

## Anti-Hyperglycemic Activity Of *Cucumis Melo* Leaf Extracts In Streptozotocin Induced Hyperglycemia In Rats

N. Babulreddy<sup>1</sup>, Shakti Prasanna Sahoo\*<sup>1</sup>, S. Ramachandran<sup>1</sup> and M.D.Dhanaraju<sup>1</sup>

\*<sup>1</sup>Department of Pharmacology, GIET School of Pharmacy, Chaitanya Knowledge City, Rajahmundry, 533296, Andhra Pradesh, India  
[sahoo.shakti@gmail.com](mailto:sahoo.shakti@gmail.com)

Subject: Pharmacology

### Abstract

*Cucumis melo* L (Cucurbitaceae), from many centuries have been used, in Indian traditional medicinal system, for the treatment of various disorders of heart such as Cardio protective and antiobesity. The present study was to investigate the possible anti-hyperglycemic activity of *Cucumis melo* leaf extracts in streptozotocin induced hyperglycemia in rats. *Cucumis melo* leaf Methanolic and aqueous extracts were administered to the streptozotocin (55mg/kg) induced hyperglycemia rats for 28 days to study anti-hyperglycemic activity. Serum obtained by immediate centrifugation of blood samples using remi ultra cooling centrifuge at 3000 rpm for 15 minutes at room temperature and was directly used for estimating serum glucose. The acute toxicity value of methanol and aqueous leaf extract after oral administration in mice were found to be 5000 mg/kg. The results concluded that *Cucumis melo* leaf methanolic extract (500 mg/kg) have greater anti-hyperglycemic activity than aqueous extract in streptozotocin induced hyperglycemia model and when compared with Glibenclamide treated group. Hence to conclude that the methanolic extract of *Cucumis melo* leaf having anti- hyperglycemic activity.

**Key words:** Anti-Hyperglycemia, *Cucumis melo*, Streptozotocin

### Introduction

Diabetes mellitus is a serious metabolic disease with several micro and macro vascular complications and diabetic patients have been rapidly increasing in number worldwide.<sup>1</sup> An increase in ageing population, consumption of diets rich in calories and fat, sedentary life style and obesity are among the common risk factors and prevention of disease advancement will be a major concern in the 21<sup>st</sup> century. It is obvious that the prevention of disease complications is possible by the control of blood glucose and improvement of hyperglycemia.<sup>2</sup>

In addition to restriction of taking energy and exercise promotion, the usefulness of herbal medicines and functional foods during the daily life has been shown. For this purpose, several studies on medicinal plants and functional foods and also their active components have been carried out to ascertain their usefulness in controlling the diabetes and their complications.<sup>3, 4</sup> *Cucumis melo* L (Syn. *Cucumis callosu* Rottl) Cogn. *Cucumis trigonus* Roxb. Family Cucurbitaceae) popularly known as Muskmelon<sup>7</sup>. *Cucumis melo* Linn (CM) is a trailing herb or

pubescent with edible, polymorphous fruits. It is used for various ailments in Indian Traditional System of Medicine. Fruit and leaves have medicinal value. The leaves are used in flatulence, fever, jaundice, leprosy, diabetes, antiobesity, cough, anemia, constipation, ascites, bronchitis other abdominal disorders and amentia.<sup>5-7</sup> In addition, fruit pulp is bitter, acrid, liver tonic, anthelmintic, cardio tonic, appetizer, thermogenic, expectorant and intellect promoting. Roots are used as purgative and emetic.<sup>6</sup>

Earlier works on *Cucumis melo* fruits showed effectiveness in preventing chemically induced hypothyroidism in rats.<sup>7</sup> Some other research studies are available on its urease inhibitory, anti-inflammatory, antioxidant, antiulcer and diuretic.<sup>8-10</sup> Phytochemical investigations revealed the presence of phenolic glycoside (E)-4-hydroxycinnmlyl alcohol 4-O-(2'-O-D-apiofuranosyl) (1"→2')-D-glycopyranoside, benzyl O-D-glycopyranoside, 3,29-O- dibenzoylmultiflor - 8-en-3 $\alpha$ ,7,29-triol and 3-O-p-amino-benzoyl-29-O-benzoylmultiflor-8-en-3  $\alpha$ , 7, 29-triol was isolated

and identified from *Cucumis melo* seeds.<sup>11</sup> *Cucumis melo* leaves are used in Indian traditional system of medicine (Ayurvedam, Unani) and also for obesity<sup>6</sup> but there is no much scientific study reported about anti-hyperglycemic activity. Therefore present study was undertaken to establish the acute toxicity study and scientifically evaluate the anti-hyperglycemic activity of the methanol and aqueous extracts of *Cucumis melo* leaf (CML) in streptozotocin induced hyperglycemia model in Albino rats.

