sci* **1996**;93:7225-30.
18. Friedwald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low density lipoprotein cholesterol in plasma, without use of preparative ultra-centrifuge; **1972**.
19. Flodstrom M, Tyrberg B, Eizirik DL, Sandler S. Reduced sensitivity of inducible nitric oxide synthase-deficient mice to multiple low-dose streptozotocin-induced diabetes. *Diabetes* **1999**;48:706-713.
20. Giugliano D, Ceriello A, Paolisso G. Oxidative stress and diabetic vascular complications. *Diabetes Care* **1996**;19: 257-67.

How to cite this article:

Kondalreddy J, et al. PHARMACOLOGICAL SCREENING OF ANTI DIABETIC ACTIVITY OF AQUEOUS EXTRACT OF LEAVES AND FLOWERS OF *BUTEA MONOSPERMA*. *J Pharmacol* 2019;4(1):48-52.

Conflict of interest: The authors have declared that no conflict of interest exists.

Source of support: Nil