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Research

Evaluation of Antidiabetic Activity Of Trigonella Foenum Graecum Leaves In Alloxan Induced Type-II Diabetis

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Abstract

Trigonella foenum graecum leaves have been used as antidiabetic remedies in many cultures for thousands of years. The aim of this review is to address the existing evidence on antidiabetic effects of the Trigonella foenum graecum. The hypoglycaemic effects, antidyslipidaemics, antioxidative effects and the safety of the Trigonella foenum graecum leaves have been scientifically validated by using in vitro and in vivo studies. Multiple cell regeneration, insulin β mechanisms responsible for hypoglycaemic effects of Trigonella foenum graecum including release and insulin like actions of some compounds isolated were identified. Epicatechin, a cell regeneration and β flavonoid isolated from the leaves has shown insulin-like effects, effects on insulin release. Several compounds including pterostilbene and marsupsin isolated from the Trigonella foenum graecum leaves were identified as compounds with hypoglycaemic effects.Few investigations focused on the antidiabetic effects of Trigonella foenum graecum latex have demonstrated strong inhibitory effects of the latex on α-amylase and α-glucosidase activities and on protein glycation. Investigations focusing on the antidiabetic effects and possible toxicity of the Trigonella foenum graecum are essential to validate its efficacy and safety.

Keywords: Anti diabetic, Neuropathy,diabetes, Trigonella foenum graecum etc.

INTRODUCTION

Diabetes mellitus is one of the most common endocrine diseases in all populations and all age groups. It is a syndrome of disturbed intermediary metabolism caused by inadequate insulin secretion or impaired insulin action, or both. Diabetes mellitus comprises of heterogeneous group of disorders characterized by hyperglycemia, altered metabolism of carbohydrates, lipids and proteins. Diabetes mellitus is associated with complications such as nephropathy, retinopathy, neuropathy and cardiovascular disease.[1]

Diabetes is mainly classified into three types as: Type-I (Insulin-Dependent Diabetes Mellitus, IDDM) and Type-II (Non- Insulin-Dependent Diabetes Mellitus, NIDDM), Type-III(Gestational diabetes. Both these types are associated with excessive morbidity and mortality. Type I diabetes accounts for 5% to 10% of diabetes, usually occurs in children or young adults. This disease is caused by autoimmune destruction of the pancreatic β-cells that secrete insulin. The process involves a smoldering destructive process that can persist for several years and ultimately leading to failure of insulin secretion. Patients with type I diabetes require insulin therapy for survival and most patients ultimately develop devastating complications of this disease[1-4].

Type II diabetes accounts for 90% to 95% of all patients with diabetes and is increasing in prevalence. Some of the known environmental factors that contribute to development of type–II diabetes are obesity, a sedentary lifestyle, and aging. Insulin resistance is a characteristic metabolic defect in the great majority of patients with type II diabetes. As a consequence of insulin resistance, the β-cell produces increased amounts of insulin, and, if sufficient, the compensatory hyperinsulinemia maintains glucose levels within the normal range.

In those individuals destined to develop diabetes, β-cell function eventually declines, and relative insulin insufficiency occurs. Thus, insulin resistance combined with β-cell failure leads to the decompensated hyperglycemic diabetic state.

Insulin stimulates stored glucose in the liver as glycogen and in adipose tissue as triglycerides and amino acid storage in muscle as protein; it also promotes utilization of glucose in muscle for energy. Insulin inhibits the breakdown of triglycerides, glycogen, and protein and the conversion of amino acids to glucose (gluconeogenesis). These pathways are increased during fasting and in diabetic states. The conversion of amino acids to glucose and of glucose to fatty acids occurs primarily in the liver.

- Sulfonylureas of 1st generation drugs (Tolbutamide and chlorpropamide) and second generation drugs (Glibenclamide and Glipizide) main action is on B cells stimulate insulin secretion; their use also results in reduction of hepatic glucose production, reversal of the post-receptor defect, and increase in the number of insulin receptors.
- Biguanides, such as Metformin, increase glucose uptake and utilization in skeletal muscle and reduce hepatic glucose production.
- The nonsulfonylurea insulin secretagogues repaglinide and nateglinide bind to a specific site on the sulfonylurea receptor and increase insulin secretion, although they are short-acting agents.
- Thiazolidinediones, such as Pioglitazone and Rosiglitazone, bind to a nuclear receptor called peroxisome proliferator-activated receptor-Y (PPAR-□□ which is complexed with retinoid X receptor (RXR) increases lipogenesis and enhances uptake of fatty acids and glucose, decrease insulin resistance by enhancing insulin-mediated glucose disposal by muscle.
- α-glucosidase inhibitors, such as acarbose and miglitol, block starch, sucrose, and maltose absorption[5-7].

