



EVALUATION OF ANTI-DIABETIC ACTIVITY OF THE COMBINATION OF BITTER GOURD AND FENUGREEK SEEDS IN RAT.

Kapil Vishwakarma^a, Namrata Gupta^b, P.K. Dubey^c, Dr.Rajendra Bapna^d

^aStudent, Swami Vivekanand College of Pharmacy, Indore(M.P.), India

^bAsst. Prof., Swami Vivekanand College of Pharmacy, Indore(M.P.), India

^cPrincipal, Swami Vivekanand College of Pharmacy, Indore(M.P.), India

^dAsso. Prof., Swami Vivekanand College of Pharmacy, Indore(M.P.), India

ABSTRACT

Fenugreek (*Trigonella foenum-graecum*L. Leguminosae) and Bitter gourd are widely used in Indian Ayurvedic medicine for the treatment of diabetes mellitus. Antihyperglycaemic effect of the two different doses (385 and 550 mg/kg) of fenugreek and bitter gourd seeds extract evaluated in this study. Blood glucose, was determined in alloxan induced diabetic rats after oral administration of a fenugreek and bitter gourd seeds extract. A comparable hypoglycemic effect was evidenced from the data obtained after 7 and 28 days of oral administration of the extract. Oral administration of the seed extract at a dose of 550 mg/kg B.W. for 28 days showed a significant decrease in fasting blood glucose. Our results were greatly lower after oral administration of aqueous extract of fenugreek and bitter gourd seeds in alloxan - induced diabetic groups than Glibenclamide treated group.

Keywords: *Bitter Melon; Momordica charantia; Diabetes, Dietary Strategies, Alloxan Induced Diabetic Rats, Fenugreek, Diabetes Mellitus.*

1. INTRODUCTION

DIABETES:

It is a metabolic disorder characterized by hyperglycaemia, glycosuria, negative nitrogen balance, and generally symptom. A widespread pathological amendment is a thickening of the capillary basement membrane, a rise in vessel wall matrix, and cellular proliferation leading to vascular complications like lumen narrowing, early arteriosclerosis, in duration of capillary capillaries, retinopathy, neuropathy, and peripheral vascular insufficiency.

It is outlined as diabetes, that could be a group of metabolic diseases in which there are high blood glucose levels over a protracted amount. Symptoms include high blood glucose, frequent urination, increased thirst, and redoubled hunger. diabetes will cause several complications. Acute complications embody diabetic acidosis, non ketotic hyperosmolar coma, or death. Serious or long-run complications embody cardiovascular disease, stroke, chronic nephropathy, foot ulcers, and injury to the eyes.

TYPES OF DIABETES:

Type 1 Diabetes It is a chronic (lifelong) disease that occurs when the pancreas does not produce enough insulin to properly control blood sugar levels.

The most common symptoms of type 1 diabetes include:

- Abnormal thirst and xerotes
- Sudden weight loss
- Frequent micturition
- Lack of energy, tiredness
- Constant hunger
- Blurred vision

Causes: The bodies own immune system attacks and destroy beta cells in the pancreas that are responsible for creating the hormone insulin.

Type 2 Diabetes: Type 2 diabetes formerly called non-insulin-dependent diabetes is a disorder that is characterized by high blood glucose in the context of insulin resistance and relative insulin deficiency.

The foremost common symptoms are:

- Excessive thirst and xerotes
- Frequent micturition
- Lack of energy, tiredness
- Slow-healing wounds
- Recurrent infections in the skin
- Blurred vision
- Tingling or numbness in hands and feet.

Causes:-Type 2 diabetes occurs when the pancreas doesn't make enough insulin or the cells of the body become resistant to insulin.

2. MATERIAL AND METHODS

- **Collection and identification:**

Collection and identification of seeds (*Trigonella foenum-graecum* & *Momordica charantia*) were collected from Indore market M.P. and the plant samples were authenticated by Dr. Dwivedi A.P.S. College Rewa.