## Materials and Methods

### Collection and Authentications of Plant Material

The fresh leaves of *Cucumis melo* L were collected from Chithur district (Andhra Pradesh, India) in the months of December-January. The trailing herb was authenticated by Dr. K. Madhava Chetty, Dept. of Botany, Sri Venkateswara University, Turupati, and A.P. Voucher specimen number 558-4 (dated 09-03-2013). The leaves are crushed and air dried under shadow and then grounded into coarsely powdered in grinder and powder material was passed through 120 meshes to remove fine powders and coarse powder was used for extraction.

### Protocol for Successive Extraction

The coarse powder of *Cucumis melo* leaf (100 gm) was extracted by using successive soxhlet extraction using solvents of varying polarity such as petroleum ether (60-80°C), methanol, distilled water (8.2, 10.1, 4.2, 8.5g respectively) for 72 hrs. After completion of extraction, solvent was distilled off and concentrated extract was air-dried.<sup>12</sup> Petroleum ether extraction was used defatting. Methanol, Aqueous extract was mixed with 0.5% Sodium Carboxy Methyl Cellulose (Sod. CMC) and which was used for the anti-hyperglycemic activity.

### Phytochemical Screening

The crude extract obtained by using various solvents were analyzed for alkaloids, tannins, saponins, steroids, flavonoids, and phenolic compound using standard procedure of analysis.<sup>13</sup>

### Chemicals

Streptozotocin was purchased from Albino labs, Hyderabad, India. Glucose kit (GOD-POD Method)(Glucose oxidase-Peroxidase), were purchased from Classic Enterprises, Rajahmundry, India. All solvents used for extraction purchased from Classic Enterprises, Rajahmundry, India.

### Animal

Wistar albino male rats weighing 200-220g and albino mice (Either sex) weighing 20-25g were selected and housed in polypropylene cages in a room where the congenial temperature was 27°C ±1°C and 12 hrs light and dark cycles were maintained. The animals were allowed to acclimatize to the environment for 7 days and supplied with a standard pellet diet (VRK nutritional solution, Pune) and water *ad libitum*. All the experiments in this study were approved institutional animal ethical committee with CPCSEA registration number 1069/AC/07/CPCSEA, GIET School of Pharmacy.

### Acute Toxicity Study

Albino mice (Either sex) of 10 animals per group and weighing 20-25 g were administered graded dose (100-5000 mg/kg body weight, p.o.) of the methanolic and aqueous extracts of *Cucumis melo* leaf. After administration of the extract the mice were observed for toxic effects for 48-72 hr. The toxicological effects were observed in terms of mortality expressed as LD<sub>50</sub>. The number of animals dying time was noted. The LD<sub>50</sub> of the extracts (methanolic and aqueous) was determined by Litchfield and Wilcoxon (Litchfield and Wilcoxon, 1949) method.<sup>14</sup>

### Anti-Hyperglycemic Activity

Hyperglycemia was induced in Wistar albino male rats by single intraperitoneal injection of Streptozotocin (55 mg/kg)(STZ) in citrate buffer solution pH 4.8 after overnight fasting for 18 hours.<sup>15</sup> The rats were divided into seven groups of six rats in each group and were treated with single dose/day (*p.o.*) of standard drug or extracts of *Cucumis melo* leaf. The first group was given Standard pellet diet, 0.5% Sod.CMC, and water (Served as normal control). The second group was given a single dose of streptozotocin (55 mg/kg), *i.p.* After 72 hours of streptozotocin injection, this group received a daily dose of 0.5 % Sod.CMC (*p.o.*). Third group was administered with Standard Glibenclamide 10 mg/kg *p.o.* for 28<sup>th</sup> days (served as standard). The fourth to seven group was administered a daily dose of *Cucumis melo* leaf Methanol and aqueous extract at a dose 300 mg/kg and 500 mg/kg suspended in 0.5% Sod.CMC, *p.o.* (served as treatment groups) for 28 days, after inducing hyperglycemia.

