Preventive Effect of Anogeissus Latifolia in High Fructose Diet Induced Insulin Resistance and Metabolic Dyslipidemia

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Abstract:

Anogeissus latifolia (Combretaceae) has been used in traditional system of medicine in India for over many years, and widely used to enhance the immune system and against various ailments. During the last decade many in-vitro, in-vivo and analytical studies witnessed and justified the traditional claim of this plant. The study was aimed to evaluate the preventive effect of Anogeissus latifolia bark in insulin resistance and metabolic dyslipidemia induced by high fructose diet feeding in rats. At the end of the treatment period, the plasma from normal control, diabetic, reference standard and Anogeissus latifolia extract (300mg/kg) treated animals were subjected to estimation of insulin, lipid profile, liver marker enzymes and kidney function test. Anogeissus latifolia was found to attenuate insulin resistance and metabolic dyslipidemia induced by adverse effects of fructose. This study validates the traditional use and shows a possible beneficial effect of the plant in the treatment of diabetes mellitus.

Key Words: Anogeissus latifolia, High Fructose Diet, Insulin Resistance, Metabolic Disorder.

1.   Introduction

Type-2 diabetic condition is prevalent worldwide and is associated with morbidity and mortality, which are secondary to complications such as myocardial infraction, stroke and end stage renal disease (Se Na Yun et al, 2004). Nutritional or dietary oxidative stress denotes a disturbance of the redox state resulting from excess oxidative load or from inadequate nutrient supply favoring prooxidant reactions. Postprandial increases of lipid and carbohydrate levels lead to increased oxidative stress and lipid hydroperoxides present in the diet are absorbed, contributing to the prooxidant load (Helmut et al, 2005). Low intake or impaired availability of dietary antioxidants weakens the antioxidant network. Postprandial oxidative stress, as a sub form of nutritional oxidative stress, ensues from sustained postprandial hyperlipidemia and/or hyperglycemia and is associated with a higher risk for atherosclerosis, diabetes, and obesity (Tsai et al, 2004). Moreover, although data are scarcer, the fact that fructose may increase intrahepatic lipids and leads to insulin resistance in experimental settings raises some concern. In animal models, the potential danger of fructose consumption and its links to various metabolic disorders have been widely documented (Luc Tappy et al, 2009). Deleterious effects of high fructose intake on body weight, insulin sensitivity/glucose homeostasis, dyslipidemia, and atherosclerotic disease has been identified, and potential mechanisms have been proposed (Luc Tappy et al, 2009). High fructose consumption is suspected to be causally linked to the epidemics of obesity and metabolic disorders. A study by Nagai et al has focused a particular gene, known as PGC-1 beta, appears to play a key role in the development of insulin resistance in response to a high-fructose diet (Nagai et al, 2009).   Currently available pharmacological agents for type-2 diabetes have a number of limitations associated with high rate of secondary failure (Inzucchi et al, 2002). In response to the enormity of the growing problem, efforts to identify and develop new pharmacological agents for type-2 diabetes medicinal herbs with antihyperglycemic and antihyperlipidemic activities are increasingly sought by diabetic patients and health care professionals. Commonly used herbs and other alternative therapies, less likely to have the side effects of conventional approaches for type-2 diabetes, are reviewed. These efforts have resulted in the successful screening of medicinal herbs with antihyperglycemic activity (Lucy Deg et al, 2002). To date, over 400 traditional plant treatments for diabetes have been reported (Bailey et al, 1989). The hypoglycemic effect of some herbal extracts has been confirmed in human and animal models of type 2 diabetes. The World Health Organization Expert Committee on diabetes has recommended that traditional medicinal herbs to be further investigated (Bailey et al, 1989). The current study was designed to investigate the antihyperlipidemic effect of *Anogeissus latifolia* bark in model of hypertriglyceridemia and insulin resistance induced by high fructose diet, which is considered as a good model for type-2 diabetes as it displays many of the characteristics of the human diseases including hyperglycemia, insulin resistance and progressive obesity (Hummel et al, 1966).

