Antihyperglycemic Effect of Mangiferin in Streptozotocin Induced Diabetic Rats

Periyar Selvam Sellamuthu,*,[a] Balu Periamallipatti Muniappan,[a] Sathiya Moorthi Perumal,[b] and Murugesan Kandasamy[a]

[a]*Lab. No. 102, Centre for Advanced Studies in Botany, University of Madras, Chennai-600025, Tamilnadu, India and* [b]*Department of Industrial Biotechnology, Dr. M.G.R. Educational and Research Institute, Dr. M.G.R. University, Chennai-600095, Tamilnadu, India.*

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The study was undertaken to evaluate the antihyperglycemic potential of mangiferin purified from methanolic root extract of *Salacia chinensis* (*S. chinensis*) in control and streptozotocin (STZ)-induced diabetic rats. The mangiferin was administered orally at a dose of 40 mg/kg weight per day (30 days) to STZ-induced diabetic rats. The mangiferin treated diabetic rats significantly decreased the level of blood glucose, glycosylated hemoglobin as well as increased level of insulin and hemoglobin. The activities of hexokinase, pyruvate kinase, glucose-6-phosphate dehydrogenase, glycogen synthase, and glycogen content level were increased to near normal in mangiferin treated diabetic rats. The activities of lactate dehydrogenase, glucose-6-phosphatase, fructose-1,6-diphosphatase and glycogen phosphorylase were significantly decreased in liver tissue of diabetic treated rats. These findings demonstrated that mangiferin possess antidiabetic activity against STZ-induced diabetic rats. The antidiabetic effect of mangiferin was compared with standard antidiabetic drug glibenclamide.

Key words —— mangiferin, *Salacia chinensis*, streptozotocin, diabetes, glibenclamide

INTRODUCTION

Diabetes mellitus is a metabolic disorder, characterized by chronic hyperglycaemia with disturbance of carbohydrate, fat and protein metabolism resulting from defects in insulin secretion, insulin action or both [World Health Organization (WHO), 1999]. Diabetes affected around 171 million people worldwide in 2000 and it may be increase to at least 366 million by 2030.[1)]

In modern medicine, there is no satisfactory effective therapy to cure diabetes mellitus. The management of diabetes mellitus by insulin therapy, have several drawbacks like insulin resistance[2)] and in chronic treatment cause anorexia nervosa, brain atrophy, and fatty liver.[3)] The oral hypoglycaemic drugs are sulphonylureas and biguanide groups have been used in the treatment of diabetes mellitus. The sulfonylureas (*e.g.*, glibenclamide, glipizide) stimulate the insulin secretion from the existing pancreatic β cells. Glibenclamide inhibits the adenosine tri phosphate (ATP) sensitive K^+ (K_{ATP}) channels in the plasma membrane.[4)] This leads to membrane depolarization, activation of voltage gated Ca^{2+} channels, a rise in cytosolic (Ca^{2+}), and release of the insulin. The streptozotocin (STZ)-induced diabetes is treated by gilbenclamide and used as a standard drug to compare the antidiabetic activity of various compounds.[5)]

Ethnobotanical information indicates that more than 800 plants are used as traditional remedies for the treatment of diabetes,[6)] but many plants do not have a scientific scruting. Herbal medicines are used for primary health care, by about 80% of the world population particularly in the developing countries, because of better cultural acceptability, safety, efficacy, potent, inexpensive, and lesser side effects.[7)] The plant drugs are frequently considered to be less toxic when compared to synthetic drugs.[8)] More than 1123 plant species have been used to treat diabetes and more than 200 pure compounds have showed, lowering blood glucose activity.[9)]

The WHO Expert Committee recommended that the important to investigate the hypoglycemic agents from plant origin, which were used in traditional medicine for the treatment of diabetes melli-

*To whom correspondence should be addressed: Lab. No. 102, Centre for Advanced Studies in Botany, University of Madras, Chennai-600025, Tamilnadu, India. Tel.: +91-9444-821490; Fax: +91-44-22202756; E-mail: periyar. india@gmail.com

tus.[10] The antihyperglycemic agents have been focused on plants used in traditional medicine because that may be a better treatment than currently used synthetic drugs.[11]