- **Plant materials:**

The powdered seeds (*foenum-graecum* & *Momordica charantia*) were kept in airtight containers in a deep freeze temperature until the time of further use. The seed extract was prepared by dissolving a known amount of seed powder in distilled water using a magnetic stirrer. It was then filtered and evaporated to dryness under reduced pressure. An aqueous suspension was prepared to facilitate easy handling. The drug suspensions were prepared freshly each time and administered orally. The dosage schedule for the drug was once a day.

- **Preliminary Phytochemical Analysis:**

Extract was subjected to preliminary phytochemical investigation for detection of Alkaloids, Carbohydrates, Tannins, Saponins and Terpenoids etc. Phytochemical screening was performed using standard procedure.

Table 1 - Table indicating presence of various phytochemical constituents.

<u>SERIAL NO.</u>	<u>TEST</u>	<u>RESULTS</u>
1.	Alkaloids	+
2.	Tannins	+
3.	Saponins	+
4.	Terpenoids	+
5.	Anthraquinones	+
6.	Flavanoïdes	+
7.	Reducing	+
8.	Sugars	+
9.	Phlobatanins	+
10.	Steroids	+
11.	Phenol	+
12.	Amino acids	+
13.	Proteins	+
	Quinones	+

Note:-

+ = Present

- = Absent

- **Pharmacological Methods:**

Animals

24 healthy adult male albino Wistar rats with a body weight of 120-180 gm obtained from the animal house of the Department of Pharmacology of the Swami Vivekanand College of Pharmacy, Indore, India, were used for this study. The animals were placed in standard cages maintained in 12-hour light/dark cycle (artificial lights, 7 a.m - 7 p.m) and air exhaustion cycle (15 min/h) under standard conditions (temperature $25\pm 2^\circ\text{C}$, relative humidity $60\pm 10\%$). All procedures were carried out in accordance with the conventional guidelines for experimentation with animals. Prior to experimental treatments, animals were fasted overnight but were allowed free access to water.

Induction of Diabetic:

Rats were injected intraperitoneally with freshly prepared solution of alloxan monohydrate in normal saline at a dose of 150 mg/kg BW. Alloxan is capable of producing fatal hypoglycemia as a result of massive pancreatic insulin release rats were treated with 20% glucose solution (5-10 ml) orally after 6 h. The rats were then kept for the next 24 h on 5% glucose solution bottles in their cages to prevent hypoglycemia. After 1 week, rats with moderate diabetes that exhibited glycosuria and hyperglycemia (i.e. blood glucose concentration >200 mg/dL) were taken for the experiment.

Note: 24 male albino Wistar rats were used, divided randomly into four groups (6 each)

Blood Collection

Under aseptic conditions, blood samples were collected on the first day before inducing alloxan and later on the 3rd, 7th, and 14th days of alloxanization from the tail vein. Blood glucose determination was performed by one touch glucometer strip test.

Statistical Analysis:

The data were expressed as mean \pm standard error (SE). The Significance of differences among the groups were assessed by using ANOVA, followed by Student-Newman-Keuls test. $P < 0.05$ (5%) were considered as significant.

As per present study, results of P value are 0.002684.

Picture – a

Anova: Single Factor						
SUMMARY						
Groups	Count	Sum	Average	Variance		
250.4	2	500.91	250.455	7.80125		
101.2	2	205.65	102.825	109.6681		
ANOVA						
Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	21794.62	1	21794.62	371.0692	0.002684	18.51282
Within Groups	117.4693	2	58.73465			
Total	21912.09	3				