Anogeissus latifolia, (A.latifolia) (Combretaceae), locally known as Dhava, is a moderate sized tree characteristic of dry deciduous forests and available throughout India. In phytotherapy, the bark, leaves, heartwood and roots of the plant is traditionally used for the treatment of dysentery, snakebite, leprosy, wounds and ulcers, skin diseases, jaundice and diabetes (Anonymous 1987). The bark is reported to have potent antioxidant activity (Govindarajan et al 2004a) and possess several biological activities like antiulcer,

antimicrobial (Govindarajan et al 2006), wound healing (Govindarajan et al 2004b), chemo protective (Khan et al,2008), anthelmentic (Hemamalini et al,2011), nephroprotective (Veenavamshee et al,2011) and hepatoprotective (Pradeep et al,2009). The bark contains tannin and phenolic compounds (Gallic acid, ellagic acid, chebulic acid), flavonoids (rutin and quercetin)(Reddy et al,1965,Deshpande et al,1976), triterpenoids and phytosterol (β-sitosterol) (Mohammad et al,2007, Uday Sing et al,2011) which might contribute to various pharmacological activities. These constituents are reported to possess potential antioxidant activity and offer protection against various diseases (Aruoma et al, 1993, Festa et al, 2001, Boyle et al,2000, Boots et al,2008, Francisco et al,2009). Beneficial effects of *A.latifolia* and its gum exudates (commonly known as Gum gatti) have been evidenced in experimental diabetic animals in Streptozotocin- induced (Type-1) diabetic model (Parvathi et al,2009a) and in atherogenic diet induced hyperlipidemic model (Parvathi et al,2009b) respectively. These pharmacological evidences and rich antioxidant phytoconstituents of *A.latifolia* promoted us to explore the antihyperglycemic activity of *A. latifolia* bark extract in high fructose diet induced model of the metabolic syndrome in rats.

2.   Materials and Methods
2.1 Composition of High Fructose Diet
High fructose diet consisted of 60 % fructose. The 60 % fructose concentration was chosen because this amount produces hippocampal insulin resistance in hamsters (Mielkej al al, 2005), leads to peripheral pathology in rats similar to the pathology associated with fructose consumption in humans (Elliotl et al, 2002, Montonenj et al,2007), and is the amount used most extensively in rodent studies (Demoura et al,2008, Suga et al,2000). The control group was fed a diet of standard rat chow consist of 60 % vegetable starch *ad libitum*. Both diets contained equal percentages of carbohydrates (65 %), proteins (20 %), and lipids (10 %), and both diets were also isocaloric on a weight basis (kcal/gm) (Anonymous 1977). The control group received standard pellet chow for 6 weeks.

Composition	High Fructose Diet g/kg (kcal)
Fructose	600 (2208)
Casein	200(800)
Corn oil	100(900)
DL Methionine	3(12)
Choline bitartrate	2(0)
AIN-76 Mineral mix	35(0)
AIN-76A vitamin mix	10(40)
Starch	50(0)

2.2 Plant Material
Bark of *A.latifolia* was collected from Nallamala *forest*, Mahaboobnagar district, Andhra Pradesh, India, during the month of May. It was authenticated from Osmania University, Hyderabad.

2.3 Extraction of Plant Material
The bark was shade dried and powdered coarsely. The coarse powder obtained was extracted exhaustively with 70% ethanol in Soxhlet apparatus and filtered. The extract was concentrated under reduced temperature and pressure to get dry residue and stored in a desiccators.