Aim And Objectives

The evaluation of Trigonella foenum graecum leaves extract for antidiabetic activity in rats.

- To determine the Trigonella foenum graecum properties
- To evaluate the protective effect on diabetic nephropathy of Trigonella foenum graecum leaves extract by biochemical estimations in rats.

MATERIALS AND METHODS

Materials	**Source**
Sodium citrate	Virat labs, Hyd, India
Diethyl ether	Finar chemicals limited, Ahmadabad.
Methanol	E-Merk, Mumbai, India.
Normal saline	Claris life sciences, Ahmadabad, India.
Formaldehyde	Finar chemicals limited, Ahmadabad, India
Chloroform	Molychem, Mumbai, India.
Alloxan monohydrate	Sigma, St Louis, U.S.A.
Metformin	MSN Formulations, HYD, India.
Equipments used	
Centrifuge	Remiequipments Pvt, Ltd, Hyd, India.
Shimadzuelectronic balance	Toshvin Analytical Pvt. Ltd, India
Shimadzu UV-spectrophotometer	Toshvin Analytical Pvt. Ltd, Mumbai.
Inverted microscope	Boeckl + co, Hamburg.

Plant description

Fenugreek is an annual plant in the family Fabaceae, with leaves consisting of three small obovate to oblong leaflets. It is cultivated worldwide as a semiarid crop. Its seeds and its leaves are common ingredients in dishes from South Asia.

Description of the Plant

An erect, annual, aromatic herb, 30-45 cm high. Leaves pinnately 3-foliolate; leaflets 2-2.5 cm long, oblanceolate-oblong toothed. Flowers 1-2, axillary, sessile, corolla much exerted. Pod 5-7.5 cm long with a long persistent beak, often falcate, 10-20 seeded.

Chemical Constituents

Trigogenin, neotrigogenin, diosgenin, yamogenin, gitogenin, 4-hydroxyisoleucine, vitexin, isovitexin, saponaretin, homoorientin, vicenin-1, vicenin-2 and two flavonoid glycosides quercetin and luteolin and steroidal saponins have been isolated from seeds. Seeds also contain essential oil, fixed oil, fatty acids, proteins, large number of amino acids, carbohydrates, vitamins A, B1 and C, nicotinic acid, minerals and several coumarins. Saponin isolated from stems on hydrolysis yield a sapogenin, while that isolated from leaves gave diosgenin, tigogenin and gitogenin. Presence of β-sitosterol, kaempferol, quercetin, saponins - graecunin A, graecunin B and graecunin C have been reported in leaves (Ghani, 2003; Rastogi & Mehrotra, 1990 & 93).

Pharmacology

Chemical Constituents

In laboratory tests, fenugreek has been found to contain 4-hydroxyisoleucine (4-OH-Ile), fat, diosgenin, iron, phenolic acids, protein, and protodioscin.

Analgesic effects

In a rat study, *Trigonella foenum-graecum* extract showed analgesic activity, which may be similar to nonsteroidal anti-inflammatory drugs (NSAIDs) via the spinal 5-HT system or purinoceptors[1, 12].

Antiadhesive properties

Bactericidal and anti-adhesive properties against *Helicobacter pylori* have been studied[5].The bactericidal activity of the extract was assessed by a standard kill-curve with seven strains of *H. pylori*. The anti-adhesive property was assessed by the inhibition of binding of four strains of FITC-labeled *H. pylori* to stomach sections. Fenugreek was found to have no bactericidal effect on any of the isolates.

In a review article, authors presented evidence that numerous agents such as diosgenin (fenugreek) identified from fruits and vegetables can interfere with several cell-signaling pathways.[32] The results of several studies indicate that a diet rich in fresh vegetables protects against several common epithelial neoplasms.This probable effect has been related to specific micronutrients contained in vegetables. A case-control study and systematic assessment of the relationship between vegetable intake and the risk of gallbladder cancer was conducted in 153 patients with gallbladder cancer and 153 controls with gallstone disease. Each patient's consumption of vegetables was assessed by using a food frequency questionnaire. The frequency of vegetable consumption was divided into three levels: ≥3 days/week, 1-2 days/week and no or rare consumption. Participants were divided into three groups according to the level of vegetable intake. Odds ratios and 95% confidence intervals were computed for subsequent levels of vegetable consumption compared with the high level of consumption. A low consumption of vegetables showed an increase in odds ratio for gallbladder cancer for

Almost all the vegetables studied. A significant inverse trend was observed for green leafy vegetables and gallbladder cancer. An inverse association was observed for amaranth with an OR of 3.45 for the low vs. high level of consumption. Corresponding values were 2.14 for spinach, 1.86 for bathua, 1.02 for bengalgram leaves, 2.26 for cabbage, 3.06 for fenugreek leaves, 1.95 for mustard leaves and 1.44 for radish leaves. An inverse relationship between the risk of gallbladder cancer and the level of vegetable consumption was observed.

