



Research Article

Antidiabetic Activity And Phytochemical Investigation On The Whole Plant Of *Commelina Benghalensis* Linn. In Male Albino Rat

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ABSTRACT

Study of phytochemical and antidiabetic activity on the whole plant of *Commelina benghalensis* in male albino rat. The methanolic extract of the whole plant of *Commelina benghalensis* (100, 200, 400 mg/kg i.p.) has shown significant antidiabetic activity in alloxan-induced diabetic rat. In addition the extract significantly reduced the elevated level of blood cholesterol ($p < 0.01$) and triglyceride ($p < 0.05$). At the same dose level, the extract significantly improved the alloxan-induced reduction of blood protein ($p < 0.01$) to normal value.

INTRODUCTION

Diabetic mellitus comprises a group of common metabolic disorders that share the phenotype of hyperglycemia. Depending on the etiology of the DM, factors contributing to hyperglycemia may include reduced insulin secretion, decreased glucose utilization, and increased glucose production¹. Diabetes mellitus, a chronic metabolic disorder of insulin deficiency or ineffectiveness, constitutes a global public health burden and predictions estimate that India, China, and United States will have the largest number of diabetic people by the year 2030². Diabetic

mellitus is a widespread disorder, which has long been recognized in the history of medicine, before the advent of insulin and oral hypoglycemic drugs, the major form of treatment involved the use of plants. More than 400 plants are known to have been recommended and recent investigations have affirmed the potential value of some of these treatments³. Diabetic mellitus is a group of endocrine syndromes characterized by hyperglycemia; altered metabolism of lipids, carbohydrates, and proteins, and an increased risk of complications from vascular disease. Most patients can be classified clinically as having

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either type I diabetes mellitus (type I DM formerly known as insulin-dependent diabetes of IDDM) or type II DM formerly known as non-insulin dependent diabetes of NIDDM)⁴. Diabetes is one of the most prevalence chronic diseases in the world. This is a chronic incurable condition due to insulin deficiency that affects 10% of the population. The number of diabetic people is expected to rise from present estimate of 150 million to 230 million in 2025. For a long time, diabetes has been treated with several medicinal plants or their extract based on the folklore medicine⁵. Now a days herbal medicines are highly recommended for the treatment of diabetes inspite of other therapeutic option, which can produce serious side effects and in addition they are not safe during pregnancy. Therefore, the search for the more effective and safer hypoglycemic agents has continued to be an important area of active research. Furthermore, after the recommendation made by WHO on diabetes mellitus, investigation on hypoglycemic agent from medicinal plant has become more important⁶. *Commelina benghalensis* Linn. Known as Kanchara, Commelina Kilimandscharica, Commelina Rufociliata, Commelina orrhigia, Uncata, Commelina Obscura, belongs to the family Commelinaceae⁷. The leaves are ovate, about 2.5-7.5 cm long, leaf margins entire. Stem can be erect or crawling along the ground. Flowers are purple in colour with 3 petals, about 3-4 mm long. Seeds are rectangular, about 1.6 - 3 mm in length and with netted appearance. Plant 60-90 cm. Annual or perennial, creeping herb. Leaves broadly ovate, upto 5 sometimes perennial, creeping herb. Leaves broadly ovate, upto 5 cm long and 4 cm wide⁸. The main chemical constitutes of the whole plant in mainly n-octacosanol, n-triocotanol, stigma sterol, compesterol, hydrocyclic acid. Ayurvedic remedy that has been mentioned in many Indian medicinal literatures for the treatment of fever, anemia,

uterine complaints, menorrhigia, blood and skin diseases, diarrhea, colitis, stomatitis, dysentery and in improvement of semen quality⁹. India has a rich traditional of plant-based knowledge on healthcare. All above ground parts of the grass plant are native medicine. Sedative and anxiolytic effects of different fractions of the *Commelina benghalensis* Linn. The leaves or herb are regarded as wound herbal eye medicine chemical tolerance anticancer hepatoprotective antimicrobial activity Commelina Herb LF Wounds. In traditional system of remedies, warm aqueous extract of its leaves have been used to alleviate the pain, swelling and for cleansing and better wound healing¹⁰. It has cooling, indigestible, galactagogue, astringent, alexiteric, uterine complaints, vata, kapha, biliousness and burning effects. Charka prescribes the bark in the treatment of snake-bite but the bark is not an antidote to snake bite¹⁰. The present study was undertaken investigate antidiabetic activity of methanolic extract on the whole plant of *Commelina benghalensis* in male albino rat.