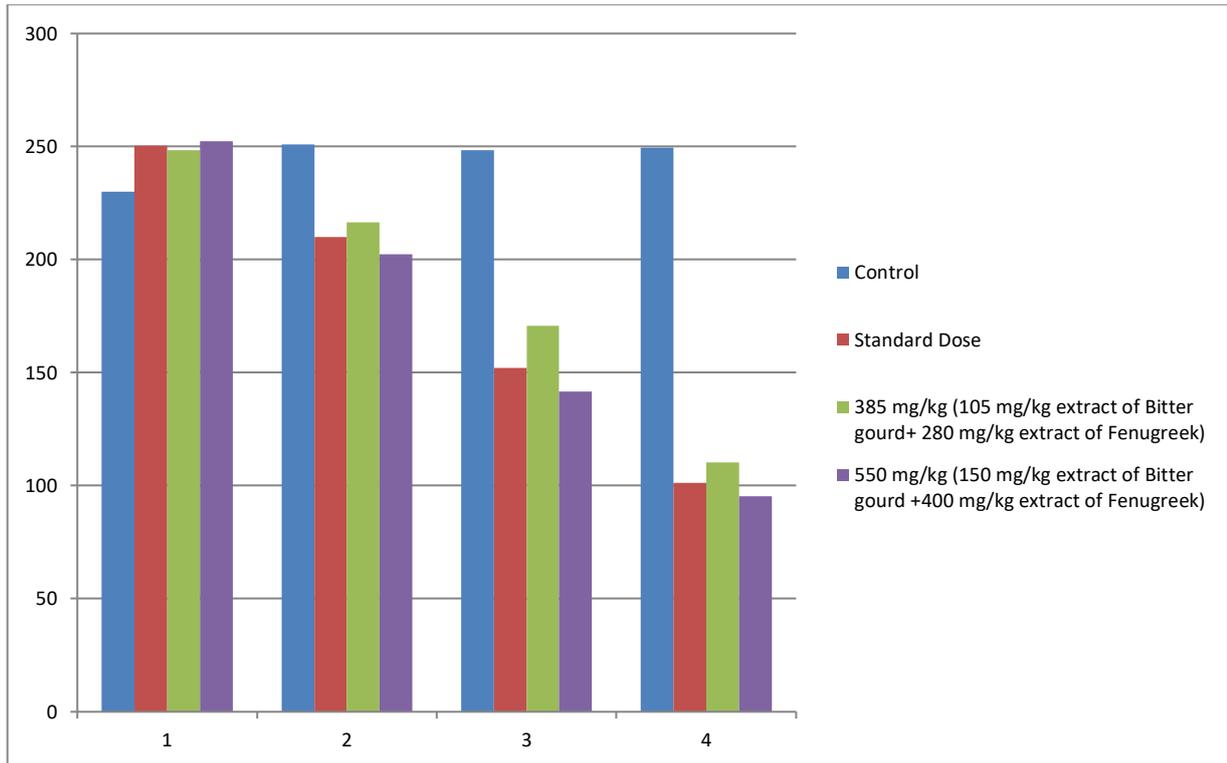


Fig. 1 - Effect of the combination of Fenugreek and Bitter gourd extract on Blood glucose (mg/dL)

The extract showed a significant reduction ($P < 0.05$) in blood glucose levels following sub-chronic administration when compared with the diabetic pre-treatment values. At day 28, the extract reduced the blood glucose level.

Treatment	Pre-treatment	Day 7	Day 14	Day 28
Control	230.2±1.35	250.99±1.34	248.48±0.35	249.67±0.24
Standard Dose	250.4±1.45	209.9±1.32	152.2±1.28	101.2±1.18
385 mg/kg (105 mg/kg extract of Bitter gourd+ 280 mg/kg extract of Fenugreek)	248.48±4.83	216.37±4.32	170.83±5.85	110.23±0.34
550 mg/kg (150 mg/kg extract of Bitter gourd +400 mg/kg extract of Fenugreek)	252.43±3.24	202.44±4.86	141.67±4.28	95.42±2.55

3. DISCUSSION

- Diabetes was induced by Alloxan and treated with the combination of *Momordica charantia* and *Trigonella foenum-graecum* did not show any alterations in behavior and no mortality was observed up to the 550 mg/kg dose level during the interventional period.
- By IP administration of *Momordica charantia* and *Trigonella foenum-graecum* with a dosage of 305 and 550 mg/kg resulted in a steady decrease in blood glucose levels of 252.43±3.24 mg/dl and 95.42±2.55 mg/dl after the trial which proves that *Momordica charantia* and *Trigonella foenum-graecum* possess anti-diabetic activity.
- By mixing each extract in varying proportions, *Momordica charantia* and *Trigonella foenum-graecum* were developed and evaluated. *Momordica charantia* and *Trigonella foenum-graecum* (305 and 550 mg/kg) anti diabetic activity was determined for Alloxan-induced diabetes in rats and Glibenclamide (5.0mg/kg body weight) was used as a standard drug.
- The investigational drug was administered for 28 days and the blood glucose level effect of the *Momordica charantia* and *Trigonella foenum-graecum* was analyzed on the 28th day after the intervention time.

