WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 7.523
Volume 6, Issue 14, 806-816. Research Article ISSN 2277–7105

ANTIDIABETIC AND HYPOLIPIDAMIC EFFECTS OF *COSTUS SPICATUS* JACQ. RHIZOME EXTRACT AGAINST STREPTOZOTOCIN INDUCED DIABETIC ALBINO RATS

S. Azhagumadhavan[1], Arjun Pandian[2], P. Vinayaga Moorthi[3] and Ganesan Sivamani[1]*

[1]PG & Research Department of Zoology & Biotechnology, A.V.V.M. Sri Pushpam College (Autonomous) Poondi, Thanjavur – 613503, Tamil Nadu, India.

[2]Department of Biotechnology, PRIST University, Thanjavur-613 403, Tamil Nadu, India.

[3]Department of Human Genetics and Molecular Biology, Bharathiar University, Coimbatore-641 046, Tamil Nadu, India.

Article Received on
11 Sep. 2017,
Revised on 01 October 2017,
Accepted on 22 October 2017
DOI: 10.20959/wjpr201714-9980

***Corresponding Author**
Ganesan Sivamani
PG & Research Department of Zoology & Biotechnology, A.V.V.M. Sri Pushpam College (Autonomous) Poondi, Thanjavur – 613503, Tamil Nadu, India.

ABSTRACT

The present study of antidiabetic effects of rhizome extracts of *C. spicatus* on STZ in diabetic rats was evaluated, important mechanism; underlying hyperglycemia in diabetes mellitus. Excess of glucose in blood leads diabetes. It reacts with haemoglobin and forms glycosylated. Therefore, the total haemoglobin level is decreased in STZ diabetic rats. Administration of *C. spicatus* ethanolic rhizome extract reversed the total haemoglobin levels in STZ diabetic rats. Our study we observed that haemoglobin level is lowered in diabetic treated rats. After twenty eight days treatment ethanolic rhizomes extract showed important reduction of blood glucose level. It showed effectively maintain blood glucose level in normal and STZ induced diabetic rats. Different group of animals were used; ethanolic extract of *C. spicatus* treated groups (200 and 300 mg/kg BW) another treated groups glibenclamide, used as standard, results in rats showed a significantly increased insulin level when compared with diabetagenic rats treated groups as well as in the group of glibenclamide.

KEYWORDS: Blood glucose, Diabetic, Ethanolic extract, Glibenclamide, Insulin level.

INTRODUCTION

Diabetes mellitus (DM) is a group of metabolic disorders in hyperglycemia owing to decreased insulin production or inefficient insulin insensitivity of target organs to insulin utilization. It's a common and very widespread disease affecting developed and developing countries. The World Health Organization (WHO) predicted it DM affects approximately 171 million people worldwide and in 2030[1] these number will be reach up to 366 million. In diabetes, hyperglycemia generates reactive oxygen species (ROS) in which causes lipid peroxidation, membrane damage and plays an important role in production of secondary complications in DM such as kidney, eye, blood vessel and nerve damages[2-3] insulin resistance and to stimulate insulin secretion. Diabetes is a metabolic disorder in humans, it didn't produce or properly us insulin, a hormone that is required to convert sugar levels, starches and other food into energy. It's characterized through constant high levels of blood glucose (BG) in human body sensitivity (sugar). In human body blood glucose levels (BGLs) very narrow range done with insulin and glucagon. Glucagon causes liver to release glucose from its cells into the blood for energy production. Type 1 diabetes leads to inability to release insulin results in low rates of glucose uptake into muscles and adipose tissue.[4] The antihyperglycemic activity of plants is mainly due to its ability or motility to restore function of pancreatic tissues through causing an increase insulin output of intestinal absorption of glucose or facilitation of metabolites in insulin dependent processes. Secondary metabolites; glycosides, alkaloids, terpenoids, flavonoids, carotenoids from plants are frequently implicated in having antidiabetic effects.[5] Hyperglycemia in DM, but these drugs are dangerous condition, alternative strategies for current modern pharmacotherapy of DM.[6] In conventional medical practice, the present therapies of DM reported have side effects. Many oral therapeutic agents are primary alternative treatments for type 2 DM.[7-8] Traditional system of Indian medicinal plants formulation and in several cases, combined extracts of plants are used as drug of choice rather than individual, many of these have shown promising effects.[9] *C. spicatus* belongs to Costaceae family, is a spiral, wild ginger. It is an important source of diosogenin and is used in many human and veterinary medicines. It is bitter, astringent, cooling, digestive, stimulant and good for the heart.[10] Based on the medicinal properties and literature plant *C. spicatus* selected for present study evaluate antidiabetic and hypolipidamic activity of *C. spicatus* in STZ induced diabetic rats.

