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# Synergistic effect of Trigonella foenum graecum with Metformin in the treatment of alloxan induced diabetic Wistar rats

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#### ABSTRACT

**Aim**: This study was aimed to assess the role of anti-diabetic activity of a combination of *Trigonella foenum greacum* and Metformin in alloxan-induced diabetic Wistar rats.

**Objectives**: To explore newer alternatives to alleviate diabetes and its complications.

**Materials and methods**: A combination of Metformin (250 mg/kg.bw) and an aqueous extract of Trigonella foenum graecum (400mg/kg.bw) was given orally over a period of 2, 4 and 8 weeks following induction of diabetes in male albino Wistar rats with alloxan. Blood glucose, bilirubin, lipid profile and renal function tests were analyzed. A description of statical data was applied for individual analysis as described in the figures. The interpretations were based on p< 0.005.

**Results**: In alloxan-induced diabetic rats, combination therapy induced a significant decrease in blood glucose level, urea, creatinine, uric acid, Total cholesterol, triglycerides, LDL and VLDL with a significant increase in HDL. On microscopy, there is an increase in the number of  $\beta$ -cells of islets of Langerhans of the pancreas.

**Conclusion:** The results showed that Trigonella foenum graecum with Metformin was far superior in the management of diabetes.

# **Keywords**: Combination therapy, Trigonella foenum graecum, alloxan, diabetes, Metformin. **INTRODUCTION**

Diabetes mellitus is a metabolic disorder resulting from insulin deficiency, characterized by altered hyperglycemia and metabolism of carbohydrates, protein and lipids. Untreated and persistent hyperglycemia leads to mortality and morbidity. The etiology is heterogeneous and includes heredity, diet, obesity, lifestyle and stress (Md Yaheya Md Ismail et al, Ravikumar et al 2010). Uncontrolled high blood glucose causes microvascular damage. Diabetes is highly prevalent in India which is often referred to as the diabetic capital of the world (Misra 2011).

The extracts of herbs have been tried since ancient times for the treatment of diabetes. Trigonella foenum greacum (TFG) has several potent ingredients which are biologically active and is recognized as highly effective in the treatment of hyperglycemia and its potentially crippling complications.

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Metformin (Met) finds the pride of place as the drug of choice amongst the oral hypoglycemic agents. It is used as a gold standard for the assessment of the efficacy of the OHA. It increases the peripheral utilization of glucose, decreases its intestinal absorption and increases the sensitivity of the receptors (Saenz, 2005; Okonkwo, 2014).

Alloxan selectively destroys insulin-secreting  $\beta$ -cells in the Islets of Langerhans (Fisher 1985). In addition, it has been widely accepted and used to induce diabetes in laboratory animals. The toxic action of alloxan on pancreatic beta cells involves oxidation of essential sulfhydryl (-SH) groups, inhibition of glucokinase enzyme, generation of free radicals and disturbances in intracellular calcium homeostasis (Szkudelski 2001, Lenzen 2008, Etuk 2010).

#### **MATERIALS AND METHODS:**

Animals: Wistar rats were procured from the National Institute of Nutrition, Hyderabad, India

Animal Care: All rats were kept under observation for 1 week prior to the experiments to permit the animals to adjust to the environment. All animals were fed standard rat pellets and tap water *ad libitum*. They were housed in a facility with 12–12 hr light– dark cycle that is maintained at  $25\pm2^{\circ}$ C. At the end of the experiment, the animals were anaesthetized with diethyl ether inhalation.

**Drugs and chemicals:** Alloxan was purchased from Pearala agencies, Hyderabad, India

Aqueous extract of Fenugreek was obtained from GR Herbals, Indore.

**Experimental Induction of Diabetes:** The animals were fasted for 18 hours and on the following day, 120 mg/kg.bw of alloxan was given intra-peritoneally (Dharmaseelan Sarasa et al, 2012). Since fatal hypoglycemia results, the rats were treated with 10% glucose per oral following the injection and 5% glucose was provided in feeding bottles. After 7 days animals with blood glucose >250mg/dL were taken up for the experiment.

Animal treatment protocol: In the present study male Wistar rats weighing 280–300gms were utilized for the study and are grouped as follows. Experiments were performed with the permission of the institutional ethics committee: Group I- Wistar rats were treated with normal saline (n=6)

Group-II: Wistar rats received single intra peritoneal injection of 120 mg/kg.bw of alloxan monohydrate (n=6).

Group-III: Aqueous extract of Trigonella foenum greacum was given at the dosage of 400 mg/kg and Metformin 250mg/kg.bw/day for 2 weeks from the third day of alloxanization (n=6).

Group-IV: Aqueous extract of Trigonella foenum greacum was given at the dosage of 400 mg/kg and Metformin 250mg/kg.bw/day for 4 weeks from the third day of alloxanization (n=6).

Group-V: Aqueous extract of Trigonella foenum greacum was given at the dosage of 400 mg/kg and Metformin 250mg/kg.bw/day for 8 weeks from the third day of alloxanization (n=6).

Statistical Analysis: All the results were expressed as Mean  $\pm$  SEM. The mean were analysed by one-way Analysis of Variance (ANOVA) with multiple comparison test of Student-Newman-Keuls test. Stastical analysis as well as plotting of graphs was carried out using Sigma Plot 13.0 (Syatat software, USA). Statistical significans is considered if the P < 0.05 (Figure: 1).

### **RESULTS:**

Biochemical parameters: There was a significant decrease of blood glucose to euglycemia and normal lipid profile and kidney function tests following treatment with 400 mg/kg.bw of TFG+ 250 mg/kg.bw of Metformin for a period of 8 weeks. The decrease was incremental over the study period as depicted in the Table:1.