- Oral administration of the seed extract at a dose of 550 mg/kg B.W. for 28 days showed a significant decrease in fasting blood glucose. Our results were greatly lower after oral administration of aqueous extract of *Momordica charantia* and *Trigonella foenum-graecum* seeds in alloxan- induced diabetic groups than Glibenclamide treated group.

4. CONCLUSION

- The preliminary phytochemical screening of *Trigonella foenum-graecum* and *Momordica charantia* were given a good results in the presence of flavonoids, alkaloids, anthraquinones, tannins phenol compounds, terpenoids, saponins, carbohydrates and proteins.
- Sub maximal dose (70 % of 150mg/kg bitter gourd seeds extract which is most effective dose + 70 % of 400 mg/kg fenugreek seeds extract which is most effective dose) values were nearby standard dose (glibenclamide 5 mg). It showed that sub-maximal dose is not more effect than glibenclamide drug.
- Maximal dose (100% of Bitter gourd extract + 100 % of Fenugreek extract) values were surpassed the standard drug values (glibenclamide) that was showing the synergetic effect which means combination of Bitter gourd and Fenugreek seeds extract are more effect.

Note: The most effective dose was selected by reviewing the research paper.

Acknowledgements

The authors are highly thankful to Dr. P.K. dubay sir principle of Swami Vivekanand College of Pharmacy for complete this project work. Special thank to Mrs. Namrata Gupta and Dr. Rajendra Bapna professor of pharmacology department for proper guidance.

REFERENCE

- [1] Tripathi K.D., "Essentials of pharmacology", Jayshree brothers medical publishers, 6th Edition, 254-274.
- [2] Loumpangou, Celestine Nkounkou, Baonda, Snelle Miakayizila, Ampa, Raoul, Ndinga, Arnold Murphy Elouma, Milandou, Longin Justin Clair Bonazaba, Bonose, Myriam, Ouamba, Jean Maurille, "Evaluation of antidiabetic activity of the ethanol extract of *Momordica charantia* L. and the identification of charantine by gas chromatography coupled with Mass spectrometry" *Journal of Medicinal Plants Research*, Vol. 13(14), pp. 321-328, 25 July, 2019.
- [3] Khan, Muhammad F, Abutaha, N, Nasr, Fahd A., Alqahtani, Ali S., Noman, Omar M., Wadaan, Noman & Mohammad A. M., "Bitter gourd (*Momordica charantia*) possess developmental toxicity as revealed by screening the seeds and fruit extracts in zebrafish embryos", *BMC Complementary and Alternative Medicine* volume 19, Article number: 184 (2019).
- [4] Chanda R, Samadder A, Banerjee J, Anti-diabetic Activity of *Momordica Charantia* or Bitter Melon, *Acta Scientific Pharmaceutical Sciences* (ISSN: 2581-5423), Volume 3 Issue 5 May 2019.
- [5] Rachana G, Anupam P, Ashish T, Pharmacological actions and potential uses of *Trigonella foenum-graecum*: A Review. *Asian Journal of Pharmaceutical and Clinical Research*.2009; 2 (4): 29-38.
- [6] Seyis, F. "Determination of Suitable Solvents for Extraction of Different Fruit Parts of Bitter Melon (*Momordica charantia* L.)" *Research Journal of Agricultural Sciences* 9(1): 18-22, 2016.
- [7] Modak D. "A Review – Antidiabetic Activity of herbal drugs", *Pharma tutor*, 2015, Volume 3, Issue 9, 36-42.
- [8] Jasim A (2014), Therapeutic Uses of Fenugreek (*Trigonella foenum-graecum* L.), *AMERICAN JOURNAL OF SOCIAL ISSUES AND HUMANITIES* 2014.
- [9] Efirid j, Choi M, Davies S, Mehra S, Potential for Improved Glycaemic Control with Dietary *Momordica charantia* in Patients with Insulin Resistance and Pre-Diabetes, *International Journal of Environmental Research and Public Health* 11(2):2328-45 • February 2014.
- [10] Joseph, Baby, Jini, D., "Antidiabetic effects of *Momordica charantia* (bitter melon) and its medicinal potency", *Asian Pacific Journal of Tropical Disease*, 2013, VL - 3, 93-102.
- [11] Kaviarasan S, Fenugreek (*Trigonella Foenum Graecum*) seeds: In health and disease, *Natural Products and Their Active Compounds on Disease Prevention*, 2012.
- [12] Anwar, Sirajudheen, Desai, Sandhya, Eidi, Maryam, Eidi, Akram, "Antidiabetic Activities of Fenugreek (*Trigonella foenum-graecum*) Seeds", 469-478, 2011.