MATERIAL AND METHODS

Animals

The present study, albino wistar male rats; 7–8 weeks old, body weight (BW) 190-220 g, were used. Animals were housed under standard conditions temperature (24±2°C) and relative humidity (30-70%) with a 12:12 (light:dark) conditions. The animals were fed with standard pellet diet. Animal handling was performed according to Good Laboratory Practice (GLP). Ethical clearance was obtained from Institutional Animal Ethics Committee and conducted according to the Indian National Science Academy guidelines for the use and care of the experimental animals.

Chemicals

Streptozotocin (STZ), Ethylene Diamine Tetra Acetic Acid (EDTA), Glibenclamide, Chloroform purchased from Prudence Pharma Chem, India.

Plant materials

Fresh plant material of *Costus spicatus* area collected from the Saliyamangalam, Thanjavur District, Tamil Nadu, India.

Plant sample extraction

The rhizome were cut into small pieces and shade dried at room temperature. The dried rhizome were subjected to size reduction to a coarse powder by using dry grinder and passed through sieve, dried rhizome powdered, rhizome powder (100 g) were continuously extracted with ethanol (95%) using Soxhlet extractor extracted up to 48 h. The extract was filtered through Whatmann filter paper then it's concentrated by using rotary evaporator at 40-60ºC under reduced pressure finally prepared crude extract.

Induction of Diabetes in Rats

After fasting, diabetes was induced by intra peritoneal, injection of streptozotocin dissolved in cold sodium citrate (0.1 M; pH 4.5) buffer, dose level 55 mg/kg.[11] The control rats received the vehicle alone. The animals are allowed to drink glucose solution (5%) over night to overcome the drug induced hypoglycemia. After a week time for the development of diabetes, the rats are having glycosuria and hyperglycemia (blood glucose range 250 mg/dl) was it's considered as diabetic rats and used for the experimental works.

Experimental Design

The animals were divided into six groups follows; control (group I) were injected with citrate buffer alone. Group II served as diabetagenic rats (control). Group III; *C. spicatus* ethanolic extract (200 mg/kg BW), IV; *C. spicatus* ethanolic extract (300 mg/kg BW) were treated orally once in a day up to 4 weeks. Group V; glibenclamide dose level (5 mg/kg BW) and served as stranded treatment continued (4 weeks). Each animal was marked for identification and regularly monitoring, per group six animals used for all the experiments.

Collection of samples

After treatments animals were anesthetized using ketamine chloride (24 mg/kg BW) and sacrificed by cervical dislocation after an overnight fast. Blood was collected with and without EDTA. Plasma and serum were separated after centrifugation it's used for various biochemical studies.

Different biochemical studies

Serum glucose estimated by oxidase method.[12] Total cholesterol was estimated through described method.[13] Triglyceride was estimated.[14] HDL cholesterol was separated by adding phosphotungsti magnesium chloride in fresh samples to precipitate other lipoproteins and HDL cholesterol was quantified.[14] The concentration of LDL cholesterol was calculated by using the Friedewald formula and VLDL cholesterol was calculated by dividing the triglycerides values (5 mg/dl). Heamoglobin estimated and insulin was assayed through solid phase system amplified sensitivity immunoassay.