2.4 Animals and Treatment Schedule
Thirty male *Wister* rats, 6 weeks old and weighing roughly above 150 g, were procured from Mahaveer Enterprises and were housed in polypropylene cages in a temperature-controlled room ($25^{o}\pm2^{o}$C) with a 12 h light/12 h dark cycle. All rats were adapted and fed a standard pellet chow diet for 1 week. Rats had unrestricted access to food and water. Food intakes were monitored daily, and body weights were measured weekly. All rats were randomly divided into four groups (n = 6 each): Group I, normal control group: fed with a standard diet with normal saline daily using an intra gastric tube, Group II, diabetic control group: fed with HFD with normal saline daily using   an intragastric tube , Group III *A. latifolia* treated group: fed with HFD and *A. latifolia* (300 mg/kg body weight) in aqueous solution treated daily using an intra-gastric tube**,** and Group IV Rosiglitazone-treated group: fed with HFD and rosiglitazone (60 µg/kg body weight) in aqueous solution treated daily using an intra-gastric tube for 6 weeks. The study protocol was approved by the Institutional Animal Ethics Committee (NIP/1330/AC/10/CPCSEA Dated 11th March), Nizam Institute of Pharmacy, (Hyderabad, Andhra Pradesh, India) and the rats were maintained in accordance with the National Institute of Nutrition, Indian Council of Medical Research, Hyderabad, India guidelines for the care and use of laboratory animals.

2.5 Oral Glucose Tolerance Test (OGTT)
OGTTs were performed at weekly intervals during the experimental period. The rats were food deprived for 12 h prior to the administration of an oral glucose load (2 g/kg body weight, 200 g/ l solution). Blood samples were drawn from the tail vein at 0, 30, 60, and 90 min after glucose feeding. Glucose levels were determined using an Accu-Check Advantage Blood Glucose Monitor (Roche Group, Indianapolis, IN, USA).

2.6 Blood and Tissue Collection

At the end of study, blood samples were collected in a heparinized tube by puncturing the orbital venous plexus of 12-h fasted and anesthetized rats. Whole blood samples were centrifuged at 4500 *rpm* for 10 min at 4°C and plasma was separated out and stored at −70°C until further analysis. All the animals were sacrificed by cervical dislocation, liver, pancreas and   epididymal fat pad were removed and rinsed with chilled phosphate buffered saline (pH 7.0) and weighed. After immediate weighing, the liver tissues were chopped into small pieces and stored at −20°C until further use.

2.7 Blood and Liver Tissue

2.7.1 Lipid Profile: Plasma total-cholesterol, Triglycerides (TG), and LDL cholesterol (LDL-C) were estimated by using enzymatic kits procured from E-Merck.

2.7.2 Liver and Kidney Function Tests: The liver function tests (aspertate transaminase (AST), alanine transaminase (ALT), plasma Albumin/globulin ratio (A/G ratio), and total bilirubin) and kidney function tests (plasma urea and creatinine) were estimated using the E-Merck kits.

2.7.3 Estimation of Plasma Insulin, Liver Glycogen Content and HMG CoA Reductase Activity

The plasma insulin was measured with an Autoanalyser (Auto lab 200) according to the manufacturer's instructions (Merck,).

Liver glycogen content was determined using the method reported by Van der Vies et al, 1954. Liver tissue (200 mg) was finely ground with 20 ml of 5% TCA in a homogenizer and protein precipitate was filtered. Clear filtrate (2 ml) was pipette out into a 20-ml calibrated test tube and then 10 N KOH (2 ml) was added. This tube was placed in a boiling water bath for 1 h. After cooling, 1 ml of glacial acetic acid was added to neutralize the excess of alkali and fluid brought up to the mark with water. Slowly, 2 ml of solution from the previous step was added to a test tube containing 4 ml of anthrone reagent, which was placed in cold water to prevent excessive heating. After thorough mixing, the tube was placed in a boiling water bath for exactly 10 min for the development of colour and cooled with running tap water. The optical density was read within 2 h in a spectrophotometer (SPECORD 200; Analyjetika, Germany) at 650 nm against a blank that was prepared by subjecting 2 ml of 5% TCA instead of sample to the same procedure. Serum was used for the estimation of HMG CoA reductase activity. Activity of HMG CoA reductase was determined using the method of Rao and Ramakrishnan et al, 1975   by determining the ratio of HMG CoA to mevalonate.