Mangiferin is a xanthone glucoside and an active phytochemical present in common principal constituent of *Salacia* species.[12] *Salacia chinensis* Linn. (*S. chinensis*), a member of the family Hippocrateaceae. The roots are used in indigenous system of medicine and treated for diabetic, astringent, abortifacient, amenorrhoea, dysmenorrhoea, and veneral diseases. Mangiferin is called as C-glucosyl xanthone[13] and 2-β-D-glucopyranosyl-1, 3,6,7-tetrahydroxy xanthone.[14] Mangiferin is recommended to treat immuno-deficiency diseases such as diabetes, hepatitis, arthritis, cardiac, and mental disorders.[15]

The three new friedelane-type triterpenes named salasones A, B, and C, a new norfriedelane-type triterpene, salaquinone A, and a new acylated eudesmane-type sesquiterpene, salasol A, were isolated from the 80% aqueous methanolic extract of the stems of *S. chinensis* collected in Thailand. Their stereo structures were elucidated on the basis of chemical and physicochemical evidence. In addition, six constituents, 3β, 22β-dihydroxyolean-12-en-29-oic acid, tingenone, tingenine B, regeol A, triptocalline A, and mangiferin, were found to show an inhibitory effect on rat lens aldose reductase.[16]

Hence, the aim of our study was to investigate the antihyperglycemic effect as well as non-toxic nature of mangiferin purified compound from the methanolic root extract of *S. chinensis* in STZ-induced diabetic rats. The results were compared with standard antidiabetic drug, glibenclamide.

MATERIALS AND METHODS

Chemicals —— The analytical graded chemicals were used for all the experiments. The STZ was purchased from Sigma Chemicals, St. Louis, MO, U.S.A.

Plant Material —— The roots of *S. chinensis* were collected from Veenangaputtu, Karumpakkam, Thangal and Kurumpuram (all areas are nearest to Pondicherry, India). The plant voucher specimen (778) has been deposited in Centre for Advanced Studies in Botany, University of Madras, Chennai, India.

Preparation of Plant Extract —— The roots of *S. chinensis* were washed thoroughly with tap water, shade dried, cut into small pieces, and were crushed to moderately coarse powder. It was extracted using 95% methanol in soxhlet apparatus for 6 hr. The extract was concentrated by using rotary evaporator at 45–50°C under reduced pressure. The methanolic root extract yield was about 23.5%.

Purification of Mangiferin by Column Chromatography —— A portion of the crude methanolic root extract was subjected to column chromatography over slicagel with chloroform gradient elution using ethyl acetate in methanol 95 : 5–15 : 85 respectively for the total amount of 17 elutes (100 ml) were collected and combined in to basis of their Thin Layer Chromatography (TLC) similarities. The mangiferin yield was eluted on the fraction ratio of 40 : 60 (ethyl acetate : methanol), which was confirmed by TLC analysis. The concentrated fraction was subjected to recolumn chromatography, while purity was confirmed by using high performance liquid chromatography analysis. The yield of mangiferin was found to be 7.8%.

Purity Analysis of Mangiferin —— High Performance Liquid Chromatography was performed to confirm the purity of the mangiferin[17] and C18 column was used to separate the mangiferin. The mobile phase of an isocratic consisting of acetonitrile and 3% of acetic acid (16 : 84) was used with a flow rate of 0.5 ml/min and the UV-visible detector wavelength was set at 254 nm. The authentic mangiferin was purchased from Sigma Aldrich Company (St. Louis, MO, U.S.A.), when compared with authentic sample; the isolated mangiferin was closely resembled with authentic purity that was found to be 99.4% (w/w).

Experimental Animal —— Male Wistar albino rats (150–200 g) were procured from Tamil Nadu Veterinary and Animal Sciences University, Chennai, India and were housed in polycarbonate cages in an animal room with 12 hr day-night cycle at temperature of 22 ± 2°C and humidity of 45–60%. They were fed with commercial pelleted rats chow and free access water during the experiment.

The experiments were designed and conducted in accordance with the ethical norms approved by Ministry of Social Justice and Empowerment, Government of India and Institutional Animal Ethics Committee Guidelines (IAEC No.02/004/06) for the investigation of experimental pain in conscious animals.

