STUDIES ON ANTI-DIABETIC ACTIVITY OF CLEOME VISCOSA IN ALLOXAN-INDUCED DIABETIC RATS

Article Review by Boyina Chaya Devi[*1]*, Chakrapani Ramesh*[2]*, India*
[1]*MSc, PhD in Clinical Research, Texila American University*
[2]*Adjunct Professor of Pharmacology and Clinical Research, Texila American University*
Email: - chaya_devi06@rediffmail.com

ABSTRACT

Cleome viscose (L.) belonging to family (Capperdiceae), commonly called "Sticky spider flower", is a terrestrial, annual, erect, aromatic herb. The plant has high medicinal value as it is traditionally used for its antiemetic, wound healing, antimicrobial, hepatoprotective and anti-oxidant properties. The aim of the proposed work is to evaluate the therapeutical potential of *Cleome viscosa* in alleviating diabetes by assessment of liver and kidney function and lipid profile parameters in alloxan-induced diabetic rats. A significant elevation ($P<0.001$) of blood glucose, SGPT, SGOT, ALP, urea, uric acid, creatinine and lipid profile in control groups was observed as compared to normal groups. However, there was significant reduction ($P<0.001$ and $P<0.001$) in the tested biochemical parameters in both the groups treated with extract as compared to the control group and the effect was compared with the standard drug, Metformin. From these results, it can be concluded that the methanol extract of *Cleome viscosa* possesses significant ability to reduce the diabetes complications

KEY WORDS

Cleome viscosa, diabetes, complications, neuropathic pain, lipid profile, liver and kidney function.

INTRODUCTION

Diabetes is a metabolic disorder resulted from the destruction of insulin secreting pancreatic β-cells, defect in insulin production, insulin action, or both, characterized by hyperglycemia. The chronic hyperglycemia results in long-term complications of diabetes include peripheral neuropathy causing foot ulcers, autonomic-neuropathy, causing stroke, ischemic heart diseases and peripheral vascular disease, sexual functions; nephropathy causing renalfailure; retinopathy with loss of vision. Quality of life is diminished. It reduces the life expectancy[1]. The metabolic

complications of diabetes are diabetic ketoacidosis, hyperosmolar non-ketotic coma, Lacticacidosis, and hypoglycemia. Ketoacidosis and hyperosmolar non-ketotic coma are due to insulin deficiency. Hypoglycemia results from the treatment, either with oral agents or insulin. Hypoglycemia common in patients treated with insulin[2]. Prolonged exposure of tissues to hyperglycemia results in various complications including premature atherosclerosis, retinopathy, nephropathy and gangrene of the limbs. It is thought to be due to reduced blood supply to these structures because of thickening of the capillary walls. Accumulation of glycosylated products in the vessel walls may be responsible for the thickening. More amount of intercellular over conversion of intracellular glucose is converted to sorbitol by the enzyme aldose reductase. This sorbitol exerts osmotic effect resulting in tissue damage particularly in the retina and peripheral nerves. Hence, it is necessary to maintain normal blood glucose levels to prevent or delay the onset of complications of diabetes[3].Cleome viscosa is a terrestrial, annual, erect, aromatic (fetid smell) herb reaches up to 120cm tall. Taproots are white or brown in colour. Stem is erect, rounded, solid, (glandular) hairy. Stiputes are absent. Leaves are compound, trifoliolate, alternate spiral, stalked, leaflets elliptic, (glandular) hairy on both side, margin entire, apex acute, base acute, pinnately veined. Flowers are bisexual, single, axiallary, stalked, yellow, petals 4 and free. Fruit is a capsule, opening by two valves[4].Gram negative species. Free radical scavenging activity of Cleome viscosa extract was studied in vitro by Lakshmi and Bindu (2013) and was found to be have significant activity

MATERIALS AND METHODS

PLANT MATERIAL

Plant sample of Cleome viscosa were collected in the month March, 2014 from S.V. University, Tirupathi, India; and verified by Prof. Dr. M. Madhava Chetty, Department of Botany, S. V. University, Tirupathi, Andhra Pradesh, India.

PREPARATION OF EXTRACT

The collected fresh plant materials were dried in shade (2 days) and then dried in a hot air oven at 25ºC for three days and they were made in to coarse powder with the use of mixer grinder. The powder of Cleome viscosa obtained were weighed separately and transferred to a round bottomed flask and then went to continuous heat extraction with soxhlet apparatus using 70% methanol for 24 hours. Then the extract of methanol was concentrated. Extract obtained was dried by placing it on a big petri plate on electric water bath (70ºC) and then kept in an oven at 30ºC for 2 hour. The extract obtained was kept for drying and stored in vacuum desiccators. The percentage yield of the extract was 6.29%.

