IJPSR (2013), Vol. 4, Issue 12 **(Research Article)**

INTERNATIONAL JOURNAL OF PHARMACEUTICAL SCIENCES AND RESEARCH

Received on 19 July, 2013; received in revised form, 11 September, 2013; accepted, 19 November, 2013; published 01 December, 2013

ANTIDIABETIC AND HYPOLIPIDEMIC EFFECTS OF METHANOLIC EXTRACT OF *FILICIUM DECIPIENS* IN STREPTOZOTOCIN INDUCED DIABETIC RATS

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Keywords:
Filicium decipiens
antihyperlipidemic
,antihyperglycemic and
streptozotocin

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ABSTRACT: This study evaluated methanolic extract of *Filicium decipiens* (MFD) on reducing hyperglycemia and hyperlipidemia in streptozotocin-induced diabetic rats. MFD was orally administered once a day after 3 days of streptozotocin -induction at 100,200 and 400 mg/kg for 45 day and the results showed that serum fasting blood glucose, Glucose – 6 – phosphotase , Fructose 1, 6 –phosphotase in hepatic tissues ,TC, free fatty acids , phospholipids, and TG in serum levels were significantly decreased, whereas Hb and HbCA1in blood and serum HDL, Glucokinase, Glucose – 6 – Phosphate Dehydrogenase in hepatic tissues and liver and muscle glycogen level were increased. The dosage of 400 mg/kg is more effective than that of 100, 200 mg/kg. These results suggest that the MFD possesses antidiabetic and hypolipidemic effects in streptozotocin -induced diabetic rats.

INTRODUCTION: Diabetes mellitus is the most common disease associated with carbohydrate metabolism. The oral therapy of non-insulin-dependent diabetes mellitus (NIDDM) presently relies upon compounds from two chemical classes, sulfonylureas and biguandies [1].

Ethnobotanical information indicates that more than 800 plants are used as traditional remedies for the treatment of diabetes [2 & 3].

The hypoglycemic activity of a large number of these plants has been evaluated and confirmed in different animal models.

QUICK RESPONSE CODE

DOI: 10.13040/IJPSR.0975-8232.4(12).4741-44

Article can be accessed online on: www.ijpsr.com

DOI link: http://dx.doi.org/10.13040/IJPSR.0975-8232.4(12).4741-44

Filicium decipiens (Family: Sapindaceae) is also called fern leaf tree. It is Slender tree 4.5–20(–30) m. tall; bark smooth, whitish. Petiole winged, 1.5–11 cm. Filicium is a genus of three tree species found in Africa and possibly, South India. The name Filicium is form the Greek word for fern. Distinguishable by the 3 habits, leaves that are alternate, pinnately compound and fern like and axillary panicles of inconspicuous fruit and flowers.

It is probably native to south-eastern Africa but was long ago introduced to India, where it is widely cultivated. It is also grown elsewhere in the topics as a shade or street tree. The flowers are seasonal, small and inconspicuous, but the feathery, dark green foli-age is attractive. Dichloromethane, methanol and *n*-butanol fractions of the methanol extracts from the leaves and the stem showed a variety of biological activities, e.g. antifungal, antibacterial and molluscicidal activities [4].

Ethanolic extract of *Filicium decipiens* showed significant anti-inflammatory activity [5]. Sitosterol, kaempferol were identified in n-butanol extract from the methanol extracts from the leaves of *Filicium decipiens* [6]. Four new saponins were isolated from the stem bark of *Filicium decipiens* [7].

MATERIAL AND METHODS:

Collection of Leaves: *Filicium decipiens* is a fern like tree belonging to family Sapindaceae was obtained from Tirupathi in August and authenticated by Dr. Madhavachety (Department of Botany) SV University, Tirupathi).

Preparation of extract: The leaves were shade-dried at room temperature for 10 days, coarsely powdered and the powder was passed through sieve No. 60. The powdered material (300 g) was extracted with methanol (1000 ml) by Soxhlet technique. The extract was then dried under reduced pressure. The dried extract (25.6 g) was stored in desiccators.

Animals: Healthy female albino rats weighing 110-130 g were obtained from King Institute, Chennai. Housed in groupl in polypropylene cages, maintained under standard conditions (12 hrs light and 12 hrs dark cycle; 21± 3^{o}C; 35-60% humidity), the animals were fed with standard rat pellet diet (Sai Durga foods and feeds, Bangalore) and provided water *ad libitum*.