Induction of Experimental Diabetes —— Diabetes was induced by administering intraperitonial injection of a freshly prepared solution of STZ

(55 mg/kg b.w.) in 0.1 M cold citrate buffer (pH 4.5) to the over night fasted rats.[18] Because of the instability of STZ in aqueous media, the solution is made using cold citrate buffer (pH 4.5) immediately before administration. Control rats were injected with citrate buffer alone. The rats were allowed to drink 5% glucose solution overnight to overcome the drug-induced hypoglycemia. The blood glucose values above 250 mg/dl on the third day after STZ injection, were considered as diabetic rats. Then the treatment was started on the fifth day after STZ injection and it was considered as first day of treatment.

Experimental Design —— The rats were divided into four groups with six animals for each group.

Group 1: Control rats

Group 2: Diabetic control rats

Group 3: Diabetic rats treated with mangiferin (40 mg/kg b.w. per day) in aqueous solution orally for 30 days[19]

Group 4: Diabetic rats treated with glibenclamide (600 μg/kg b.w. per day) in aqueous solution orally for 30 days

At the end of the treatment (30 days), the animals were deprived of food overnight and anesthetized and sacrificed by cervical dislocation. Blood was collected in heparinised tubes and used for the estimation of hemoglobin and glycosylated hemoglobin.

The liver tissue was excised and rinsed in ice-cold physiological saline. The tissue was homogenized by tissue homogenizer with a Teflon pestle at a 4°C in 0.1 M Tris-HCl buffer at a pH 7.4. The supernatant was kept in −20°C for further use. The homogenate (liver) was centrifuged in a cooling centrifuge ($500 \times g$) to remove the debris and supernatant was used for the biochemical analysis. The total proteins were estimated by Lowery *et al.*[20] method.

Toxicity Study —— The mangiferin was orally administrated at a concentration of 25, 50, 100, 250 and 500 mg/kg of body weight/day for a period of 30 days. The toxic effect of mangiferin was measured by body weight gain and morphological changes.

Glucose Tolerance Test (GTT) —— Diabetic rats were administered with mangiferin up to 30 days and after treatment a fasting blood sample was taken from all groups. After 30 min of the drug administration all the rats were orally administered with glucose solution (2 g/kg). The blood samples were collected at 30, 60, 90 and 120 min intervals after the administration of glucose.[21] The blood samples were collected with potassium oxalate and sodium fluoride solution for the estimation of glucose.

Estimation of Blood Glucose, Hemoglobin, Glycosylated Hemoglobin, and Plasma Insulin —— The blood glucose level was estimated by the method of O-toludine by Sasaki *et al.*[22] Hemoglobin was estimated by Drabkin and Austin[23] method. The method of Sudhakar Nayak and Pattabiraman[24] was used for assay the glycosylated hemoglobin. The plasma insulin was estimated by radio immuno assay kit purchased from Stat Diagnostics (Linco Research Inc.), Mumbai, India.

Hexokinase was assayed by the method of Brandstrup *et al.*[25] Lactate dehydrogenase was assayed according to the method of King.[26] Glucose-6-phosphatase was assayed according to the method of Koide and Oda.[27] Pyruvate kinase was assayed by Pogson and Denton[28] method. Fructose-1,6-bisphosphatase was assayed by the method of Gancedo and Gancedo.[29] Glucose-6-phosphate dehydrogenase was assayed by the method of Ell and Kirkman.[30]

For the estimation of glycogen, the extraction was carried out by the method of Morales *et al.*[31] Glycogen synthase was assayed by the method of Leloir and Goldenberg.[32] Glycogen phosphorylase was assayed by the method of Cornblath *et al.*[33]

Statistical Analysis —— All data were expressed as mean ± standard deviation for six animals in each group. All the grouped data were statistically evaluated with SPSS/7.5 software. Hypothesis testing methods included one way analysis of variance (ANOVA) followed by least significant difference (LSD) test. P values of less than 0.05 were considered to indicate statistical significance.

RESULTS

Acute Toxicity Studies

The normal rats were treated with mangiferin for a period of 30 days for toxicity study. The toxicity studies revealed nontoxic nature of the mangiferin at a concentration of 25, 50, 100, 250, and 500 mg/kg of body weight/day for a period of 30 days. There was no morphological change like respiratory distress, hair loss, restlessness, convulsions, laxative, coma, weight loss, *etc*. There was no lethality or any toxic reactions found at any of the doses selected till the end of treatment period.