PHYTOCHEMICAL SCREENING

The crude extract was investigated for the presence of various phytoconstituents by using the standard methods.

EXPERIMENTAL ANIMALS

The Wister albino rats of either sex (200-250g) were obtained from the central animal house of Sigma Institute of Clinical Research & Administration Pvt. Ltd., Hyderabad, India. Animals were housed at a temperature of 24±2°C and relative humidity of 30-70% environment with a constant 12 h light and dark cycle was followed. All animals had free access to water and standard pellet laboratory animal diet. Animals were acclimatized to laboratory conditions before the experiment. All experiments and protocols described in present study were approved by the (IAEC) Institutional Animal Ethics Committee (769/2011/CPCSEA) approved the study protocol.

ACUTE ORAL TOXICITY STUDY

Acute toxicity studies were performed according to OECD-423 guidelines[9] Category IV substance (acute toxic class method). Albino mice (n=3) of either sex selected by random sampling technique were employed in this study. The animals were fasted for 4 h with free access to water only. The Sandy and freely draining soils in open wood land scrub and on screes slopes in dry areas. Occasionally, a noxious weed. In Spain, it is a troublesome weed on irrigated crops in arid areas[5]. Plant contains flavonoids, tannins, saponins, and alkaloids and steroids, tannins, fatty acids. Plants contain volatile principle with a smell similar to mustard[6]. The leaves are diaphoretic, rubefacient and vesicant. They are used as an external application to wounds and ulcers. The juice of the leaves has been used to relieve earache. The seeds are anthelmintic, carminative, rubefacient and vesicant. The seed contains 0.1% viscosic acid and 0.04% viscosin.

In the Unani system of medicine, the seeds of the plant are documented as anthelmintic and detergent, and are given to treat fever and diarrhea. The seeds are used for anthelmintic while the leaves are useful for healing wounds. A poultice made from the plant is efficacious as a counterirritant in chronic painful joints. Panduraja et al., studied the wound healing activity of methanol extract of Cleome viscosa on rats[7] and it was found to have significant wound healing activity. Upadhyay et al., 2008[8] carried on the antimicrobial assay on aqueous extract of Cleome viscosa on various microbial species and found that they have significant activity on Gram positive andphytosterols, flavonoids, steroids and alkaloids are present and gums and mucilages, proteins and amino amino acids.

ACUTE ORAL TOXICITY STUDY

The extract of Cleome viscosa did not show any mortality and toxicity even at highest dose of

2000mg/kg body weight employed. The present research study was carried out using two different doses (low and high). The methanol extract of Cleome viscosa such as 200 and 400mg/kg body weight for diabetic complication.

EFFECT OF THE EXTRACT ON BLOOD GLUCOSELEVELS

Effect of methanol extract of *Cleomeviscosa*on blood sugar levels (BSL) in Alloxan-induced diabetic rats was studied[14]. Blood sugar levels of normal group did not alter significantly throughout study. In control group blood glucoselevels increased steadily on 1st day (250.50±1.54) to 14th day (261.33±1.05). On treatment of MCV at 200mg/kg., gradual reduction of BSL was observed from 1st day (181.00±1.32) to 14th day (121.50±1.00), similarly on treatment with MCV at 400mg/kg., the BSL were from 1st day (200.3±2.96) to 14th day (101.30±0.85).

Table 4: Effect of methanol extract of *Cleome viscosa* on blood sugar

Treatment	Glucose (mg/dl)			
	0 day	**1st day**	**7th day**	**14th day**
Normal (0.9% saline)	44.66±1.43	122.83±1.17	176.80±1.64	181.10±1.11
Control (Alloxan 120mg/kg)	99.33±1.33*	250.50±1.54*	254.3±1.28$^{@}$	261.33±1.05$^{@}$
Standard (Metformin 14.2mg/kg)	37.27±1.04*	245.83±0.95*	214.50±1.4$^{\&}$	145.85±0.79$^{\&}$
MCV 200mg/kg	69.83±1.25*	181.00±1.32*	180.00±1.08$^{\&}$	121.50±1.00$^{\&}$
MCV 400mg/kg	59.00±1.17*	200.30±2.96*	199.60±0.88$^{\&}$	101.30±0.85$^{\&}$

Data expressed was Mean ± S.E.M. n=6, * = not significant,

ANOVA followed by Dunnett's multiple comparison test $^{\&}P<0.001$ as compared control group; and $^{@}P<0.001$ as compared normal group.