Histological Assay

On 28th day, pancreatic tissues were taken from animals which were fasted over night under ether anesthesia. The whole pancreas from each animal was removed after killing the animals, was placed in formulation (10%) solution and immediately processed by the paraffin technique section thickness (5 µm) were cut and strained by haematoxylin and eosin (H and E) for histological examination. The photomicrographs of histological studies noted.

Statistical Analysis

All results are presented as mean ± SEM Data were analyzed by the student's T test. Groups for the pair of observations depended upon each other. Results were considered statistically at $P < 0.001$.

RESULTS AND DISCUSSION

Estimation of Body Weight

The BW changes in control and experimental groups were described (Table 1 and Fig. 1). BW in diabetic rats compared to control group, in treated it's decreased significantly. Supplementation of ethanolic extract showed significant improvement in BW of diabetic rats. There were no significant changes observed between control treated groups.

Estimation of Blood glucose, Plasma insulin and Hemoglobulin

The blood glucose plasma insulin and total hemoglobulin levels in normal and experimental rats, it showed (Table 2 and Fig. 2). There was asignificant increased level of blood glucose and plasma insulin was observed in diabetes animals compared othe control. Ethanolic extract restored levels of blood glucose and plasma insulin of diabetic group of rats and its effect was more pronounced in the group of rats.

Estimation of Serum Lipid Profile

The ethanolic rhizomes extracts of *C. spicatus*, administration in diabetic rats serum lipids levels. The triglycerides, total cholesterol, VLDL, LDL increased and HDL levels were significantly decreased in STZ treated rats (Table 3 and Fig. 3). Oral administration of ethanolic extract ranged (200 and 300 mg/kg BW) restored the altered parameters, which was compared to that of glibenclamide group. However, no significant changes were observed control treated groups.

Histological Assay

Multiple section of pancreas were taken and studied for histological changes in the plant treated and control group (Fig. 4). Results observed that histological findings of pancreas tentatively similar. STZ induced diabetic rats showed extensive damage on the islets of langer hanscells (Fig. 4-b). The orally administered ethanolic extract (200 and 300 mg/kg BW) and commercial drug (Fig. 4-c, d), Glibenclamide (5 mg/kg BW) (Fig. 4-e) were showed restoration of normal cellular population and enlarged size of β-cells with hyperplasia found in islets of langer hans cells in pancreas.

The pancreas present in the group of animals treated with the ethanolic extract (300 mg/kg BW) clearly showed that the partial restoration of normal cellular population and enlarged size of β-cells with hyperplasia on 30^{th} day. The islets were normal in size, shape and number comparatively similar to that of standard treated drug.

The present investigates antidiabetic effect of rhizomes extract of *C. spicatus* on STZ diabetic rats. The fundamental mechanism underlying hyperglycemia in DM. During diabetes the excess of blood glucose reacts with haemoglobin to form glycosylated haemoglobin. Therefore, the total haemoglobin level is decreased in STZ treated rats. So the total haemoglobin level is lowered in diabetic rats. Administration of *C. spicatus* reversed the total haemoglobin levels in STZ diabetic rats decreased.

Twenty eight days administration of ethanolic extract *C. spicatus* resulted in significant reduction in fasting BGL compared to diabetic rats. The differences observed between initial and final fasting levels of different groups revealed a significant elevation in BG in diabetic control group compared to normal. It is evident from these investigations rhizomes extract is effectively maintaining the BGLs in normal and STZ induced diabetic rats. Ethanolic extract of *C. spicatus* treated groups III, IV and solution of *C. spicatus* treated groups V rats showed a significantly increase insulin level when compared with group II as well as group V. STZ, a,β-cytotoxin, induces chemical diabetes in a wide variety of animal species by damaging the insulin-secreting β–cells of pancreas.

