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Hypoglycemic Effect of Fenugreek Nanoparticles *In* Alloxan Induced Diabetic Albino Mice (*Mus musculus* L.)

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ABSTRACT: Diabetes mellitus is the most common endocrine disorder that impairs glucose homeostasis resulting in severe diabetic complications. A study was conducted to evaluate the hypoglycemic effect of *fenugreek Nanoparticles* at a dose of 15 mg/kg body weight (BW) once a day to diabetic mice daily for 15 days. Mice were made diabetic by subcutaneous injection of alloxan (150 mg/kg BW) which induced diabetes in albino mice after 5 days. Male albino mice (*Mus musculus* Linn.) of age 3 months and weighing between 28-30 gm were divided in 3 equal groups. Group A served as control, group B was diabetic and was not given fenugreek whereas group C mice were diabetic and given fenugreek nanoparticles. The results showed that the fenugreek nanoparticles dose of 15mg/kg b.wt has significant antihyperglycemic effect in experimental models of diabetes mellitus.

KEYWORDS: Alloxan Diabetes, Blood glucose, *Trigonella foenum graecum*

INTRODUCTION: Diabetes mellitus (DM), one of the major metabolic disorders, is characterized by high blood glucose levels due to the inability of body cells to utilize glucose properly. Diabetes is the world's largest endocrine disease associated with increased morbidity and mortality rate (Edem, 2009) affecting at least 15 million people having complications that include hypertension, atherosclerosis and microcirculatory disorders (Scoppola et al., 2001). India has today become the diabetic capital of the world with over 20 million diabetes and this number is set to increase to 57 million by 2025 (Sridhar et al., 2000). Diabetes mellitus is a multifactorial disease which is characterized by hyperglycemia (Ugochukwu et al., 2003), lipoprotein abnormalities (Scoppola et al., 2001), raised basal metabolic rate (Owu et al., 2006), defect in reactive oxygen species scavenging enzymes (Kesavulu et al., 2000.) A wide number of traditional medicinal plants are still being used to treat diabetes mellitus. Several beneficial role such as correcting altered carbohydrate metabolism, maintaining integrity and function of β-cells, insulin secreting activity, enhancing glucose uptake and utilization and antioxidant properties present in traditional medicinal plant and their constituents offer exciting opportunity to develop them in to novel therapeutics (Rajagopal & Sasikala, 2008).

Many plants reported useful for the treatment of diabetes in the ayurvedic system of medicine. One of most promising amongst them is *Trigonella foenum-graecum* L. commonly

known as methi (Sanskrit), Greek hay fenugreek (English), Kasuri methi consists of dried seeds of the plant. Fenugreek seeds have shown potential as a dietary supplement and scientifically used for wounds, inflammation, and gastrointestinal ailments, cholesterol-lowering agent (Khare, 2004), bronchitis, chronic cough, liver disorders (WHO, 2008), as an antifertility agent (Ahirwar, 2010). *Trigonella foenum graecum* is reported to contain several possible active chemical constituents such as alkaloids, saponins, steroids, tannins, flavonoids, amino acids, and trigonelline (Ahirwar, 2010). The present study was conducted to investigate the antidiabetic activity of fenugreek nanoparticles (*Trigonella foenum-graecum*) on the pancreas in alloxan-induced diabetic mice.

Preparation of fenugreek seed extract:

Fenugreek seeds were collected from the local market of Kolhapur. They were (10g) cleaned and ground into a fine powder using a grinding machine. Ethanol was used for extraction by the Soxhelt extraction method (Lim Cheung 2002). The extract was evaporated to dryness under reduced pressure at 60^0 C by rotary evaporator. Extract was placed in a dark bottle and stored at -8^0C.

Animals: Male albino mice (*Mus Musculus* L.) were used for present study. They were bred and reared in a departmental animal house in separate cages under proper conditions of light, temperature and humidity. They were supplied with Amrut mice feed (Pranav Agro industries) and water *ad libitum* during the experiments. The institutional Animal ethical committee permitted the study.

Induction of diabetes:

Diabetes mellitus was induced by single subcutaneous injection of freshly prepared solution of Alloxan monohydrate (150mg/kg b.wt) dissolved in physiological saline in overnight fasted albino mice. The diabetes was assessed in alloxan induced mice by determining the blood glucose concentration. The mice with blood glucose level above 250mg/dl were selected for the present study.

Experimental design*:* Mice were divided into 3 groups:

1) Control Group: Three months male mice were given a subcutaneous injection of 0.15m acetate buffer pH 5.4 for 15 days.