Streptozotocin-induced diabetic rats: Streptozotocin-induced diabetic rats, Streptozotocin (STZ) were purchased from Sigma Aldrich, was dissolved in ice-cold normal saline immediately before use. Diabetes was induced in rats by intra peritoneal (i.p) injection of streptozotocin at a dose of 45 mg/kg, dissolved in normal saline. 3 days after streptozotocin administration, the blood samples were drawn from the tail of the rats and glucose levels determined to confirm the presence of diabetes.

The diabetic rats exhibiting blood glucose levels higher than 300 mg/dl were selected for the studies. These diabetic rats were divided into five groups as follows. Group I, Normal control group received food and water. Group II Untreated (Diabetic control) received 0.5 ml of 5% Tween 80. Group III, Group IV and Group V received 100,200 and 400 mg/kg of MFD, respectively.

The treatment was continued daily for 45 days. Blood was collected from the tail for glucose estimation just before drug administration on the first day and 1 h after sample administration on day 45. The animals were killed after blood collection on day 45. Blood samples were collected and centrifuged to separate serum for estimation of diabetic and lipid marker. Glucose, Total cholesterol, Free fatty, acids, Phospholipids, HDL, LDL, VLDL, and triglycerides were analyzed from serum. Hb and HbAC1 were determined using blood sample. Glucokinase, Glucose-6-Phosphate Dehydrogenase, Glucose-6-phosphotase, Fructose 1, 6-phosphotase in hepatic tissues and glucogen in liver and muscle were estimated.

Statistical analysis: The results were expressed as mean ± S.E.M. Statistical comparisons were made by means of One-way ANOVA and the results were considered statistically significant when $p < 0.05$.

RESULT: The hyperglycaemic animals showed significant decrease in the glucose level on long-term treatment for the 45-day model at the doses of 100, 200 and 400 mg/kg of methanolic extract of FD **(Table** 1). Body weight slightly increased in the normal control rats, compared to initial body weight. Extract treated rats; there was a significant increase in the body weight on the 45th day compared to untreated diabetic rats (Table 1). The effect of MFD on glucose levels in streptozotocin-induced diabetic rats is shown in Table 1. MFD (100, 200 and 400 mg/kg) treated rats, the blood glucose levels at three doses steadily decreased and were found to be 192,160 and 107 mg/100 ml, respectively, and Hb level was significantly increased and HbAC1 level was significantly decreased in methanolic extract of FD compared to untreated diabetic rats on the 45th day.

The Glucokinase, Glucose-6-Phosphate Dehydro-genase and liver, muscle glycogen was significantly decreased in untreated diabetic rats compared to control group. After the treatment with methanolic extract Glucokinase, Glucose-6-Phosphate Dehydrogenase and liver, muscle glycogen was significantly increased in treated diabetic rats compared to untreated diabetic rats. Glucose-6-phosphotase and Fructose 1, 6-phosphotase were significantly decreased compared to untreated diabetic groups (**Table 2**).

The serum TG, total cholesterol, free fatty acids, phospholipids, LDL and VLDL levels were significantly higher in untreated diabetic rats compared to those in normal rats, while the HDL levels were significantly decreased in the diabetic rats compared to those in normal rats.

After treatment with MFD in diabetic rats, a significant reduction in serum level of cholesterol, free fatty acids, phospholipids, TG, LDL, VLDL and a significant increase in HDL was observed (**Table 3**).

TABLE 1: EFFECT OF MFD ON BODY WEIGHT AND DIABETIC MARKERS OF CONTROL AND EXPERIMENTAL RATS

Groups	Control	STZ 50mg/kg	STZ+100mg of MFD	STZ+200mg of MFD	STZ+400mg of MFD
			Body weight		
0 day	182.00±7.43	185.00±7.43	183.00±6.29	184.00±7.27	183.00±7.43
45 day	203.00±8.25^a	138.00±6.19^b	152.00±6.26^c	170.00±7.90^d	187.00±4.46^e
			Blood glucose		
0 day	89.01±4.08	253.34±10.70	260.62±9.69	258.80±10.07	255.87±9.95
45 day	91.66±3.73^a	280.77±11.57^b	192.20±5.67^c	160.56±4.81^d	107.10±4.46^e
			Diabetic markers		
Insulin	16.82±0.74^a	6.09±0.23^b	8.63±0.26^c	9.95±0.86^d	12.62±0.49^e
Hb	14.33±0.58^a	8.98±0.33^b	9.89±0.66^c	10.78±0.63^d	11.06±0.45^e
HbA_1C	0.48±0.02^b	1.87±0.08^b	0.90±0.04^c	0.70±0.04^d	0.56±0.02^e

All the values are expressed as mean ± SD of 6 rats in each group.