3. Statistical Analysis

Data are expressed as the mean ± SEM. The results of the study were subjected to analysis of variance (ANOVA) using graph pad prism followed by Dunnett's t-test for multiple comparisions. P value <0.05 was considered statistically significant.

4. Results

4.1 Oral Glucose Tolerance Test (OGTT)

The results of glucose intolerance induced by high-fructose diet and comparative delaying effect of *A.latifolia* as well as rosiglitazone in rats during 6 weeks of the experimental period. In Glucose Tolerance Test, the Glucose levels were estimated before drug treatment and at different intervals thereafter. In the control group the blood glucose was found to increase linearly from basal value of 101.4 mg/dl to 191.8 mg/dl in the first 30 minutes. After 60 minutes of glucose loading, the blood glucose was increased further. The maximum value of 312 mg/dl was seen at the 90th minute. Whereas in the extract treated animals, only a little elevation in the blood glucose were seen from basal value of 97.4 mg/dl to 113.8 mg/dl in the first 30 minutes, 78.2 mg/dl in the 90 minutes and 89.4mg/dl at 150 minutes and maximum glucose tolerance was observed at 90th minute. In rosiglitazone treated animals blood glucose rises to 106 mg/dl from basal value of 93.3 mg/dl in the first 30 minute, 72.4 mg/dl in 90 minutes and 79.6 mg/dl at 150 minutes and maximum glucose tolerance was observed at 90th minute.

4.2 Effect on Body, liver and Pancreas Weight

Table 1 shows the values of body weight, weight of liver, pancreatic tissue and epididymal fat of normal and diabetic animals and the comparative effect of *A.latifolia* as well as rosiglitazone in HFD-fed rats. The body weight and epididymal fat of the diabetic control animals was significant higher than those of all the other groups; however, significant decrease was observed in body weight and epididymal fat content in *A.latifolia* and rosiglitazone-treated animals in comparison to the normal animals. The weights of liver and pancreatic tissue among all the groups were nearly similar.

4.3 Effect on Hyperglycemia, Plasma Insulin, Liver Glycogen and HMG-CoA Reductase Activity

Administration of *A.latifolia* showed a significant antihyperglycemic activity in HFD-fed rats similar to standard rosiglitazone. The results in Table 2 and 3 show that blood glucose level, plasma insulin and liver glycogen in the diabetic control group were significant higher than those in the normal group (P<0.05). The levels of blood glucose level, plasma insulin and liver glycogen content were significantly lower in the *A.latifolia* treated animals than those of the diabetic control group (P<0.05) Similarly, rosiglitazone- treated animals also showed significantly lower values of blood glucose, plasma insulin and liver glycogen content as

compared with the diabetic control group of animals (P<0.05). Diabetic rats exhibited a highly significant increase in liver HMG-CoA reductase activity as compared with the non-diabetic group. Administration of both *A.latifolia* and reference standard led to marked amelioration of HMG-CoA reductase activity.

4.4 Effect on Plasma Lipid Levels

The changes in plasma lipid levels in the diabetic control, A.latifolia treated and rosiglitazone treated groups are summarized in Table 4. It demonstrates that the plasma total cholesterol, TG and LDL-C were significantly higher in the diabetic control animals than that in the normal animals. However, these variables were significantly lower in the *A.latifolia* treated rats than diabetic control rats (P<0.05). However, these values were almost similar to those of rosiglitazone-treated animals (P<0.05). In rosiglitazone-treated rats, plasma total cholesterol, TG and LDL-C, were also lower than those of diabetic control animals (P<0.05).