2*) Diabetic Group:* Three months male mice were given a subcutaneous injection of alloxan 150 mg /kg body weight for 15 days. (Al-Shamaony *et al.*, 1994).

3) Recovery group II: Three months male mice were given a subcutaneous injection of fenugreek nanoparticles at a dose of 15mg/ kg body weight to diabetic mice daily for 15 days.

Measurement of parameters:

1) Blood Glucose:

The blood glucose was measured by collecting a drop of blood from the tail after incision with a sharp blade. The blood glucose level was determined by using a rapid glucose analyzer with a glucose strip inserted in Accu Chek blood glucose monitoring glucometer

(Roche diagnostics India Pvt. Ltd.). The results were expressed in terms of milligram per deciliter of blood (Bopanna et al., 1997).

RESULT: Table.1 displayed a significantly high fasting blood glucose value (226.2222 ± 19.2404) as compared to the control group (113.1111 ± 12.3839) (1:2, P< 0.01). The blood glucose level in the fenugreek seed extract received group was 113.2222 ± 5.0442 which was significantly decreased as compared to alloxan-induced diabetic mice group (2:3, P< 0.01) (Figure. 1).

Sr. No.	Treatment (n=5)	Blood Glucose	Statistical Significance
1	Control	113.1111± 12.3839	1:2, P< 0.01
2	Diabetic	226.2222± 19.2404	2:3, P< 0.01
3	Recovery	113.2222± 5.0442	

Table.1 Fenugreek nanoparticles effect on blood glucose levels (mg/dl) of alloxan-induced diabetic mice. Values are mean ± S.D. (Numbers in parentheses denote the number of animals). P<0.01= Significant

DISCUSSION: Alloxan is a β cell cytotoxin that induces chemical diabetes in a wide variety of animal's species including rats or mice by damaging the insulin secreting β-cells of the pancreas. Alloxan causes time and concentration-dependent degradation lesions of the pancreatic β- cell leading hyperglycemia (Badol et al., 2006). In the present study, alloxan administration resulted in an increase in the blood glucose level indicating hyperglycemia. These observations are similar to those of Dixit *et al.*, 1986, Al- Hader, 1994 and Nagappa *et al.*, 2003 who have used alloxan to induce diabetes in a variety of species. Seeds of fenugreek have multiple benefits in patients with diabetes such as reduction of blood sugar and its complications (Hobori, and Raman 1998; Anuradha and Ravikumar1998; Madar *et al.*, 1988; Rao *et al.*, 1996; Sharma *et al.*, 1996; Basch *et al.*, 2003). The reduction in blood sugar was may be due to 4 hydroxyisoleucine present in fenugreek seeds which show insulinotropic activity (Sauvaire et al., 1978).

The results in present study indicate that fenugreek nanoparticles were found to reduce the glucose level in animals made diabetic with alloxan. Alloxan has been shown to induce free radical production and cause tissue injury. In the present investigation, fenugreek nanoparticles demonstrated significant anti-diabetic activity. Overall results showing the antidiabetic activity of fenugreek seeds, the activity may be due to presence of chemical constituents alkaloids, saponins, steroids, tannins, flavonoids, amino acids, trigonelline (Ahirwar, 2010) in seeds.

CONCLUSION: From this study, we can state that the fenugreek nanoparticles have beneficial effects on blood glucose level as well as improving metabolic aberrations. It has the potential to impart therapeutic effects in diabetes. From this study, we can state that fenugreek nanoparticles have beneficial effects on blood glucose. The present investigation provides a new possibility for synthesis of fenugreek nanoparticles.

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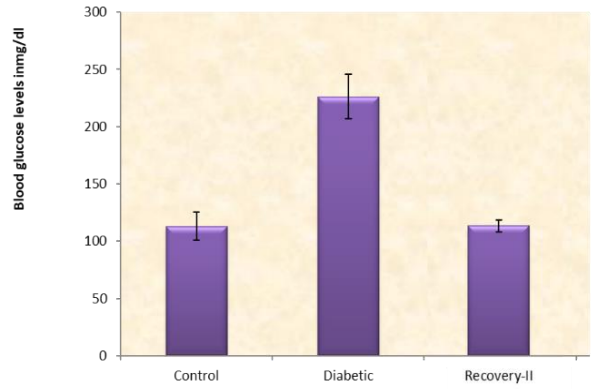

Figure. 1 Fenugreek nanoparticles effect on blood glucose levels (in mg/dl) of alloxan induced diabetic mice