-
- [13] Klomann, Sandra, Müller, Andreas, Pallauf, Josef, Krawinkel, Michael, "Antidiabetic effects of bitter gourd extracts in insulin-resistant db/db mice", *The British journal of nutrition*, (2010), VL - 104, 1613–1620.
- [14] Nibras, Auroba, Al-Abassi, N, Ibrahim, Orooba, Abdulhamza, Nibras, "Study Antidiabetic Effect of Momordica Charantia (bitter gourd) seeds on Alloxan Induced Diabetic Rats", *Iraqi Journal of Veterinary Sciences*, 2010, 165-170.
- [15] Tsai, C.-H, Chen E, Tsay H, Huang C, Bitter gourd (*Momordica charantia*): A review of its efficacy and safety for glucose homeostasis, VL - 35, *Nutritional Sciences Journal* (2010).
- [16] Lucas E, Dumancas G, Smith B, Clarke S, Arjmandi B, Health Benefits of Bitter Melon (*Momordica charantia*), *Bioactive Foods in Promoting Health* (2010).
- [17] Sharma N., "Potential antidiabetic herbal Drugs; A Comparative Review of Marketed Products", *Research journal pharmacognosy and phytochemistry*, 2010, Volume 2, Issue 2, 115-121.
- [18] Mowla A, Alauddin M, Rahman Md Atiar, Ahmed K. "Antihyperglycemic Effect of *Trigonella Foenum-Graecum* (Fenugreek) Seed Extract in Alloxan-Induced Diabetic Rats and Its Use in Diabetes Mellitus: A Brief Qualitative Phytochemical and Acute Toxicity Test on the Extract", *Afr. J. Trad. CAM* (2009) 6 (3): 255 – 261.
- [19] Helmy, Neveen, Abou El-Soud, Neveen, Khalil, Mona, Hussein, Jihan, Oraby, F.S.H, Farrag, Abdel Razik, "Antidiabetic Effects of Fenugreek Alkalioid Extract in Streptozotocin Induced Hyperglycemic Rats", *Journal of Applied Sciences Research*, 3(10): 1073-1083, 2007.
- [20] Srinivasan K, Fenugreek (*Trigonella foenum-graecum*): A Review of Health Beneficial Physiological Effects, *Food Reviews International - FOOD REV INT*, VL - 22, 2006.
- [21] Kitukale M. D., Chandewar A.V., "An overview on some Recent Herbs Having AtidiabeticPotential", *Research journal of pharmaceutical, Biological and chemical Sciences*, 2014, Volume 5, Issue 6, 190-196.
- [22] Cheekati R, Rao A, R V, A histological study of alloxan-induced diabetes on experimental male Wistar rats, "National Journal of Physiology, Pharmacy and Pharmacology " vol-7, 2017, 1329-1334.
- [23] Ramesh k, Yogesh HL, Kantikar SM and Prakash KB, Antidiabetic and Histopathological analysis of fenugreek extract on alloxan induced diabetic rats, "International Journal of Drug Development & Research" vol-2, 2010.
- [24] Cheekati R, Rao A, R V, A histological study of alloxan-induced diabetes on experimental male Wistar rats, "National Journal of Physiology, Pharmacy and Pharmacology " vol-7, 2017, 1329-1334