The STZ causes time and concentration dependent degenerative lesions of pancreatic β-cells. The lipid profiles in control and experimental rats are depicted in STZ-induced diabetic rats; there was a significant increase of total cholesterol, triglycerides and low-density lipoprotein (LDL) and very low-density lipoprotein (VLDL) cholesterol and decrease high density lipoprotein (HDL) cholesterol in serum compared with normal control. The plant extracts used in the study significantly decreased the levels of cholesterol, triglycerides, phospholipids, and LDL and VLDL cholesterol and increase HDL cholesterol. This indicates that the rhizomes extract had favorable effects, on lipid metabolism of diabetic rats. Derangement of glucose, fat, and protein metabolism in diabetes results in the development of Hyperlipidemia.[15,16]

Table 1: Effect of *C. spicatus* ethanolic rhizome extract on the changes of body weight (BW) of control and treated rats.

Groups	BW (g)	
	Initial (0 day)	Final (4 weeks)
Control	198.22± 19.10	228.21 ± 19.01
Diabetic control	187.13 ± 18.11*	149.11 ± 11.4*
Diabetic + ethanolic extract(200 mg/kg BW)	182.04 ± 12.2	180.11±12.21*
Diabetic+ ethanolic extract(300 mg/kg BW)	197.06 ± 19.43*	228.02±11.41**
Glibenclamide (5 mg/Kg)	202 ± 11.25**	225 ±12.35**

Values are given as mean ± S.D (n=6 rats).

*P<0.01 Vs control, **P<0.001Vs control by students 'T' test.

Table 2: Effect of *C. spicatus* ethanolic rhizomes extract on the levels of blood glucose, plasma insulin and hemoglobin in control and experimental rats

Groups	Blood glucsoe (mg/dL)	Plasma insulin (µg/mL)	Total Hemoglobin (g/dL)
Control	85.45 ± 8.81*	7.24 ± 1.54	13.43 ± 1.35*
Diabetic control	280.11± 19.33	4.24 ±0.13	8.93 ± 0.49
Diabetic + ethanolic extract (200 mg/kg BW)	108.12±4.57*	11.02±1.21*	12.87± 0.45*
Diabetic + ethanolic extract(300mg/kg BW)	90.01±3.12*	17.21±1.05*	13.99±1.58**
Glibenclamide (5 mg/kg)	84.21 ±6.23	15.12±2.43 **	13.88±3.12 **

Values are given as mean ± S.D (n=6)

* P<0.01 Vs control, **P<0.001Vs control by students 'T' test.

Table 3: Effect of *C. spicatus* ethanolic rhizome extract on lipid profile in control and experimental rats.

Treatment	TGL (mg/dl)	HDL-C (mg/dl)	VLDL-C (mg/dl)	LDL-C (mg/dl)	Total Cholesterol (mg/dl)
Control	75.15±5.03	38.6±1.83	15.03±1.06	41.9±4.37	96.16±6.44
Diabetic control	124.32±6.5	42.56±5.52	32.14±1.7	96.47±3.2	215.2±7.4
Diabetic + ethanolicextract (200 mg/kg BW)	97.52±4.67*	38.27±2.56	30.19±2.8*	88.41±2.34	172.3±5.*
Diabetic+ ethanolicextract (300mg/kg BW)	91.49±2.45**	33.46±1.36*	25.52±3.4*	81.27±5.28*	152.6±6.9*
Glibenclamide standard (5 mg/kg)	82.04±6.7*	25.38±4.75*	16.92±1.34*	33.9±2.66*	96.2±4.8*

Values are given as mean ± S.D (n=6)

* P<0.01 Vs control, **P<0.001Vs control by students 'T' test.