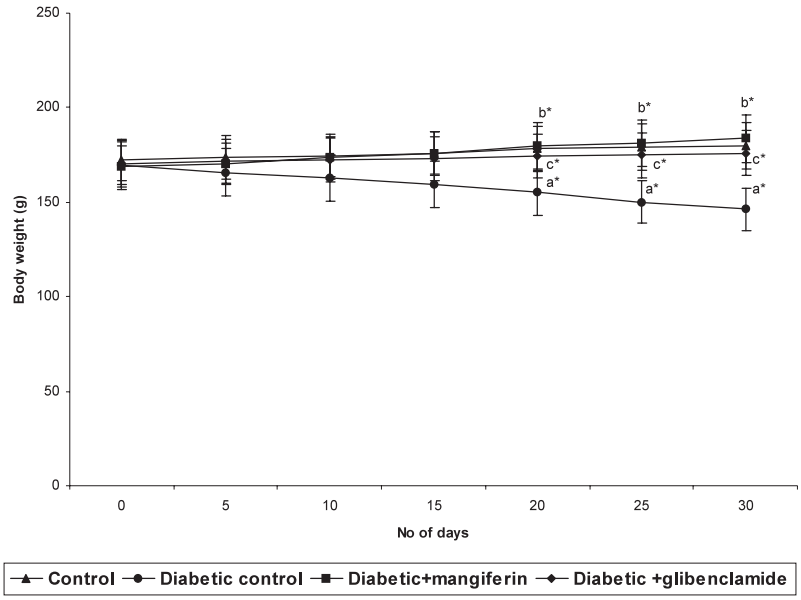

Fig. 1. Effect of Mangiferin on Body Weight in Control and Experimental Group of Rats

Data were given as mean ± standard deviation for six animals in each group. Values are statistically significant at $^{*}p < 0.05$. [a]Diabetic control rats were compared with normal control rats. [b]Mangiferin treated diabetic rats were compared with diabetic control rats. [c]Glibenclamide treated diabetic rats were compared with diabetic control rats.

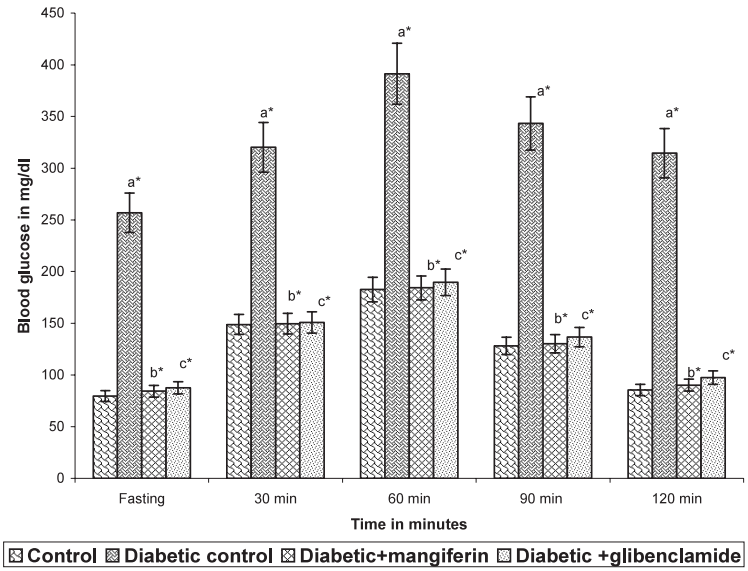

Fig. 2. Effect of Mangiferin on Glucose Tolerance Test in Control and Experimental Groups of Rats

Data were given as mean ± standard deviation for six animals in each group. Values are statistically significant at $^{*}p < 0.05$. [a]Diabetic control rats were compared with normal control rats. [b]Mangiferin treated diabetic rats were compared with diabetic control rats. [c]Glibenclamide treated diabetic rats were compared with diabetic control rats.

The body weight of control and experimental groups of rats were checked up to 30 days and represented in Fig. 1. The body weight was decreased in diabetic group of rats, when compared to control rats. In the initial days of the treatment, there was no significant difference in the body weight, but in later the body weight was significantly increased in mangiferin and glibenclamide treated rats, when compared to diabetic group of rats.

Glucose Tolerance Test

The blood glucose level, after oral administration of glucose in control and experimental rats were represented in Fig. 2. In the control rats, the blood glucose was reached maximum during 60 min and returned back near to the normal levels after

120 min. Whereas in diabetic rats, the blood glucose was increased to its peak at 1 hr, which remained high up to 120 min. The mangiferin was enhancing glucose utilization, so blood glucose level was significantly decreased in glucose-loaded rats.