Antioxidant activity

In an ethanol toxicity rat study, an aqueous extract of fenugreek seeds prevented the rise in lipid peroxidation and enhanced antioxidant potential.[9] These results are supported by *in vitro* evidence in diabetic human erythrocytes, that polyphenol acids from fenugreek seeds showed a concentration-dependent inhibition of lipid peroxidation[2].

Antiplatelet activity

In a rat study, a fenugreek extract inhibited ADP (10^{-5}M) induced platelet aggregation (IC_{50}=1.28mg/mL)[12].

Exercise recovery effects

trained male cyclists, a glucose beverage and 4-hydroxyisoleucine isolated from fenugreek seeds significantly increased muscle glycogen concentration 63% from immediately post exercise to four hours after exercise compared to the control[28].

Hepatoprotective activity

In an *in vitro* study using Chang liver cells treated with ethanol, a polyphenolic extract of fenugreek seeds significantly and dose-dependently increased cell viability by reducing oxidation and apoptosis[8].

Hypoglycemic effects

Hypoglycemic effects of fenugreek observed in animal studies have been associated with a fraction that contains the testa and endosperm of the defatted seeds, called the "A" subfraction. These effects have not been observed with lipid extracts.Hypoglycemic effects have been attributed to several mechanisms: Sauvaire et al. demonstrated that the amino acid 4-hyroxyisoleucine in fenugreek seeds increases glucose-induced insulin release *in vitro* in human and rat pancreatic islet cells[16].This amino acid appeared to act only on pancreatic beta cells, since somatostatin and glucagon were not altered in the study. However, another *in vitro* study indicates that fenugreek

Seed extract phosphorylates a number of proteins, including the insulin receptor, insulin receptor substrate 1 and p85 subunit of PI3-K, in both 3T3-L1 adipocytes and human hepatoma cells, HepG2[17].These results suggest that fenugreek's effects may be due to activation of the insulin-signaling pathway in adipocytes and liver cells. In human studies, fenugreek reduced the area under the plasma glucose curve (AUC) and increased the number of insulin receptors via an unclear mechanism[10].Also, a combination of bittergourd, jamun seeds, and fenugreek seeds significantly reduced fasting and postprandial glucose level of the diabetic patients[15]. Fenugreek seeds have also been postulated to exert hypoglycemic effects by stimulating glucose-dependent insulin release by beta cells[13], or via inhibition of α-amylase and sucrase activity[14]. A unique major free amino acid, 4-hydroxyisoleucine (4-OH Ile), has also been characterized as one of the active ingredients for blood glucose control[11].

Insulin sensitization effects

When administered to type 2 diabetic rats, the amino acid 4-hydroxyisoleucine extracted from fenugreek seeds increased peripheral glucose utilization and decreased hepatic glucose production, thereby improving insulin resistance[6].Chronic ingestion of 4-hydroxyisoleucine significantly reduced insulinemia.

Pharmacodynamics/Kinetics

Pharmacokinetic data are not available for all components of fenugreek, or for the compound as a whole. Saponins present in fenugreek are believed to be primarily absorbed in the terminal ileum[34]

In a rabbit study by Zhao et al., after post-intragastric injection of fenugreek extract, the pharmacokinetic parameters of one compartment model were half-life, t½ (Ka) = 0.9 hr, t½ (Ke) = 2.2hr, volume of distribution = 0.64L/kg and AUC = 1.93mg*min/L.[37] After intravenous injection, the two compartment open model parameters were t½ (Ka) =10.8 min, t½ (Ke) = 44 min, K2,1 = 0.044/min, K1,0 = 0.026 min, K1,2 = 0.017/min, and the AUC = 931mg*min/L.

Methodology

Collection and Authentification of Plant Material

The Aerial Parts of *Trigonella foenum graecum* were collected and authenticated

Extraction of Plant Material

The plant is grinded in to a coarse powder with the help of suitable grinder.

Cold Extraction (Ethanol Extraction)

In this work the cold extraction process was done with the help of ethanol. About 200gms of powdered material was taken in a clean, flat bottomed glass container and soaked in 750 ml of ethanol. The container with its contents were sealed and kept for period of 7 days accompanied by continuous shaking with the shaker. The whole mixture then went under a coarse filtration by a piece of a clean, white cotton wool[34-36].

Evaporation of Solvent

The filtrates (ethanol extract) obtained were evaporated using Rotary evaporator in a porcelain dish. They rendered a gummy concentrate of greenish black. The extract was kept in vaccum dissecator for 7 days.