### Collection of Blood Samples

On 0, 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup>, 28<sup>th</sup> day blood was collected by retro orbital sinus puncture, under mild ether

anesthesia in plane capillary tubes. Serum obtained by immediate centrifugation of blood samples using remi ultra cooling centrifuge at 3000 rpm for 15 minutes at room temperature and was directly used for estimating serum glucose. All samples were stored at 4°C until analysis.

#### Biochemical analysis

Serum glucose levels were carried out using respective diagnostic commercial kits (Transasia Bio-medicals Ltd, Solan, India) in semi auto analyzer (EON one vital diagnostics).

#### Statistical Analysis

The results were expressed as mean  $\pm$  SEM. The Streptozotocin control was compared with normal and the experimental results were compared with Streptozotocin control. Statistical analysis was carried out using one-way ANOVA followed by Dunnett test. Differences below  $P < 0.05$  implied statistically significance.

## Results

### Phytochemical screening

The results of phytochemical screening are given in Table No1. Indicated the presence of maximum amount of Carbohydrate, Tannins, Saponins, Flavonoids, steroids is present in Methanolic extract and aqueous extract contains Carbohydrate, Flavonoids, Saponin and Tannins.

### Acute toxicity study

The acute toxicity studies of chloroform, methanolic and aqueous extract of the CML were found to be non-toxic up to the dose 5000 mg/kg and did not show any mortality.

### Anti-Hyperglycemic Activity

The results of present study are given in Table No 2. The rats treated with streptozotocin showed significant increase in serum glucose level from 70 mg/dl to 120 mg/dl in rats. Treatment with CML different extract at the doses of 300 mg/kg and 500 mg/kg reduced the serum glucose levels when compared to the control and standard group rats.

**Table No 1: Phytochemical screening of *Cucumis melo* leaf extracts**

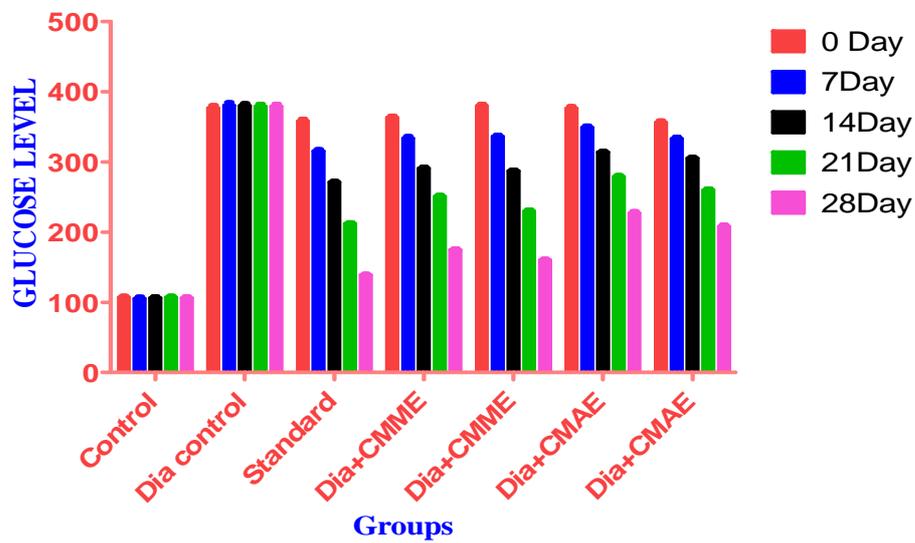
Chemical constituents	Methanolic extract	Aqueous extract
Carbohydrates	++	++
Tannins	++	++
Alkaloids	-	-
Saponins	++	++
Flavonoids	++	+
Glycosides	+	-
Steroids	++	+
Amino acids	+	+
Proteins	+	+

+ve Presence of Chemical constituents, -ve Absence of Chemical constituents, ++: Maximum Presence of Chemical constituents.