4.5 Effect on Liver and Kidney Function Tests

HFD fed rats showed significant increase in AST, ALT and Total bilirubin level and decrease in albumin/globulin ratio compared to rats fed standard diet indicating liver dysfunction. The oral administration of *A.latifolia* as well as rosiglitazone showed significant improvement in the liver function tests as compared to the parameters in diabetic control animals. However there was no significant variability observed in the kidney function tests of HFD, standard fed, A.latifolia and rosiglitazone treated animals (Table 5).

5. Discussion

Fructose is a potent reducing sugar that promotes the formation of toxic advanced glycation end-products, which appear to play a role in the aging process; in the pathogenesis of the vascular, renal, and ocular complications of diabetes; and in the development of atherosclerosis. In addition, excessive fructose consumption may be responsible in part for the increasing prevalence of obesity, diabetes mellitus, and non-alcoholic fatty liver disease characterized by an impaired glucose tolerance test. With a few exceptions, the relatively small amounts of fructose that occur naturally in fruits and vegetables are unlikely to have deleterious effects (Alan et al,2005). Study results revealed that treatment of diabetic rats with *A. latifolia* lead to significant amelioration of glucose tolerance. A decrease in elevated glucose levels is in agreement with the results of Vessal *et al*, 2003 and Kamalakkannan *et al*, 2006 who demonstrated similar effects of flavonoid, rutin in diabetic rats and also with those reported by Nuraliev et al, 1992 on a hypoglycemic effect of quercetin in diabetic animals. Thus, the hypoglycemic effect of *A. latifolia* may be due to the presence of flavonoids such as rutin and quercetin present in bark extract. Administration of *A.latifolia* and rosiglitazone delayed the appearance of an impaired glucose tolerance test. This beneficial effect shown for glucose absorption is postulated to be attributed to the activity of specific constituents, flavonoids in the *A. latifolia*, such as quercetin, which is recognized as an inhibitor of glucose absorption (Kwon et al, 2007). In addition, the other antioxidant constituents (Gallic acid, ellagic acid, sterols) of the *A. latifolia* may have also played a role in its effects. The results of present study showed that administration of *A.latifolia* to rats for six weeks inhibited the disturbance in glucose metabolism in the liver by reducing the accumulation of glycogen in liver, which might be due to induced glycogenolysis and/or inhibited gluconeogenesis.

Obese and type-2 diabetes are characterized by adipose tissue over production of pro inflammatory cytokines. The reduced body and epididymal fat weight in *A.latifolia* supplemented rats is probably due to reduced portal collagen deposition and fat deposition in the liver. Fructose-induced insulin resistant states are commonly characterized by a profound metabolic dyslipidemia, which appears to result from hepatic and intestinal overproduction of atherogenic lipoprotein particles. A high flux of fructose to the liver, perturbs glucose metabolism and glucose uptake pathways, and leads to a significantly enhanced rate of lipogenesis and triglyceride synthesis, resulting in high flux of glycerol and acyl portions of triglyceride molecules from fructose catabolism. These metabolic disturbances appear to underlie the induction of insulin resistance commonly observed with high fructose feeding in both humans and animal models (Heathe et al, 2005). The ability of *A.latifolia* to reduce plasma cholesterol and triglycerides in diabetic animals could be explained by the insulin releasing capacity of flavonoid constituents (Quercetin & Rutin) present in the bark extract (Hii et al, 1985). The hypolipidemic activity of *A.latifolia* may also be mediated, at least in part, *via* inactivation of hepatic HMG-CoA reductase, a key enzyme, in cholesterol synthesis. The observed activity may be due to flavonoids, rutin, quercetin of the extract which are reported to possess HMG-CoA reductase inhibiting activity (Bok et al, 2002, Osama et al, 2010). In concurrence with this attribution, Raz *et al*, 2005 state that inhibitors of hepatic HMG-CoA reductase are well established drugs for the treatment of hypercholesterolemia and decrease the incidence of dyslipidemia in diabetic subjects. This also coincides well with the work of Jung *et al*, 2006 who state that flavonoids decrease liver HMG-CoA reductase activity in type 2 diabetic mice. Moreover, rutin has been reported to lower hepatic and blood cholesterol levels, as stated by Park *et al*, 2002. It can be concluded that the ameliorative effect of *A.latifolia* extract on serum lipid variables may be attributed to their insulin releasing capacity and insulin binding affinity and decreasing intestinal cholesterol absorption and activity of hepatic HMG-CoA reductase. Hepatic damage in fructose-fed rats was evident from the increased plasma