% Yield value of Ethanol Extract from Aerial Parts of Trigonella foenum graecum Plant

- Powder taken for extraction = 200gm
- Weight of the empty china dish = 53.70gm
- Weight of the china dish with extract = 73.24gm
- Weight of the extract obtained = (73.24-48.70) gm = 24.54 gm
- % yield of ethanol extract = (weight of extract)/(powder taken for extraction) × 100
  = 24.54/200 ×100 = 12.27 %.

Phenolic Constituents Extracts

Expereimental Animals

Healthy Adult Male wistar rats of 8-10 weeks old with Average weight in the range of 150-180gms were selected. Animals are housed 4 per cage in temperature controlled (27 ^{0}C ±3 ^{0}c) room with light/dark cycle in a ratio of 12:12 hrs is to be maintained. The Animals are allowed to acclimatize to the environment for seven days and are supplied with a standard diet and water *ad libitum*. The prior permission was sought from the Institutional Animal Ethics Committee (IAEC) for conducting the study[36].

Acute toxicity studies

The Acute oral toxicity test of the extracts was determined prior to the experimentation on animals according to the OECD (Organisation for Economic Co-operation and Development) guidelines no 423. Female Albino wistar rats (130-200 g) were taken for the study and dosed[41]

Once with 1000 mg/kg. The treated animals were monitored for 14 days to observe general clinical signs and symptoms as well as mortality. No mortality was observed till the end of the study revealing the 1000 mg/kg dose to be safe. Thus, 1/10 and 1/20 doses of 1000 mg/kg i.e. 100 mg/kg and 50 mg/kg were chosen for subsequent experimentation.

Induction procedure

Diabetes mellitus or hyperglycemia was induced in rats by administration of alloxan monohydrate (2,4,5,6-tetraoxypyrimidine; 2,4,5,6-primidinetetrone) at dose of 120mg/kg intraperitoneally in normal saline[23]. After one hour of alloxan administration the animals were given feed ad libitum. The animals were kept fasting overnight and blood glucose levels were estimated before and after 72hrs of alloxan treatment. Animals showing blood glucose levels of >200mg/dl is considered as diabetic and were used for study.

Experimental Study Design for Diabetic screening

Diabetic rats were divided in to five groups with each group four animals.
Group-I: Rats served as normal control group.
Group-II: served as diabetic/disease control.
Group-III: Diabetic rats treated with *Trigonella foenum graecum* at a dose 50mg/kg.
Group-IV: Diabetic rats treated with *Trigonella foenum graecum* at a dose of 100mg/kg
Group V: Diabetic rats treated with Metformin (standard drug) at 450mg/kg[24].
The treatment was given for 14days and blood samples were collected at different intervals.

Collection of blood samples

Blood samples were collected from all the groups of animals at 0, 7,15th day intervals through puncture of retro orbital plexus and were centrifuged at 3000 revolutions per minute (rpm) for 15 minutes. Serum was separated and stored at -20ºc and then used for estimating blood glucose levels.

Experimental Study Design for Diabetic neuropathy screening

Group-I: Rats served as normal control group.
Group-II: served as diabetic/disease control.
Group-III: Diabetic rats treated with *Trigonella foenum graecum*, at a dose 50mg/kg (low dose).
Group-IV: Diabetic rats treated with *Trigonella foenum graecum* at a dose of 100mg/kg (high dose).
Group V: Diabetic rats treated with Diclofenac sodium (standard drug) at 100mg/kg.
All the animals are tested for tail flick and thermal hypoalgesia Eddies plate method response to find out the peripheral neuropathy[25]

Statistical analysis

All the values will be expressed as mean ±standard deviation (S.D). Statistical comparisons between different groups will be done by using one way analysis of variance .P value <0.05 will be considered as statistically significant.

Evalution parameter
Glucose Method: GOD/POD method
Principle

$$\text{D-glucose} + H_2O + O_2 \xrightarrow{\text{glucose oxidase (GOD)}} \text{gluconic acid} + H_2O_2$$

$$H_2O_2 + \text{4-AAP} + \text{Phenol} \xrightarrow{\text{peroxidase (POD)}} \text{Quinoneimine dye} + H_2O$$

Procedure

- Wavelength/filter : 505 nm (Hg 546 nm) / Green
- Temperature: 37º C / R.T.
- Light path : 1 cm
- Pipette into clean dry test tubes labeled as Blank (B), Standard (S) and Test (T).

Addition Sequence	B (ml)	S (ml)	T (ml)
Glucose Reagent L1)	1.0	1.0	1.0
Distilled Water	0.01	--	--
Glucose Standard S)	--	0.01	--
Sample	--	--	0.01

Mix well and incubate at 37ºC for 10 min or at R.T. (25ºC) for 30 mins. Measure absorbances of the Standard (Abs.S) and Test Sample (Abs.T) compare these against the Blank within 60 mins.