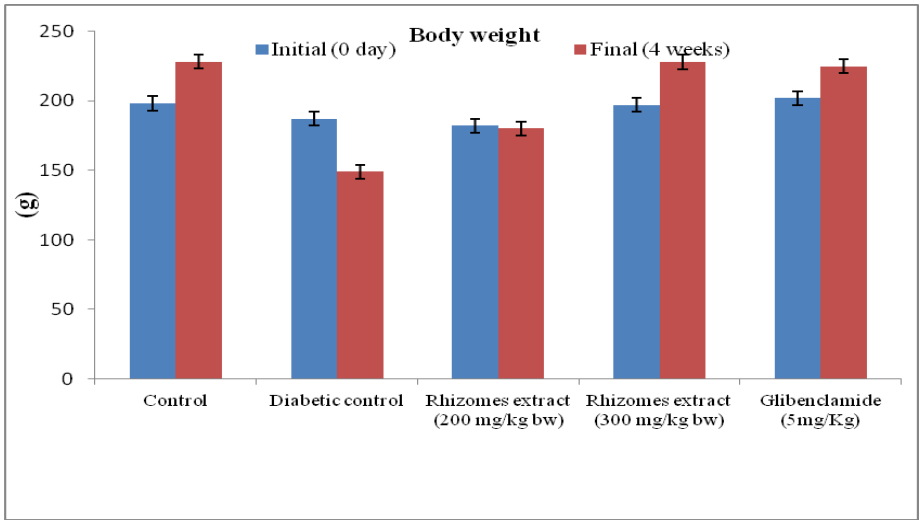


Figure 1: Effect of *Costus spicatus* ethanolic rhizomes extract on the changes of body weight of control and experimental rats.

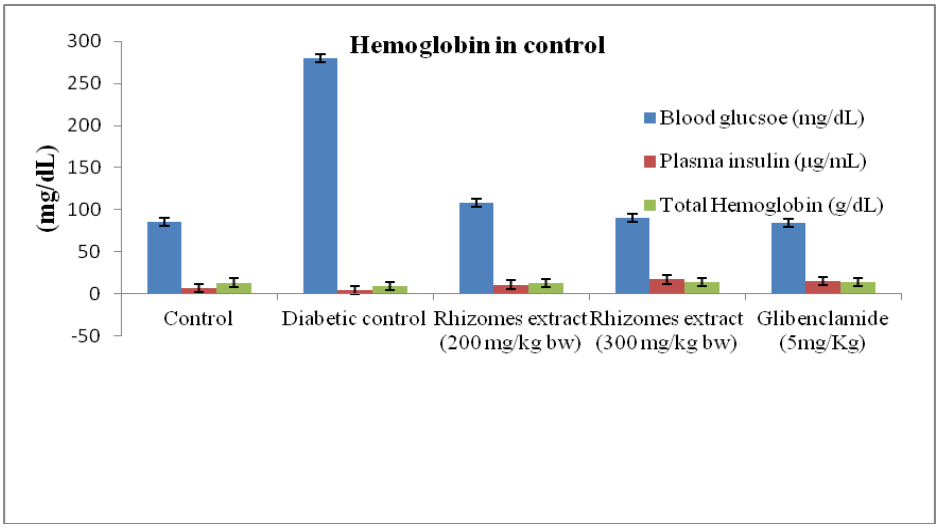


Figure 2: Effect of *Costus spicatus* ethanolic rhizomes extract on the levels of blood glucose, plasma insulin and Hemoglobin in control and experimental rats.