**Table 2: Showing decrease in blood glucose level in all Extracts**

S.No	Groups	Dose	0 Day	7 Day	14 Day	21 Day	28 Day
1	<b>Control</b>	CMC	106.8±2.0	105.6±2.0	106.2±1.8	107±2.0	106.2±2.0
2	<b>Diabetic control</b>	CMC	377.0±3.4*	380.4±3.9*	379.8±3.1*	379±3.4*	378.9±3.1*
3	<b>Diabetic +standard</b>	10mg/kg	358.2±2.7* *	315.5±2.1**	271.2±1.5**	212±1.8**	138.6±2.0**
4	<b>Diabetic + CMME</b>	300mg/kg	363.3±1.9* *	333.9±2.5**	291.0±1.6**	251±1.6**	174.9±1.1**
5	<b>Diabetic + CMME</b>	500mg/kg	380.0±2.0* *	336.0±2.2**	287.1±1.3**	230±1.7**	160.2±1.6**
6	<b>Diabetic + CMAE</b>	300mg/kg	376.2±3.1* *	348.8±2.2**	314.1±1.4**	279.0±1.9**	227.0±2.4**
7	<b>Diabetic + CMAE</b>	500mg/kg	356.4±2.2**	333.0±2.3**	305.4±1.2**	260±1.6**	208.2±2.4**

Values are expressed as Mean ± SEM of 6 rats in each group. \*P < 0.05 is compared to control group. \*\*P < 0.05 is compared to diabetic control group.



**Figure.1: Showing decrease in blood glucose level in all Extracts**

**Fig.1: Showing decrease in blood glucose level in all extracts.**

## Discussion

The present work has detected the antidiabetic effect of the methanolic and aqueous extract of *Cucumis melo* leaf in streptozotocin induced type-II diabetic rats. Streptozotocin injection caused diabetes mellitus, probably due to destruction of the  $\beta$ -cells of the islets of Langerhans of the pancreas.<sup>16</sup> Overproduction of glucose and decreased utilization by the tissues form the fundamental basis of hyperglycemia in diabetes mellitus.<sup>17</sup> Induction of diabetes with STZ is associated with a characteristic loss of body weight, which is due to increased muscle wasting<sup>18</sup> and loss of tissue proteins.<sup>19</sup> Diabetic rats treated with the *Cucumis melo* leaf methanolic and aqueous extract showed an increase in body weight as compared to the diabetic control, which may be due to its effect in controlling muscle wasting, i.e., by reversal of antagonizing.<sup>20</sup>

*Cucumis melo* leaf methanolic extract (500 mg/kg) has shown maximum anti-hyperglycemic activity comparing to other extracts (CMME 300mg/kg, CMAE 300mg/kg, 500mg/kg) which is equivalent to the Glibenclamide in streptozotocin induced diabetic rats (Figure.1).

## Conclusion

The methanolic and aqueous extract of the leaves of *Cucumis melo* has antidiabetic activity as it lowers serum glucose levels in diabetic rats (Figure.1). But methanolic extract is more active than aqueous extract. Hence to conclude that methanolic extract of *Cucumis melo* leaf having more anti- hyperglycemic activity. It also increases the body weight of diabetic rats. Hence, long-term studies of *Cucumis melo* leaf and its isolated compounds are necessary to elucidate the exact mechanism of action so as to develop it as a potent antidiabetic drug.

## Acknowledgement

The authors sincerely thank the management of GIET School of Pharmacy, Chaitanya Knowledge City, Rajahmundry, 533296, Andhra Pradesh, India, for providing the necessary facilities for carrying out this work.

## “Cite this article”

N. Babulreddy, S. P. Sahoo, S. Ramachandran M.D.Dhanaraju “Anti-Hyperglycemic Activity Of *Cucumis Melo* Leaf Extracts In Streptozotocin Induced Hyperglycemia In Rats ” Int. J. of Pharm. Res. & All. Sci.2013; Volume 2, Issue 4, 22-27