Histopathological examination: Control rats showed normal architecture of pancreatic tissue. Diabetic rats showed necrosis and atrophy of  $\beta$ -cells in the Islets of Langerhans of the pancreas. Following treatment with 400 mg/kg.bw of TFG+ 250 mg/kg.bw of Metformin, there was an incremental increase in the number of functioning  $\beta$ -cells in sections studied at the end of 2 and 4 weeks. At the end of 8 weeks, there was a complete restoration of the  $\beta$ -cells which is shown by a normal uptake of Hemotoxylin & Eosin stain (Figure: 2).

DISCUSSION:

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This study was undertaken to ascertain the pharmacological action of a combination of Trigonella foenum greacum and Metformin on alloxan-induced diabetic Wistar rats. A synergistic effect was observed following treatment with Fenugreek and Metformin. Fenugreek contains bioactive organic compounds that can effectively cause hypoglycemia. The anti-diabetic effect was amplified when given with Metformin.

Treatment with a combination of Trigonella foenum greacum and Metformin reduced the catabolic nitrogen products moderately in diabetic rats. However the biochemical values of urea, uric acid and creatinine were found to be on the high normal side. There was a marked improvement of the levels of HDL, LDL, VLDL, cholesterol and triglycerides due to the synergistic effect of TFG + Metformin in the present experimental study.

Histology of the pancreas showed degenerative and necrotic changes of islets of Langerhans in diabetic

rats. There was no necrosis or fibrotic changes but regeneration of  $\beta$ -cells was seen when Metformin with TFG for 2 and 4 weeks. Normal regeneration of  $\beta$ -cells was noted at the end of 8 weeks. Thus there was a marked restoration of histological features due to the synergistic effect of Fenugreek and Metformin.

So far there is a paucity of literature regarding the use of a combination of TFG and Metformin in pharmacological doses. This study proves the synergistic effect of two individual drugs given as a combination to produce a very effective anti-diabetic result without any side effects. The various biochemical assays used in this study show a marked decrease in the levels of glucose.

# CONCLUSION:

Metformin alone is a potent oral hypoglycemic agent. However, a dramatic synergy was noticed when it is used in combination with TFG with fewer or no side effects in the management of diabetes.

Table.1: Effectiveness of Trigonella Foenum greacum (TFG) and Metformin (Met) combination on biochemical parameters						
S.NO	Parameters	G-1	G-2	G-3	G-4	G-5
		Control	Diabetic	TFG+MET-	TFG+MET-	TFG+MET-
			rats	2 weeks	4 weeks	8 weeks
1	Glucose	94.7 ± 5.1	328.7 ± 11.9	214.5 ± 9.9	137.7 ± 12.8	113.5 ± 7.6
2	Cholesterol	76.2 ± 10.6	177.3 ± 14.6	128.9 ±18.2	99.5 ±9.5	87.6 ±5.1
3	Triglycerides	78.3 ± 8.4	167.2 ± 11.8	138.6 ±7.8	115.4 ± 9.5	91.3 ± 7.9
4	HDL	$40 \pm 2.6$	21 ± 2.6	27.8 ±3.5	$31.2 \pm 2.6$	35.3 ± 2.4
5	LDL	20.6 ± 8.8	122.9 ± 12.8	73.3 ± 18.6	45.3 ± 11.4	33.98 ± 7.4
6	VLDL	$15.6\pm1.7$	$33.4\pm2.4$	$27.7 \pm 1.6$	23.1 ± 1.9	$18.3\pm1.6$
7	Bilirubin	$0.8 \pm 0.1$	$2.6 \pm 0.3$	$1.8 \pm 0.1$	$1.3 \pm 0.0$	$0.9\pm0.6$
8	Urea	$23.2\pm4.6$	80.3 ± 3.3	$58.5\pm9.7$	$47.0\pm5.9$	33.7 ± 3.4
9	Uric acid	$2.8 \pm 0.2$	8.5 ± 0.1	6.3 ± 0.3	$4.5\pm0.2$	3.7 ± 0.3
10	Creatinine	$0.6 \pm 0.2$	$1.8 \pm 0.1$	$1.5 \pm 0.1$	$1.2 \pm 0.1$	$0.9 \pm 0.3$

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## **REFERENCES:**

- Md Yaheya Md Ismail, Ravikumar.R et al (2010). Traditional Indian spices useful in Diabetes Mellitus, Research Journal of Pharmaceutical, Biological and Chemical Sciences.1 (3) Page No. 30-34.
- 2. Misra P. (2011) A review of the epidemiology of diabetes in rural India.
- 3. Saenz A (2005). Alpha-glucosidase inhibitors in the early treatment of type 2 diabetes, Vascular Health Risk Management.
- 4. Okonkwo PO (2014). Comparative Effects of Antidiabetic Drug, Metformin and Deferoxamine, on Serum Lipids, Serum Ferritin and Endocrine Indicators of Diabetes Mellitus Complications in Sreptozotocin Diabetic Rats.

- 5. Fisher J.(1985). Drugs and chemicals that produce diabetes.Trends Pharmacol. Sci. 6, 72–75.
- 6. Szkudelski (2001). The Mechanism of Alloxan and Streptozotocin Action in B Cells of the Rat Pancreas, Res. 50: 536-546.
- Lenzen S (2008). The mechanisms of alloxan and streptozotocin-induced diabetes.Diabetologia51: 216-26.
- Etuk EU (2010). Animals models for studying diabetes mellitus. Agric Biol J N Am. 1:130-4.
- 9. Dharmaseelan Sarasa, Sekaran Sridhar and EgambaramPrabakaran (2012): Effect of an antidiabetic extract of Trigonella foenum graecumon normaland alloxan induced diabetic mice. International Journal of Pharmacy and Pharmaceutical Sciences; ISSN-0975-1491; Vol 4, Issue1.