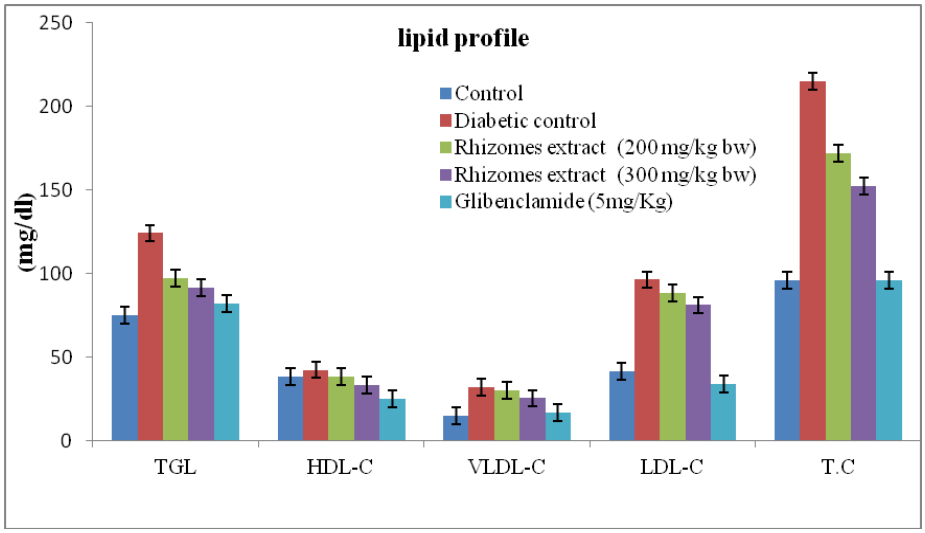
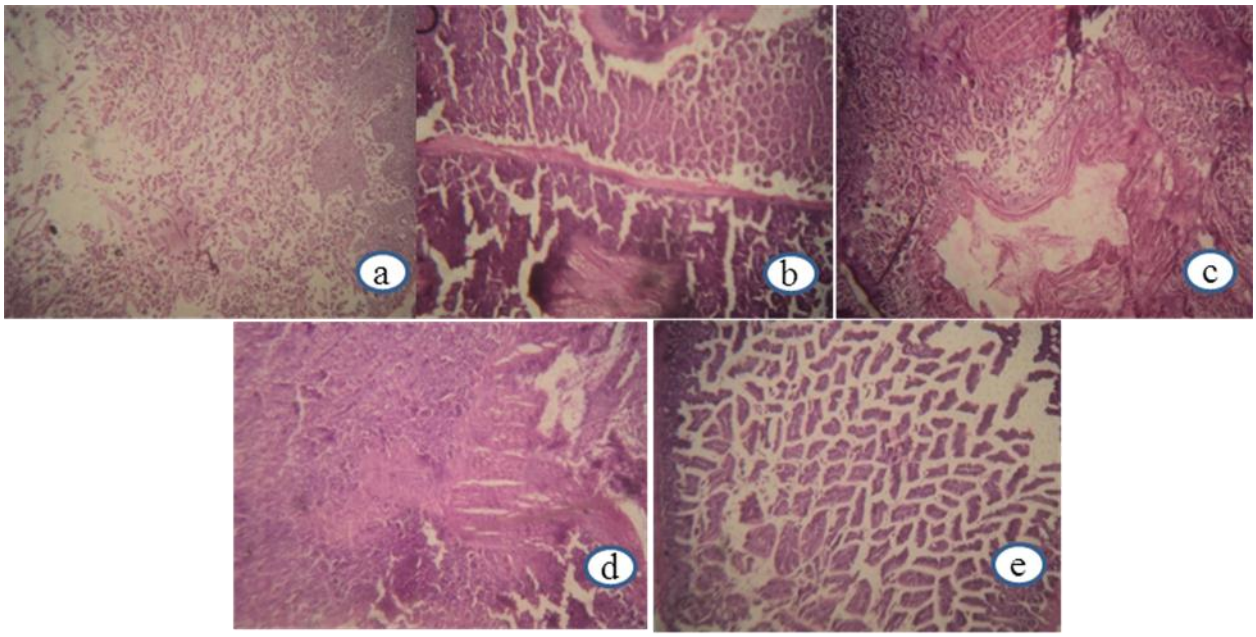


Figure 3: Effect of *Costus spicatus* ethanolic rhizomes extract on lipid profile in control and experimental rats.



a. Histopathology of Islets of Langerhans normal Rat (Control) .
b. Histopathology of Islets of Langerhans of diabetic Rat (STZ-Induced)
c. Histopathology of Islets of Langerhans of treated with *C. spicatus* (200 mg/kg) diabetic rats.
d. Histopathology of Islets of Langerhans of treated with *C. spicatus* (300 mg/kg) diabetic rats.
e. Histopathology of Islets of Langerhans of diabetic rats treated with Gibenclamide (5 mg/kg).