The levels of blood glucose and plasma insulin in control and experimental rats were represented in Table 1. The diabetic rats showed a significant increase in blood glucose and decrease in plasma insulin compared with control rats. The mangiferin and glibenclamide treated diabetic rats were found to be similar to that of control rats.

The hemoglobin and glycosylated hemoglobin in control and experimental rats were represented in Table 2. The level of glycosylated hemoglobin was significantly increased and a concomitant decrease in the level of glycosylated hemoglobin in the diabetic rats, when compared with control rats. The mangiferin and glibenclamide treated diabetic rats significantly decreased the glycosylated hemoglobin level and increased the level of hemoglobin.

Table 1. The Levels of Blood Glucose and Plasma Insulin in Control and Experimental Groups of Rats

Groups	Blood glucose (mg/dl)	Plasma insulin (μU/ml)
Control	86.42 ± 5.53	16.37 ± 0.75
Diabetic control	263.18 ± 19.47$^{a)*}$	5.04 ± 0.29$^{a)*}$
Diabetic + mangiferin	92.54 ± 6.01$^{b)*}$	14.58 ± 0.65$^{b)*}$
Diabetic + glibenclamide	98.14 ± 6.57$^{c)*}$	12.39 ± 0.58$^{c)*}$

Data were given as mean ± standard deviation for six animals in each group. Values are statistically significant at $^{*}p < 0.05$. *a*) Diabetic control rats were compared with normal control rats. *b*) Mangiferin treated diabetic rats were compared with diabetic control rats. *c*) Glibenclamide treated diabetic rats were compared with diabetic control rats.

Carbohydrate Metabolizing Enzymes

The carbohydrate metabolizing enzymes activities (liver) of control and experimental groups of rats were represented in Tables 3 and 4. There was a significant decrease in the activities of hexokinase, pyruvate kinase, glucose-6-phosphate dehy-

Table 2. The Levels of Hemoglobin and Glycosylated Hemoglobin in Control and Experimental Groups of Rats

Groups	Hemoglobin (g/dl)	Glycosylated hemoglobin (%Hb)
Control	14.02 ± 0.67	5.6 ± 0.14
Diabetic control	10.25 ± 0.56$^{a)*}$	12.7 ± 0.45$^{a)*}$
Diabetic + mangiferin	13.08 ± 0.61$^{b)*}$	6.9 ± 0.17$^{b)*}$
Diabetic + glibenclamide	12.94 ± 0.59$^{c)*}$	7.2 ± 0.20$^{c)*}$

Data were given as mean ± standard deviation for six animals in each group. Values are statistically significant at $^{*}p < 0.05$. *a*) Diabetic control rats were compared with normal control rats. *b*) Mangiferin treated diabetic rats were compared with diabetic control rats. *c*) Glibenclamide treated diabetic rats were compared with diabetic control rats.

Table 3. Activities of Carbohydrates Metabolizing Enzymes in Liver of Control and Experimental Groups of Rats

Groups	Hexokinase	Lactate dehydrogenase	Pyruvate kinase
Control	258.62 ± 16.55	245.92 ± 16.23	9.24 ± 0.24
Diabetic control	128.34 ± 9.75$^{a)*}$	455.16 ± 34.59$^{a)*}$	3.68 ± 0.13$^{a)*}$
Diabetic + mangiferin	251.86 ± 16.37$^{b)*}$	251.31 ± 16.33$^{b)*}$	8.36 ± 0.21$^{b)*}$
Diabetic + glibenclamide	247.52 ± 15.84$^{c)*}$	265.79 ± 17.01$^{c)*}$	8.19 ± 0.19$^{c)*}$

Data were given as mean ± standard deviation for six animals in each group. Values are statistically significant at $^{*}p < 0.05$. *a*) Diabetic control rats were compared with normal control rats. *b*) Mangiferin treated diabetic rats were compared with diabetic control rats. *c*) Glibenclamide treated diabetic rats were compared with diabetic control rats. The enzyme activities are expressed as hexokinase-μ moles glucose-6-phosphate formed in hour/mg of protein, lactate dehydrogenase-μ moles of pyruvate formed/hr/mg of protein, and pyruvate kinase-μ moles of pyruvate formed in minute/mg of protein.