## References

1. Zimmet P, Alberti KG, Shaw J. Global and social implications of the diabetes epidemic. *Nature* 2001; 414:782-787.
2. Rang HP, Dale MM, Ritter JM, Moore PK. Pharmacology. 5th ed. London: Churchill Livingstone; 2003; 386- 387.
3. Khan A, Safdar M. Role of diet, nutrients, spices and natural products in diabetes mellitus. *Pakistan J Nutr?* 2003; 2:1-12.
4. Narendhirakannan RT, Subramanian S, Kandaswamy M. Biochemical evaluation of antidiabetic properties of some commonly used Indian plants on streptozotocin-induced diabetes in experimental rats. *Clin Exp Pharmacol Physiol* 2006; 33:1150-1157.
5. Kirtikar KR, Basu BD, Indian medicinal plants, M/S Periodical Experts, New Delhi, 1984; 1585-88.
6. Vijayvergia VA, Coughcide kachri. Rajasthan Patrika, Jaipur Issue, Available at: [www.actahort.org/members/showpdf?booknrar=1999;752](http://www.actahort.org/members/showpdf?booknrar=1999;752).
7. Hamendra Singh Parmar, Anand K, Protective role of *Mangifera indica*, *Cucumis melo* and *Citrullus vulgaris peel* extracts in chemically induced hypothyroidisma, *Chemico-Biological Interactions*, 2009; 177: 254–258.
8. Malhotra OP, Rani I, Occurrence, isolation & characterization of urease inhibitor from melon (*Cucumis melo*) seeds, *Ind. J. Biochem. Biophysics*, 1978; 15:229–231.
9. Vouldoukis I, Lacan D, Kamate C, Coste P, Calenda A, Mazier D, Conti M, Dugas B, Antioxidant and anti-inflammatory properties of a *Cucumis melo* LC. Extract rich in superoxide dismutase activity, *Journal of Ethnopharmacology*, 2004; 94:67–75.
10. Singh RC, Sisodia CS, Pharmacodynamic investigations into the diuretic activity of *Cucumis melo* seed (ether extract), *Ind. J. Med. Res.*, 1970; 58:505–12.
11. Simona De Marino, Carmen Festa, Franco Zollo, Maria Iorizzi, Phenolic glycosides from *Cucumis melo* var. inodorus seed, *Phytochemistry Letter* 2, 2009:130–33.
12. Mukharjee PK, Quality Control of Herbal Drug, first ed., *Business Horizon Publication*, New Delhi, 2002; p. 405-06.
13. Kokate CK, Practical Pharmacognosy, 4<sup>th</sup> ed. Vallabh Prakashan, Pune, 1996:107.

14. Litchfield JT, Jr. and Wilcoxon F, A simplified method of evaluating dose effect, *J. Pharmac.exp. Ther.*, 1949; 96: 99-113.
  15. Salahuddin M, Jalalpure SS. Antidiabetic activity of aqueous fruit extract of *Cucumis trigonus* Roxb. In Streptozotocin-induced diabetic rats. *J Ethnopharmacol* 2010; 127:565-567.
  16. Kavalali G, Tuncel H, Goksel S, Hatemi MH. Hypoglycemic activity of *Urtica pilulifera* in streptozotocin-diabetic rats. *J Ethnopharmacol.*2002; 84:241-5.
  17. Chattopadhyay R. Hypoglycemic effect of *Ocimum sanctum* leaf in normal and streptozotocin diabetic rats. *Indian J Exp Biol.*1993; 31:891-3.
  18. Swanston-Flat SK, Day C, Bailey CJ, Flatt PR. Traditional plant treatment for diabetes: Studies in normal and streptozotocin diabetic mice. *J Diabetol.*1990; 33:462-4.
  19. Chatterjea MN, Shinde R. Text Book of Medical Biochemistry. New Delhi: *Jaypee Brothers Medical Publishers*; 2002. Diabetes mellitus; 317
  20. Whitton PD, Hems DA. Glycogen synthesis in perfused liver of streptozotocin diabetic rats. *Biochem J.*1975; 21:150-153.
  21. Nolte MS, Karam JH. Pancreatic hormones and antidiabetic drugs. In: Katzung BG, editor. Basic and clinical pharmacology. 9th Ed. New York: *McGraw Hill Companies*; 2004; 708.
  22. Davis SN, Granner DK. Insulin, oral hypoglycemic agents, and the pharmacology of the endocrine pancreas. In: Hardman JG, Limbird LE, Gilman AG, editors. Goodman & Gilman's the pharmacological basis of therapeutics. 10th ed. New York: *McGraw Hill Companies*; 2002: 1687-1690.
-