EFFECT OF THE EXTRACT ON BIOCHEMICAL PARAMETERS[15-21]

The effect of methanol extract of *Cleomeviscosa* on SGOT, SGPT, ALP, Urea, Uric acid, and Creatinine in Alloxan-induced diabetic rats was studied and the results were given in Table 5. There was a significant increase in these enzyme levels in control group of animals (P<0.001)

compared to the normal group of animals. There was significant reduction (P<0.05 and P<0.05) on treatment of MCV at the dose of 200mg/kg and 400mg/kg. There was a significant reduction in the standard group of animals (P<0.001) compared to the control group of animals. However, there was significant reduction (P<0.001) and (P<0.001) in both treated groups with MCV at 200mg and 400mg as compared to the control group of animals.

EFFECT OF THE EXTRACT ON LIPID PROFILE

The effect of methanol extract of Cleome viscosa on cholesterol, triglycerides, and HDL in Alloxan-induced diabetic rats were studied using standard models and the results were illustrated in Table 6. A significant elevation (P<0.001) was observed in the control group compared to normal group. There was significant reduction (P<0.001) in the standard group compared to control group.at the dose of 200mg/kg and 400mg/kg. There was a significant reduction in the standard group of animals (P<0.001) compared to the control group of animals. However, there was significant reduction (P<0.001) and (P<0.001) in both treated groups with MCV at 200mg and 400mg as compared to the control group of animals.

However, there was a significant reduction (P<0.001 and P<0.001) in both treated group at the dose of MCV 200mg, MCV 400mg/b.w., respectively, compared to the control group.

STATISTICAL ANALYSIS

The Statistical analysis software Graphpad Prism Version 5 was used for statistical analysis of the present studies.

Table 5: Effect of methanol extract of *Cleome viscosa* on biochemical parameters on liver

Treatment	SGOT (U/L)	SGPT (U/L)	ALP (U/L)	Urea (mg/dl)	Uric acid (mg/dl)	Creatinine (mg/dl)
Normal(0.9% saline)	24.3±1.03	43.7±0.80	25.0±1.01	68.91±1.18	3.2±0.50	5.2±0.64
Control (Alloxan 120mg/kg)	42.52±0.63$^{@}$	48.2±1.09$^{\wedge}$	42.1±1.06$^{@}$	66.89±1.25$^{@}$	3.38±0.53$^{@}$	5.9±1.46
Standard (Metformin 14.2mg/kg)	36.5±1.02*	34.14±0.77$^{\&}$	32.6±0.69$^{\&}$	56.24±0.63$^{\&}$	3.03±0.73$^{\&}$	5.0±0.69$^{\#}$
MCV 200mg/kg	39.4±1.23$^{\#}$	27.3±1.06$^{\&}$	33.3±0.82$^{\&}$	49.51±3.06*	2.00±0.85$^{\&}$	4.8±0.68$^{\#}$

MCV 400mg/kg	38.6±1.26$^{\#}$	41.5±1.14$^{\&}$	30.8±0.75$^{\&}$	44.83±1.05$^{\&}$	3.23±0.66$^{\&}$	4.5±0.67$^{\#}$

Data expressed was Mean ± S.E.M. n=6,

ANOVA followed by Dunnett's multiple comparison test $^{\#}$P<0.05; *P<0.01; $^{\&}$P<0.001 as compared to control group; and $^{@}$P<0.001, $^{\$}$P<0.01, and $^{\wedge}$P<0.05 as compared normal group.

Table 6: Effect of methanol extract of *Cleome viscosa* on biochemical parameters on liver

Treatment	**Cholesterol (mg/dl)**	**Triglycerides (mg/dl)**	**HDL (mg/dl)**
Normal (0.9% saline)	32.88±0.73	66.79±0.88	75.81±1.03
Control (Alloxan 120mg/kg)	83.83±0.36$^{@}$	92.84±0.74$^{@}$	54.46±0.73$^{@}$
Standard (Metformin 14.2mg/kg)	41.63±0.80$^{\&}$	50.60±0.71$^{\&}$	77.99±0.63$^{\&}$
MCV 200mg/kg	45.6±0.92$^{\&}$	54.84±0.77$^{\&}$	66.10±0.46$^{\&}$
MCV 400mg/kg	53.81±0.80$^{\&}$	54.50±0.54$^{\&}$	65.76±0.91$^{\&}$

Data expressed was Mean ± S.E.M. n=6,

ANOVA followed by Dunnett's multiple comparison test $^{\#}$P<0.01; $^{\&}$P<0.001 as compared to control group; and $^{@}$P<0.001 as compared normal group.