transaminase (Hult et al, 1986), as well as the decline in hepatic function from the elevated bilirubin level and the decreased A/G ratio. Increased aspartate transaminase (AST) activity is considered as markers of increased metabolic activity (Lottja et al, 1980). This biochemical changes reflect hepatocellular damage in fructose-fed rats. Fructose is known to induce renal hypertrophy and tubulointerstitial disease in rats (Nakayama et al, 2010, Kang et al, 1977). Reduction in AST activity therefore suggests decreased metabolic activity in *A.latifolia* administered rats. Moreover drug being Hepatoprotective may also have a role in attenuating hepatic changes brought about by high fructose feeding. The kidney function tests were screened based on the reports of Nakayama *et al*, 2010 and Kang *et al*, 1977. However there was no significant variability observed in the kidney function tests in HFD feeding, may be due to the shorter duration of the experiment. Further works on these aspects are expected to reveal the reason for discrepancies. Above results demonstrates the regenerative antioxidant effect of *Anogeissus latifolia* during progression of diabetes induced by metabolic changes in fructose fed rats. Although the antioxidant and free radical scavenging properties of *A.latifolia* and its constituents have been previously described (Govindarajan et al,2004a), this is the first study to our knowledge that demonstrates the preventive role of *A.latifolia* in high fructose diet induced dyslipidemia in rats. Phenolic compounds along with flavonoids as both insulin potentiating factors and antioxidants are postulated to act in preventing the metabolic syndrome, which is characterized by insulin resistance, dyslipidemia, and increased oxidative stress.

6. Conclusion: The findings of the present study are of merit in revealing that *A.latifolia* possesses the potential to reverse the dyslipidemia caused by metabolic changes induced by high fructose diet in rats. This study validates the traditional use and shows a possible beneficial effect of the plant in the treatment of diabetes mellitus.

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Table 1: Effect of *A.latifolia* on Body, Liver and Pancreas Weight.

Parameters	Normal	HFD Control	*A. latifolia*-Treated	Rosiglitazone-Treated
Initial Body Weight (g)	178.60±5.7	180.19±3.32	180.10±3.9	189.75±5.1
Body weight gain (g)	13.30±4.5	15.38±5.0 [a]	11.89±10.11[a]	11.38±4.98[a]
Liver weight (g)	7.49±2.56	7.11±2.10	7.54±2.0	7.38±3.4
Pancreas	0.375±0.04	0.38±0.01	0.350±0.02	0.371±0.02
Epididymal Fat (mg/100g)	368.80 ± 67.20	451.20 ±58.29[abc]	378.30 ± 44.38[a]	387.20 ± 37.38[a]

The values are means ± S.E.M. of 6 animals in each group during and/ or after 48 days of the experimental period. Values quoted with a, in comparison to normal control group (P<0.05); b, in comparison to the *A. latifolia*-treated group (P<0.05); and c, in comparison to the rosiglitazone-treated group are significantly different (P<0.05).

Table 2: Effect of *A. Latifolia* on Blood Glucose Levels (mg/dl) on High Fructose Diet Induced Diabetic Rats after Repeated Doses.