Tail Flick Method

Healthy albino rats weighing about 150-200gm were taken. The animals were divided into five groups of 6 animals each. Group I served as control and received drugless (0.5%, w/v, CMC; 10 ml/kg) vehicle. Group II served as standard and were injected Diclofenac sodium (9 mg/kg) intraperitoneally. Group III, IV and V were treated orally with EGCG 50, 100 mg/kg body weight respectively. They were divided into different groups, numbered and placed into individual restraining cages leaving the tail hanging out freely. The animals are then allowed to adapt in the cages for 30 minutes before testing. The lower 5cm portion of the tail was marked and immersed in a cup of freshly filled warm water of exactly 55 o C. Within a few seconds the rat reacts by withdrawing the tail. The reaction time was recorded by a stop watch. After each determination the tail was carefully dried. The reaction was determined before oral administration of respective treatments which was recorded as zero minutes reading. After the drug was administered the reaction time was recorded at an interval of 30, 60, 90, 120 and 150 mins. The cut off time of the immersion is 15 seconds. The mean reaction time was recorded for each group and compared with the value of standard drug.

Eddy's hot plate method Thermal hypoalgesia

Male albino mice weighing 22-25g were taken. The animals were divided into five groups of 6 animals each. Group I served as control and received drugless (0.5%, w/v, CMC; 10 ml/kg) vehicle orally. Group II served as standard and were injected Diclofenac sodium (9 mg/kg) intraperitoneally. Group III, IV and V were treated orally with EGCG of 50, 100 mg/kg body weight respectively. The animals were individually placed on the hot plate maintained at 55°C, one hour after their respective treatments. The response time was noted as the time at which animals reacted to the pain stimulus either by paw licking or jump response, whichever appeared first. The cut off time for the reaction was 15 seconds 15. The test was carried before the treatment and at 30, 60 and 90 min after administration.

RESULTS & DISCUSSION

Table 1: Effect of *Trigonella foenum graecum (EETFG)* on serum glucose levels (mg/dl) in diabetic rats

Groups/Interval	0^{th} Day	7^{th} Day	15^{th} Day
Normal	83.3±4.23	79.1±5.36	77.7±5.62
Diabetic control	283.8±5.01	286.4±12.4	300.3±8.64
EETFG (50mg/kg)	293.1±9.83	192.1±12.3**	100.3±12.5**
EETFG (100mg/kg)	280.5±42.4	185.2±11.2***	94.2±7.2***
Metformin (450mg/kg)	271.0±13.5	80.2±6.4***	70.1±6.3**

*All the values of mean±SD; n=6; ** indicates p<0.01, *** indicates [a]p<0.001 vs diabetic control.*

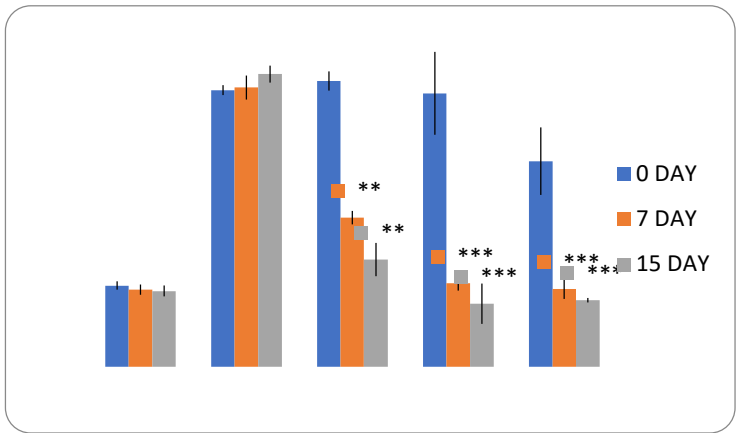

Fig 1: Effect of EETFG on serum glucose levels (mg/dl) in diabetic rats
*All the values of mean±SD; n=6; ** indicates p<0.01, *** indicates ªp<0.001 vs. diabetic control.*

Table 2: Diabetic Neuropathy screening by tail flick response

Group	Mean latency period			
	0	30	60	90
Normal	2.18±0.12	2.31±0.20	2.43±0.27	2.49±0.32
EETFG (50mg/kg)	2.79±0.20	2.94±0.34	3.10±0.45	3.19±0.40
EETFG (100mg/kg)	3.11±0.36	3.28±0.28	3.42±0.1	3.55±0.20
Diclofenac sodium (100mg/kg)	2.25±0.35	6.91±0.20*	9.74±0.34*	11.32±0.22*

*All the values of mean±SD; n=6; ** indicates p<0.01, *** indicates ªp<0.001 vs. diabetic control.*