MATERIALS AND METHODS

Plant Material

The fresh Whole plant of *Commelina benghalensis* Linn was collected during the month of September 2012, from village Chandera, Distt-Tikamgarh (M.P.) The plant materials was identified and authenticated by Dr. Gaurav Nigam, Botany Department, Bundelkhand University, Jhansi. (Herbarium and Museum Division with reference no. (BU/BOT/875/24-10-2012).

Plant Profile



Figure.1 *Commelina benghalensis* linn

| | |
|----------|------------------------|
| Kingdom: | <u>Plantae</u> |
| Clade: | <u>Angiosperms</u> |
| Clade: | <u>Monocots</u> |
| Clade: | <u>Commelinids</u> |
| Order: | <u>Commelinales</u> |
| Family: | <u>Commelinaceae</u> |
| Genus: | <u>Commelina</u> |
| Species: | <i>C. benghalensis</i> |

Drugs and chemicals

The following drugs and chemicals were used Glibenclamide tablet (Nandi Pharma, Vidyanagar, hubli-31) Chemicals: tween-80 (Central drug house, Pvt. Ltd.), Normal Saline (0.9% NaCl solution). The plant extract was suspended in tween-80 and subjected for antidiabetic activity. Glibenclamide tablet were also dissolved in tween-80.

Preparation of Extract The whole plant of *Commelina benghalensis* Linn was air-dried and

then these are made into coarsely powdered form. The powdered drug 150 gm was packed in soxhelt apparatus and continuously extracted with methanol (95%) at 60-70°C till complete extraction. The solvent was removed by distillation and the conc. Extract was dried under reduced pressure at a temperature not exceeding 40°C in rotator evaporator. A green colour extract was obtained. The Extractive value was found to be 21.62 gm. The extract is kept in petridish and stored in a dessicator at room temperature¹¹.

Preliminary Phytochemical Screening

A preliminary phytochemical screening of the extract revealed the presence of different types of chemical constituents were applied. Phytochemical tests on the extract give positive reactions for Stigma sterol, alkaloids, carbohydrate, glycosides, flavonoids, amino acid and phenolic compound¹².

Table 1: Qualitative Phytochemical Analysis

| S. No. | Tests | Methanolic extract |
|--------|--|--------------------|
| 1. | Alkaloids Dragendorff's test Wagner's test Mayer's test Hager's test | + + + + |
| 2. | Carbohydrates Molisch's test Fehling's test | + - |
| 3. | Proteins Biuret test Xanthoprotein test | - + |
| 4. | Amino acid Ninhydrin test | + |
| 5. | Phenolic compounds Lead acetate test Iodine test Dil.HNO ₃ test | - - + |
| 6. | Glycoside Keller Killiani test Borntrager's test | + + |
| 7. | Saponin Foam test | - |
| 8. | Steroids and Sterols Salkowski test | + |
| 9. | Flavonoids Shinoda test | + |



Animals

Male albino rats (160-200 gm) of either sex (bred in D.R.D.O Gwalior, M.P.) The animals were obtained from animal house of the Institute of Pharmacy, Bundelkhand University, Jhansi; India. The animals were housed in standard cages with free access of food (standard laboratory rodent's chow) and water. The animal's house temperature was maintained at $23 \pm 3.0^{\circ}\text{C}$ with a 12 hrs light / dark cycle. All the experimental procedures and protocols used in this study were reviewed by the Institutional Animal Ethics Committee (IAEC) of the Institute with reference no. BU / Pharm / IAEC / 12 / 0213 (approved by CPCSEA Regd No. 716 / 02 / a / CPCSEA).