Groups	Day 1	Day 12	Day 24	Day 36	Day 48
Normal	71.66±1.30	73.4±2.0	77.83±1.72	83.16±1.75	82±1.06
HFD Control	282.50±1.76	278±1.18	286±1.42	294.14±1.49	302.27±1.52
A.latifolia-Treated	307.83±1.84	138.16±2.29**[a]	114.83±2.16**[a]	110±2.25**[a]	96±2.23**[a]
Rosiglitazone-Treated	291.68±1.04[b]	82.33±3.56**[b]	84.03±1.36**[b]	74.74±1.05**[b]	72.17±2.2**[b]

Significance compared within the groups as follows: **a**. A. latifolia treated rats Vs. diabetic control rats.
b. Rosiglitazone treated rats vs. diabetic control rats. Values are given as mean ± SEM for groups of six animals in each group. Values are statistically significant at *p<0.05 and **p<0.01.

Table 3: Effect of *A.latifolia* and Rosiglitazone Treatment on Plasma Insulin, Liver Glycogen Content and HMG-CoA Reductase Activity after 6 weeks of Drug Treatment.

Parameters	Normal	HFD Control	*A.latifolia* Treated	Rosiglitazone-Treated
Plasma Insulin (U/ml)	238.3± 11.20	465.2 ± 24.12^a	321.8 ± 18.58^{abc}	309.9 ± 30.17^{ab}
Liver Glycogen Content (mg/ g of liver tissue)	6.91 ± 1.85	11.05 ± 1.58^a	8.55 ± 2.0^{abc}	7 .58 ± 3.0^{ab}
HMG-CoA Reductase	1.13± 0.53	1.43± 0.74	1.32± 0.74^{abc}	1.26± 0.02^{abc}

The values are means ± S.E.M. of 6 animals in each group and significantly different (P<0.05): a, in comparison to the normal group; b, in comparison to the diabetic control group; and c, in comparison to the rosiglitazone-treated group.

Table 4: Effect of *A. latifolia* and Rosiglitazone Treatment on Plasma Lipid Levels after 6- weeks of Drug Treatment.

Parameters	Normal	HFD Control	*A.latifolia*-Treated	Rosiglitazone-Treated
Total Cholesterol(mg/dl)	72.18 ± 7.09	118.30 ± 16.19^a	97.38 ± 7.32^{abc}	93.93 ± 10.2^{ab}
Triglyceride (mg/dl)	59.20 ± 4.33	86.15 ± 2.89^a	73.29 ± 8.00^{abc}	68.39 ± 3.98^{ab}
LDL-Cholesterol(mg/dl)	40.11 ± 3.81	70 .06 ± 12.0^a	51.0 ± 3.53^{abc}	48.91 ± 8.30^a

The values are means ± S.E.M. of 6 animals in each group and significantly different (P<0.05): a, in comparison to the normal group; b, in comparison to the diabetic control group; and c, in comparison to the rosiglitazone-treated group.

Table 5: Effect of *A.latifolia* and Rosiglitazone Treatment on Liver and Kidney Function Tests after 6 Weeks.

Parameters	Normal	HFD Control	*A.latifolia*-Treated	Rosiglitazone-Treated
Liver Tests				
AST(IU/L)	75.6±4.4	168.3±6.8^a	118.7±1.1^{abc}	127. 5±5.6^b
ALT(IU/L)	72.1±3.4	150.4±8.1^a	109.4±4.5^{abc}	119.6±5.3^b
A/G Ratio (mg/dl)	1.5±1.1	0.8±0.04^a	1.2±4.9^{abc}	1.3±3.2^b
Total bilirubin(mg/dl)	0.5±0.04	1.02±0.06^a	0.9±0.03^{abc}	0.7±0.05^b
Kidney Function Tests Urea (mg/dl)	14.92 ± 3.25	15.20 ± 4.0	15.30 ± 4.51	14.76 ± 4.56
Creatinine(mg/dl)	0.84 ± 0.11	0.91 ± 0.18	0.92 ± 0.11	0.88 ± 0.08

The values are means ± S.E.M. of 6 animals in each group and significantly different (P<0.05): a, in comparison to the normal group; b, in comparison to the diabetic control group; and c, in comparison to the rosiglitazone-treated group.