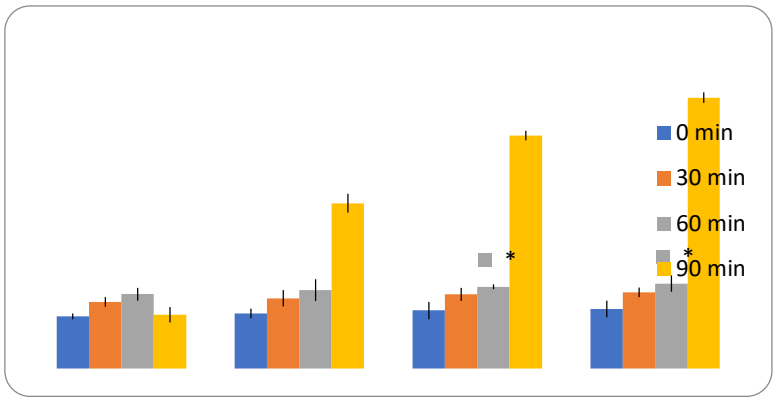

Fig 2: Effect of EETFG on Diabetic neuropathy by tail flick in diabetic rats
*All the values of mean±SD; n=6; ** indicates p<0.01, *** indicates p<0.001 vs. diabetic control.*

Table 3: Diabetic Neuropathy screening by Thermal hypoalgesia response

Group	Mean latency period				
	0	30	60	90	120
Normal	3.08± 0.4	3.1± 0.33	3.13± 0.6	3.01± 0.51	2.95± 0.34
EETFG (50mg/kg)	3.31± 0.29	3.48± 0.30	3.51± 0.34	3.54± 0.42	3.55± 0.25
EETFG (100mg/kg)	3.01± 0.35	3.19± 0.30	3.22± 0.40	3.27± 0.37	3.25± 0.32
Diclofenac sodium (100mg/kg)	2.70± 0.40	4.08± 0.38*	4.35± 0.45*	4.70±0.19*	5.40±0.22*

*All the values of mean±SD; n=6; ** indicates p<0.01, *** indicates ªp<0.001 vs. diabetic control.*

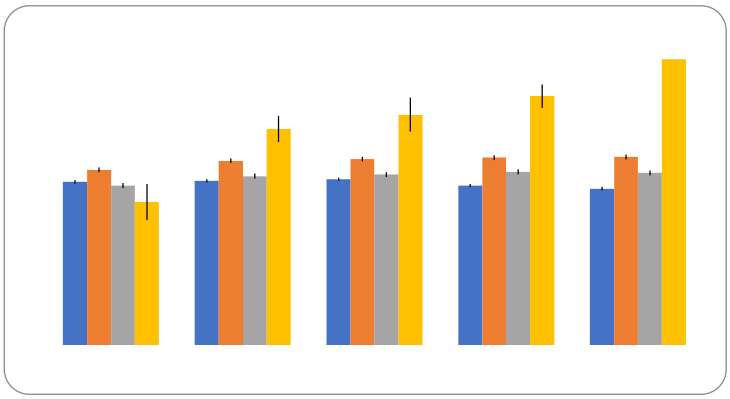

Fig 3: Effect of EETFG on Diabetic neuropathy by Thermal hypoalgesia method in diabetic rats

The present study was aimed to evaluate the anti diabetic activity of *Trigonella foenum graecum aerial parts*. The activity was measured by estimating various biomarkers like blood glucose levels, in experimental rats.

In the previous studies it was shown that alloxan monohydrate induced to diabetes mellitus. When given in a dose of 120mg/kg to rats intraperitoneally as evidenced in study[23] In the present study alloxan was administered in a single dose to induce diabetes mellitus in rats at the dose of 120mg/kg.The *Trigonella foenum graecum* has reported anti-microbial properties but the effect of the plant extract on antidiabetic, were not reported yet and so the plant was chosen for the study.

Alloxan forms an increased glucose levels that generates diabetes. Pretreatment with *Trigonella foenum graecum* produced significant decrease in glucose levels indicating the protective effect of tissue. On alloxan treatment a dose dependent decrease in glucose levels were observed. Pretreatment with *Trigonella foenum graecum* and metformin produced significant alteration in levels.

Diabetic neuropathy alterations were tested using thermal hypoalgesia and Tail flick response[25] as mentioned by wattez *et al* that neuropathy can be tested by these experimental procedures and results in comparision to that of the standard drug show that, *Trigonella foenum graecum* is Neuro protective in diabetic animals.

CONCLUSION

Trigonella foenum graecum have different medicinal properties and may able to treat diabetes & diabetics complications.Subjected to acute oral toxicity studies and found that the *Trigonella foenum graecum* is safe to use up to the dose of 1000mg/kg .The *Trigonella foenum graecum* was found to be in dose dependent way against alloxan induced diabetes in rats. The reduction of the elevated blood glucose levels in diabetic rats on treatment with the extract at two different concentrations confirmed that methanolic extract of *Trigonella foenum graecum* posses Antidiabetic activity & has shown significant effect when compared to Alloxan administration.*Trigonella foenum graecum* had shown protection in neuropathy of diabetes and effective peripheral protection as shown by results. It needs comprehensive investigations for developing a safe and effective drug. Further research is required to confirm the antidiabetic and antidiabetic complications.