CONCLUSION

The findings of the study reveal that oral administration of methanol extract of Cleome viscosa exhibited significant antidiabetic and hepatoprotective effect and the protective effect was compared with the standard drug (Metformin). Further, oral use of the extract might positively affect the functional capacities of various rat tissues, particularly blood, kidney and nerves against toxic action of Alloxan at the dose of 120mg/kg b.w. These results clearly support the traditional use of Cleome viscosa in the treatment of diabetes mellitus and further on diabetic complications shedding more light in the efficacy of the plant. Thus, Cleome viscosa appears to a valuable plant and ideally suited to be used in treatment of diabetes mellitus and further studies are required to assess the usefulness of the plant in diabetic complications, since this is a non-toxic plant.

REFERENCES

1. Bowlers LD. *et al*. *Clinical Chemistry.* 1980; 26: 655.
2. Expert Committee on the Diagnosis and Classification of Diabetes Mellitus. Report of the Expert Committee on the Diagnosis and Classification of Diabetes Mellitus. Diabetes Care.1997; 20: 1183–119.
3. Fossati P, Principe L, Berti G. Use of 3,5-dichloro-2-hydroxybenzene sulphonic acid-4-aminophenazone chromogenic system in direct assay of uric acid in serum and urine. *ClinicalChemistry.* 1980; 26: 227-231.
4. Galigher AE, Kozloff EN. Essentials o f Practical Microtechnique. 2ed, Lea and Febiger, Philadelphia, 1971; 77.
5. Govindarajan R, Vijayakumar M, Singh M, Rao CV, Shirwaikar A, Rawat AKS, Pushpangadan P. Antiulcer and antimicrobial activity of Anogeissuslatifolia. Journal of Ethanopharmacology. 2006: 106: 57-61.
6. Ghosh T, Maity TK, Das M, Bose A, Dash GK. Hepatoprotective activity of Bacopamonnieri L. against ethanol-induced hepatotoxicity in rats. Pharmacognosy Magazine. 2007: 3(10): 95-100.
7. JeyarajAnovraj, et al. In vitro regeneration of Cleome viscosa – an important medicinal herb.
8. Johnson D, Palumbo P, Chu C. Diabetic ketoacidosis in a community-based population Mayo Clinic Proc. 1980; 55: 85-88.
9. Lakshmi SP, Bindu RN. Proximate composition, mineral elements and anti-nutritional factors in Cleome viscosa L. and Cleome burmanni W. & A *International Journal of Pharmacy and Pharmaceutical Sciences*. 2013; 5(1): 398-402.
10. Marklund A, Marklund G. Involvement of superoxide anion radical in the autoxidation of pyrogallol and a convenient assay for superoxide dismutase. European Journal of Biochemistry. 1974; 47: 469-474.
11. Niramathi KL, Karunanithi M, Brindha P. Phytochemical and *in vitro* screening of aerial parts of *Cleome viscosa* extracts. *International Journal of Pharmacy and Pharmaceutical Sciences*. 2012; 4(2):
12. Padmaja Udaykumar. Medicinal Pharmacology.CBS publishers & Distributors, New Delhi.2013; 574.
13. Panduraju T, Paravthi B, Rammohan M. Wound healing propertied of Cleome viscosa. Hygeia Journal for Drugs and Medicines. 2011; 3(1): 41-45.

14. Panduraju T, Paravthi B, Rammoha M. Wound healing propertied of *Cleome viscosa*. *Hygeia Journal for Drugs and Medicines*. 2011; 3(1): 41-45.

15. Pomory CM. Color development time of the Lowry protein assay. Analytical Biochemistry. 2008; 378:216-7.

16. Rotruck JT, Pope AL, Ganther HL, Swanson AB. Selenium: biochemical role as a component of glutathione peroxidase. Science 1973; 179: 588-590.

17. Sinha AK. Colorimetric assay of catalase. Analytical Biochemistry. 1972; 47: 389-394.

18. Trinder P. Determination of glucose in blood using glucose oxidase with an alternative oxygen acceptor. *Annals of Clinical Biochemistry*. 1969; 6: 24-25.

19. Upadhayay, *et al*. Antimicrobial effects of *Cleome viscose* and *Trigeonellafoenum* see dextracts. *Journal of Cell and Tissue Research.* 2008; 8(2): 1355-1360.

20. World Health Organization 2000. Guidelines on Standard Operating Procedures for Clinical Chemistry. 69-7 Bowers LD. *Clinical Chemistry.* 1980; 26: 551.10.