Figure 4: Antidiabetic and hypolipidamic effects of rhizome extracts of *Costus spicatus.*

CONCLUSION

The present study suggests that the *C. spicatus* rhizomes extracts had synergetic hypoglycemic effect revealed by decreased serum lipid levels restored heamoglobin and therefore attribute to therapeutic values combat the diabetic condition in rats. Among the two

doses extract showed potential antidiabetic activity. Based on secondary metabolites; flavonoids, terpenoids etc. Hence, it might help in preventing diabetic complications and serves as a good adjuvant in the present armamentarium of antidiabetic drugs.

REFERENCE

1. World Health Organization; http://www.who.int/features/factfiles/diabetes/en/. 2010.
2. Aslan M, Orhan N, Orhan DD, Ergun F. Hypoglycemic activity and antioxidant potential of some medicinal plants traditionally used in Turkey for diabetes. *J Ethnopharmacol*, 2010; 128: 384–389.
3. Fenercioglu AK, Saler T, Genc E, Sabuncu H, Altuntas Y. The effects of polyphenol-containing antioxidants on oxidative stress and lipid peroxidation in Type 2 diabetes mellitus without complications. *J Endocrinol Invest*, 2010; 33: 118–124.
4. Welihinda J, Arvidson G, Gylfe E, Hellman B, Karlsson E. The insulin-releasing activity of the tropical plant *momordica charantia*. *Acta Biol Met Ger*, 1982; 41: 1229.
5. Malviya N, Jain S, Malviya S. "Antidiabetic potential of medicinal plants," *Acta Poloniae Pharmaceutica*, 2010; 67: 113–118.
6. Rao BK, Sudarshan PR, Rajsekher MD, Nagarju N, Rao CA. Antidiabetic activity of *Terminalia pallida* fruit inalloxan induced diabetic rats. *J. Ethnopharmacol*, 2003; 85: 169–172.
7. Wright Jr. E, Scism-Bacon JL, Glass LC. Oxidative stress in type2 diabetes: The role of fasting and postprandial glycaemia. *International J Clinical Practice*, 2006; 60: 308–314.
8. Ceriello A, Quagliaro L, Catone B, et al. Role of hyperglycemia innitrotyrosine postprandial generation. *Diabetes Care*, 2002; 25: 1439–1443.
9. Pari L, Saravanan G. Antidiabetic effect of cogent db, a herbal drug in alloxan induced diabetes mellitus. *Comparative Bioche and Physiology Part C*, 2002; 131: 19–25.
10. Pitchai D, James E, Ignacimuthu S. Influence of *Costus speciosus* (Koen.) Sm. Rhizome extracts on biochemical parameters in streptozotocin induced diabetic rats. *J Health science*, 2008; 54: 675–681.
11. Kakkar P, Das B, Visvanathan PN. A modified spectrophotometric assay of superoxide dismutase. *Indian J Biophys*, 1984; 21: 130–132.
12. Sasaki T, Matsy S, Sorae A. Effect of acetic acid concentration on the colour reaction in the O-toluidine boric acid method for blood glucose estimation. *Rinsho Kagarku*, 1972; 1: 346–353.

13. Allian CC, Poon LS, Chan CSG, Richm W, Fu P. Determination of total cholesterol by enzymatic method. *Clinical Chemistry*, 1974; 20: 470–475.
14. Fossat Lorenzo P. Serum triglycerides determined calorimetrically with an enzyme that produce hydrogenperoxide. *Clin. Chem*, 1982; 28: 2077–2080.
15. Freidmann N. Hormonal regulation of hepatic gluconeogenesis. *Physiol. Rev*, 1984; 64: 170–176.
16. Kanivalan N, Rajakumar R, Mani P. antidiabetic and Hypolipidamic activity of *Costus spicatus. Acta Biomedica Scientia*, 2014; 1(2): 74–79.