Acute Toxicity Study of the Extract

Acute toxicity study was performed for the extract according to the acute toxic classic method as per OECD guidelines (**OECD Guidelines for the testing of chemical, 2001**). In house bred male albino rat of either sex weighing between 160-200 gm were used during investigation. The animals were Kept fasted overnight. As per following the OECD guidelines fixed dose method procedure, the effective dose for both extract was found to be 200 mg/kg body weight. The Methanolic extract of whole plant of *Commelina benghalensis* (Linn.F.) cause one mortality up to 200 mg/kg during the observation period of 24 hrs then 72 hrs and thereafter once daily for 21 days and were considered as safe. The oral LD_{50} of the Methanolic extract estimated in rats must be > 5000 mg/kg p.o.

Assessment of antidiabetic activity

Alloxan induced model

Alloxan induced model was used to evaluate the antidiabetic activity of extract. The experimental diabetes in overnight fasted rats was induced by single intraperitoneal administration of 120 mg/kg alloxan monohydrate. After one hour of alloxanisation the animals were given feed ad libitum and 5% dextrose solution for a day to avoid early hypoglycemic phase. The blood glucose was monitored after every 24 hrs of alloxanisation. Rats with blood glucose level more than 200 mg/dl were include in study. The rats were divided into five groups of six animals each (n= 5).

Group I received Normal saline

Group II received Glibenclamide tablet 10 mg/kg body weight.

Group III received 100 mg/kg of CBME suspended in tween-80

Group IV received 200 mg/kg of CBME suspended in tween-80

Group V received 400 mg/kg of CBME suspended in tween-80

The blood samples were collected from retro-orbital plexus on day 1, 15 and 21 of extract administration. The blood levels and triglyceride levels as well as total cholesterol and total proteins were determined.

Statistical analysis

The data subjected to the analysis of variance (one way ANOVA) to determine the significance of changes, Dunnett multiple comparisons were made to analyze the significance of difference within the experimental groups. P values of 0.05 or less were taken as significant.

RESULTS

A preliminary phytochemical screening of the extract revealed the presence of different types of stigma sterol, alkaloids, carbohydrate, glycosides, flavonoids, amino acid and phenolic compound.

DISCUSSION

The result of the present study indicates that Methanolic extract of the whole plant of *Commelina benghalensis* possesses antidiabetic activity in male albino rat. The repeated administration (for 21 days) of Methanolic extract of *Commelina benghalensis* exhibited a marked antihyperglycemic activity in alloxan-induced diabetic rats by lowering the plasma glucose levels. The Methanolic extract of *Commelina benghalensis* at a dose of 400 mg/kg showed significant improvement in oral glucose tolerance in glucose fed hyperglycemic diabetic male albino rats. Such effects may be accounted for, in part, by a decrease in the rate of intestinal glucose absorption, achieved by an extra pancreatic action including the stimulation of peripheral glucose utilization or enchancing glycolytic and glycogenic process with concomitant decrease in glycogenolysis and glyconeogenesis¹³. From the results it is assumed that the observed decreased plasma glucose lowering effect of the extracts could also possibly be due to increased peripheral glucose utilization¹⁴. Further, the previous reports on methanolic extract of the whole plant of *Commelina benghalensis* and its major constituent roseoside shown



probable insulinotropic activity and enhancement of insulin release from β -cells in dose-dependent manner. The data from the present work is in line with the reported work of Frankish *et al*¹⁵. So that it may be predicated similar chemical nature of the extract might be responsible for such activity of the whole plant of *Commelina benghalensis*. It is well known fact that alloxan induces diabetes mellitus in rats by selective necrotic action on the beta cells of pancreas leading to insulin deficiency. Insulin deficiency leads to various metabolic aberrations in animals like increased blood glucose level, increased levels of antihyperglycemic effect of methanolic extract on alloxan induced diabetic rats and alloxan glucose tolerance test¹⁶.

As expected in alloxan treated rats, there was significant increase in blood glucose, alloxan alloxan glucose levels. The diabetic animals showed significant decrease in blood glucose level after 21 days treatment. Moreover it also decreased the levels of alloxan induced diabetic rats and alloxan treatment. Alloxan treatment of the rats has showed the loss in body weight as compared to normal rats. However, the methanolic extract was more effective and results are comparable with that of reference drug, Glibenclamide. From the

collective spectral data, the isolated compound was found to be stigma sterol. Hence, we can say that presence of stigma sterol in the methanolic extract may be responsible for antidiabetic activity¹⁷.