REFERENCES

1. Parvizpur, A., Ahmadiani, A., Kamalinejad, M. Spinal serotonergic system is partially involved in antinociception induced by Trigonella foenum-graecum (TFG) leaf extract. *J Ethnopharmacol*. 2004;95(1):13-17.
2. Kaviarasan, S., Vijayalakshmi, K., Anuradha, C. V. Polyphenol-rich extract of fenugreek seeds protect erythrocytes from oxidative damage. *Plant Foods Hum Nutr* 2004;59(4):143-147.
3. Devasena, T. and Menon, V. P. Fenugreek affects the activity of beta-glucuronidase and mucinase in the colon. *Phytother Res*2003;17(9):1088-1091.
4. Raju, J., Patlolla, J. M., Swamy, M. V., Rao, C. V. Diosgenin, a steroid saponin of Trigonella foenum graecum (Fenugreek), inhibits azoxymethane-induced aberrant crypt foci formation in F344 rats and induces apoptosis in HT-29 human colon cancer cells. *Cancer Epidemiol Biomarkers Prev* 2004;13(8):1392-1398.
5. O'Mahony, R., Al Khtheeri, H., Weerasekera, D., Fernando, N., Vaira, D., Holton, J., Basset, C. Bactericidal and anti-adhesive properties of culinary and medicinal plants against Helicobacter pylori. *World J Gastroenterol* 12-21-2005;11(47):7499-7507.