TABLE 2: EFFECT OF MFD ON CARBOHYDRATE METABOLIC ENZYME ACTIVITY AND GLYCOGEN LEVELS OF CONTROL AND EXPERIMENTAL RATS

Groups	Control	STZ 50mg/kg	STZ+100mg of MFD	STZ+200mg of MFD	STZ+400mg of MFD
			Carbohydrate metabolic enzyme		
Glucokinase	0.36±0.01^a	0.17±0.01^b	0.19±0.01^c	0.22±0.02^d	0.31±0.01^e
Glucose – 6 – Phosphate Dehydrogenase	5.08±0.23^a	2.66±0.08^b	3.02±0.12^c	3.80±0.15^d	4.44±0.19^e
Glucose – 6 – phosphotase	0.24±0.01^a	0.48±0.02^b	0.38±0.01^c	0.32±0.02^d	0.27±0.01^c
Fructose 1, 6 –phosphotase	39.04±1.41^a	72.04±2.89^b	60.04±1.03^c	55.90±2.53^d	48.93±2.03^c
			Glycogen		
Liver	50.09±2.11^a	22.85±0.84^b	34.86±1.11^c	40.80±1.23^d	46.03±1.86^c
Muscle	6.12±0.57^a	2.78±0.20^b	3.40±0.63^b	4.90±0.69^c	5.90±0.51^a

All the values are expressed as mean ± SD of 6 rats in each group.

TABLE 3: EFFECT OF MFD ON LIPID PROFILE OF CONTROL AND EXPERIMENTAL RATS

Groups	Control	STZ	STZ+100mg of MFD	STZ+200mg of MFD	STZ+400mg of MFD
Total cholesterol	81.93±3.32^a	159.92±6.53^b	140.60±5.63^c	120.68±6.84^d	98.83±4.01^e
Triglycerides	58.12±2.08^a	129.85±5.33^b	110.90±5.93^c	80.46±6.93^d	65.92±2.52^e
Free fatty acids	60.90±2.88^a	121.81±5.09^b	100.68±6.82^c	89.65±0.63^d	68.93±2.57^e
Phospholipids	84.13±2.94^a	151.11±5.83^b	130.87±4.70^c	110.78±7.84^d	91.01±3.71^e
HDL	47.12±1.70^a	24.50±0.88^b	32.40±0.75^c	38.68±0.69^d	46.25±1.75^a
LDL	23.19±0.96^a	109.45±4.21^b	60.98±6.84^c	52.34±8.61^d	39.40±1.25^e
VLDL	11.62±0.41^a	25.97±0.87^b	20.68±0.56^c	15.86±0.48^d	13.18±0.50^e

All the values are expressed as mean ± SD of 6 rats in each group.

DISCUSSION: Antihyperglycemic effect of the MFD in diabetic rats has been indicated here by the study of fasting blood glucose levels as the important basal parameter for monitoring of diabetes and the dosage of 400 mg/kg is more effective than that of 100 and 200mg/kg. There was a significant weight loss in the diabetic rats, and the treatment with MFD in the treated diabetic group resulted in an improvement in their body weights. The ability of the MFD to protect the body weight loss seems to be due to its antidiabetic activity. The treatment with MFD has also enhanced the rate of glycogenesis as indicated by higher amounts of hepatic glycogen in the diabetic treated group. Similar observations, i.e. hypoglycaemic activity and improved levels of

hepatic glycogen, were reported. Diabetes is also associated with hyperlipidemia. The levels of TC and TG have been decreased significantly in diabetic mice after the MFD administration. These effects may be due to low activity of cholesterol biosynthesis enzymes and or low level of lipolysis which are under the control of insulin.

The MFD also results to the significant attenuation in the level of serum HDL toward the control level which again strengthens the hypolipidemic effect of this extract. There are reports that other medicinal plants have hypoglycemic and hypolipidemic effects that could prevent or be helpful in reducing the complications of lipid profile seen in some cases of diabetes in which hyperglycemia and hypercholesterolemia coexist [8].

CONCLUSION: In conclusion, the data obtained from the present study indicates that MFD may have both antihyperglycemic and antihyper-lipedemic activity in STZ diabetic rats. In future we will be carried out the isolation of active principle from MFD and elucidate the mechanism of antihyperglycemic and antihyperlipedemic effect of the isolated compound from MFD.

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How to cite this article:
Sasikala C. and Sudhakar Y. : Antidiabetic and hypolipidemic effects of methanolic extract of *Filicium decipiens* in streptozotocin induced diabetic rats. *Int J Pharm Sci Res* 2013; 4(12): 4741-44. doi: 10.13040/IJPSR. 0975-