Table I: The characteristics of Methanolic extract of *Commelina benghalensis*

| S. No. | Characteristics | Methanolic extract |
|--------|----------------------|--------------------|
| 1. | Extractive value (%) | 21.62 % |
| 2. | Physical appearance | Semisolid mass |
| 3. | Colour | Green |
| 4. | Odour | Odourless |
| 5. | Taste | bitter |

Table II: Effect of *Commelina benghalensis* (Linn.F.) extract on the determination of acute lethal (LD₅₀) p.o.

| Doses (mg/kg) | No. of Animals | Death |
|---------------|----------------|-------|
| 100 | 03 | 00 |
| 200 | 03 | 01 |
| 400 | 03 | 00 |
| 2000 | 03 | 00 |
| 5000 | 03 | 00 |

*Results are expressed as mean \pm SEM * P < 0.001 when compared to vehicle*

Table III: The Antihyperglycemic effect of Methanolic Extract on Alloxan induced Diabetic rats.

| GP | Dose | Blood Glucose Level (mg/dl) at hr | | | |
|-----|----------------------------|-----------------------------------|-------------------------|-------------------------|-------------------------|
| | | 0 hr | 1 hr | 2 hr | 3 hr |
| I | N.C | 72.14 \pm 3.63 | 74.56 \pm 2.10 | 75.88 \pm 1.20 | 76.15 \pm 1.17 |
| II | D.C | 343.41 \pm 7.95 | 342.31 \pm 5.99 | 340.87 \pm 5.39 | 334.61 \pm 4.48 |
| III | CBL (200mg/kg) | 339.94 \pm 4.47 | 288.96 \pm 2.99*** | 271.40 \pm 3.76*** | 259.01 \pm 5.09*** |
| IV | CBL (400mg/kg) | 346.52 \pm 4.95 | 292.18 \pm 2.78*** | 270.40 \pm 2.42*** | 255.04 \pm 2.50*** |
| V | Glibenclamide (10mg/kg) | 345.31 \pm 4.31 | 286.83 \pm 2.47*** | 252.35 \pm 2.74*** | 237.77 \pm 2.33*** |

All values are mean \pm SEM * P < 0.001, compared to vehicle control

Table IV: The Antihyperglycemic effect of Methanolic Extract on Alloxan Glucose tolerance test

| GP | Dose | Blood Glucose Level (mg/dl) at minutes | | | |
|-----|---------------------|--|------------------------|-------------------------|-------------------------|
| | | 0 minutes | 30 minutes | 60 minutes | 120 minutes |
| I | N.C | 82.14 \pm 3.63 | 84.56 \pm 2.10 | 89.88 \pm 1.20 | 92.15 \pm 1.17 |
| II | Control (4mg/kg) | 75.01 \pm 2.19 | 176.58 \pm 4.36 | 150.90 \pm 3.53 | 125.35 \pm 3.59 |
| III | CBL (200mg/kg) | 70.94 \pm 1.39 | 158.42 \pm 3.73** | 129.79 \pm 2.39*** | 100.74 \pm 1.61*** |



| | | | | | |
|----|----------------------------|----------------|--------------------|--------------------|--------------------|
| IV | CBL (400mg/kg) | 76.30± 3.08 | 152.40± 2.49*** | 129.77± 2.42*** | 101.06± 1.83*** |
| V | Glibenclamide (10mg/kg) | 80.90± 2.55 | 145.93± 1.99*** | 118.86± 2.84*** | 86.00± 3.01*** |

All values are mean ± SEM * P<0.001, compared to vehicle control

CONCLUSION:

It can be concluded from the study that the antidiabetic effects of the Methanolic extract on the whole plant of *Commelina benghalensis* may be via non-specific mechanisms. However, extensive studies are needed to evaluate the precise mechanism (s), active principles and the safety profile of the whole plant as a medicinal remedy for inhibiting hepatic gluconeogenesis. Result from the phytochemical analysis of *Commelina benghalensis* Linn revealed the presence of flavonoids, which has also been isolated from the other plant and found to stimulate secretion or possess an insulin-like effect.

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