6. Broca, C., Breil, V., Cruciani-Guglielmacci, C., Manteghetti, etal., Insulinotropic agent ID-1101 (4-hydroxyisoleucine) activates insulin signaling in rat. *Am J Physiol Endocrinol Metab* 2004;287(3):E463-E471.
7. Hibasami, H., Moteki, H., Ishikawa, K., Katsuzaki, H., Imai, K., Yoshioka, K., Ishii, Y., Komiya, T. Protodioscin isolated from fenugreek (Trigonella foenumgraecum L.) induces cell death and morphological change indicative of apoptosis in leukemic cell line H-60, but not in gastric cancer cell line KATO III. *Int J Mo Med* 2003;11(1):23-26. 12469212
8. Kaviarasan, S., Ramamurty, N., Gunasekaran, P., Varalakshmi, E., Anuradha, C. V. Fenugreek (Trigonella foenum graecum) seed extract prevents ethanol-induced toxicity and apoptosis in Chang liver cells. *Alcohol Alcohol* 2006;41(3):267-273.
9. Thirunavukkarasu, V., Anuradha, C. V., Viswanathan, P. Protective effect of fenugreek (Trigonella foenum graecum) seeds in experimental ethanol toxicity. *Phytother Res* 2003;17(7):737-743.
10. Raghuram TC, Sharma RD, Sivakumar B, and et al. Effect of fenugreek seeds on intravenous glucose disposition in non-insulin dependent diabetic patients. *Phytotherapy Research* 1994;8(2):83-86.
11. Flammang, A. M., Cifone, M. A., Erexson, G. L., Stankowski, L. F., Jr. Genotoxicity testing of a fenugreek extract. *Food Chem Toxicol*2004;42(11):1769-1775.
12. Parvizpur, A., Ahmadiani, A., Kamalinejad, M. Probable role of spinal purinoceptors in the analgesic effect of Trigonella foenum (TFG) leaves extract. *J Ethnopharmacol* 3-8-2006;104(1-2):108-112.
13. Ajabnoor MA, Tilmisany AK. Effect of trigonella foenum graecum on blood glucose levels in normal and alloxan-diabetic mice. *J Ethnopharm* 1988;22:45-49.
14. Amin R, Abdul-Ghani AS, Suleiman MS. Effect of Trigonella feonum graecum on intestinal absorption. Proc. of the 47th Annual Meeting of the American Diabetes Association (Indianapolis USA). *Diabetes* 1987;36(Supp 1):211a.
15. Kochhar, A. Nagi, M. Effect of supplementation of traditional medicinal plants on blood glucose in non-insulin-dependent diabetics: a pilot study. *J Med Food* 2005;8(4):545-549.
16. Sauvaire, Y., Petit, P., Broca, C., Manteghetti, M., et al., G. 4-Hydroxyisoleucine: a novel amino acid potentiator of insulin secretion. *Diabetes* 1998;47(2):206-210.
17. Stark, A. Madar, Z. The effect of an ethanol extract derived from fenugreek (Trigonella foenum-graecum) on bile acid absorption and cholesterol levels in rats. *Br J Nutr* 1993;69(1):277-287.
18. Petit, P., Sauvaire, Y., Ponsin, G., Manteghetti, M., Fave, A., and Ribes, G. Effects of a fenugreek seed extract on feeding behaviour in the rat: metabolic-endocrine correlates. *Pharmacol Biochem Behav* 1993;45(2):369-374.
19. Al-Habori M, Al-Aghbari AM, Al-Mamary M. Effects of fenugreek seeds and its extracts on plasma lipid profile: a study on rabbits. *Phytother Res* 1998;12(8):572-575.
20. Al-Habori M and Raman A. Antidiabetic and hypocholesterolaemic effects of fenugreek. *Phytother Res* 1998;12(4):233-242.
21. Valette, G., Sauvaire, Y., Baccou, J. C., Ribes, G. Hypocholesterolaemic effect of fenugreek seeds in dogs. *Atherosclerosis*1984;50(1):105-111.
22. Sauvaire, Y., Ribes, G., Baccou, J. C., Loubatieres-Mariani, M. M. Implication of steroid saponins and sapogenins in the hypocholesterolemic effect of fenugreek. *Lipids* 1991;26(3):191-197.
23. Varshney IP and Sharma SC. Saponins sapogenins: part XXXII. Studies on Trigonella foenum-graecum Linn. Seeds. *J Indian Chem Soc*1966;43(8):564-567.
24. Yoshikawa, M., Murakami, T., Komatsu, H., Murakami, N., Yamahara, J., Matsuda, H. Medicinal foodstuffs. IV. Fenugreek seed. (1): structures of trigoneosides Ia, Ib, IIa, IIb, IIIa, and IIIb, new furostanol saponins from the seeds of Indian Trigonella foenum-graecum L *Chem Pharm Bull (Tokyo)* 1997;45(1):81-87.
25. Venkatesan, N., Devaraj, S. N., Devaraj, H. Increased binding of LDL and VLDL to apo B,E receptors of hepatic plasma membrane of rats treated with Fibernat. *Eur J Nutr* 2003;42(5):262-271.
26. Shishodia, S. Aggarwal, B. B. Diosgenin inhibits osteoclastogenesis, invasion, and proliferation through the downregulation of Akt, I kappa B kinase activation and NF-kappa B-regulated gene expression. *Oncogene* 3-9-2006;25(10):1463-1473.
27. Shishodia, S. Aggarwal, B. B. Diosgenin inhibits osteoclastogenesis, invasion, and proliferation through the downregulation of Akt, I kappa B kinase activation and NF-kappa B-regulated gene expression. *Oncogene* 3-9-2006;25(10):1463-1473. 16331273
28. Ruby, B. C., Gaskill, S. E., Slivka, D., Harger, S. G. The addition of fenugreek extract (Trigonella foenum-graecum) to glucose feeding increases muscle glycogen resynthesis after exercise. *Amino Acids* 2005;28(1):71-76. 15719265
29. Yadav, S. K. Sehgal, S. Effect of domestic processing and cooking methods on total, HCl extractable iron and in vitro availability of iron in bathua and fenugreek leaves. *Nutr Health* 2003;17(1):61-63.

30. Singh, U. P., Singh, D. P., Maurya, S., Maheshwari, R., Singh, M., Dubey, R. S., Singh, R.B. Investigation on the phenolics of some spices having pharmacotherapeuthic properties. *J Herb Pharmacother* 2004;4(4):27-42.
31. Hooda, S. Jood, S. Physicochemical, rheological, and organoleptic characteristics of wheat-fenugreek supplemented blends. *Nahrung*2003;47(4):265-268.
32. Aggarwal, B. B., Shishodia, S. Molecular targets of dietary agents for prevention and therapy of cancer. *Biochem Pharmacol* 5-14-2006;71(10):1397-1421.
33. Rai, A., Mohapatra, S. C., Shukla, H. S. Correlates between vegetable consumption and gallbladder cancer. *Eur J Cancer Prev*2006;15(2):134-137.
34. Ribes, G., Sauvaire, Y., Baccou, J. C., Valette, G., Chenon, D., Trimble, E. R., Loubatieres-Mariani, M. M. Effects of fenugreek seeds on endocrine pancreatic secretions in dogs. *Ann Nutr Metab* 1984;28(1):37-43.
35. Ribes, G., Sauvaire, Y., Da Costa, C., Baccou, J. C., Loubatieres-Mariani, M. M. Antidiabetic effects of subfractions from fenugreek seeds in diabetic dogs. *Proc Soc Exp Biol Med* 1986;182(2):159-166.
36. Sidhu, G. S. Oakenfull, D. G. A mechanism for the hypocholesterolaemic activity of saponins. Br J Nutr. 1986; 55(3): 643-649.