Table 4. Activities of Carbohydrates Metabolizing Enzymes in Liver of Control and Experimental Groups of Rats

Groups	Glucose-6-phosphatase	Fructose-1,6-bisphosphatase	Glucose-6-phosphate dehydrogenase
Control	1022.42 ± 86.90	491.65 ± 31.95	542.17 ± 35.24
Diabetic control	1898.27 ± 172.74$^{a)*}$	769.46 ± 56.94$^{a)*}$	265.43 ± 20.70$^{a)*}$
Diabetic + mangiferin	1084.12 ± 91.06$^{b)*}$	508.14 ± 32.52$^{b)*}$	498.72 ± 34.29$^{b)*}$
Diabetic + glibenclamide	1109.53 ± 95.41$^{c)*}$	531.07 ± 35.05$^{c)*}$	481.54 ± 32.26$^{c)*}$

Data were given as mean ± standard deviation for six animals in each group. Values are statistically significant at $^{*}p < 0.05$. *a*) Diabetic control rats were compared with normal control rats. *b*) Mangiferin treated diabetic rats were compared with diabetic control rats. *c*) Glibenclamide treated diabetic rats were compared with diabetic control rats. The enzyme activities are expressed as glucose-6-phosphatase and fructose-6-phosphatase-μ moles phosphate liberated in hour/mg of protein and glucose-6-phosphate dehydrogenase-units/min per mg of protein.

Table 5. Glycogen Level and Activities of Glycogen Phosphorylase and Glycogen Synthase in Liver of Control and Experimental Groups of Rats

Groups	Glycogen	Glycogen phosphorylase	Glycogen synthase
Control	52.47 ± 3.41	647.82 ± 42.10	831.15 ± 53.19
Diabetic control	18.26 ± 1.37[a)*]	886.10 ± 69.11[a)*]	552.74 ± 42.56[a)*]
Diabetic + mangiferin	49.32 ± 3.35[b)*]	697.4 ± 44.63[b)*]	805.67 ± 52.36[b)*]
Diabetic + glibenclamide	46.28 ± 3.10[c)*]	719.34 ± 48.91[c)*]	786.49 ± 50.33[c)*]

Data were given as mean ± standard deviation for six animals in each group. Values are statistically significant at $^{*}p < 0.05$. *a*) Diabetic control rats were compared with normal control rats. *b*) Mangiferin treated diabetic rats were compared with diabetic control rats. *c*) Glibenclamide treated diabetic rats were compared with diabetic control rats. The enzyme activities are expressed as glygogen phosphorylase-μ moles of phosphate liberated in hour/mg of protein, glygogen synthase-μ moles of UDP formed in hour/mg of protein, and the level of glycogen expressed as mg of glucose/g of liver tissue.

drogenase, and also an increase in the activities of lactate dehydrogenase, glucose-6-phosphatase, and fructose-1,6-bisphosphatase in diabetic rats as compared to control rats. The mangiferin and glibenclamide treated diabetic rats were restored to near normal activities.

The liver glycogen metabolism in control and experimental groups of rats were showed in Table 5, where a significant decrease in liver glycogen content and glycogen synthase activity was observed. An increase in glycogen phosphorylase was observed in diabetic rats when compared with control rats. The mangiferin and glibenclamide treated diabetic rats were brought back to near control.

DISCUSSION

The STZ is a broad-spectrum antibiotic extracted from *Streptomyces acromogenes*. The STZ-induced diabetes causes the destruction of β cells of the islets, which leads to a reduction in insulin release.[34)] An insufficient release of insulin, that leads high blood glucose namely hyperglycemia.

The body weight of STZ-induced diabetic rats were reduced and also recovered after hypoglycemic treatment.[35)] In our study also the body weight was gain in mangiferin and glibenclamide treated diabetic rats. The enhancement of body weight in STZ-induced diabetic treated rats because of increases glucose metabolism. The preliminary acute toxicity revealed the nontoxic nature of mangiferin.

The treatment of medicinal plant extract to the STZ-induced diabetic rats, that activated the β cells and granulation return to normal, like to be insulinogenic effect.[36)] The glibenclamide is a standard antidiabetic drug, used to compare the antihyperglycemic property in experimental rats. Glibenclamide have been involved in stimulating insulin secretion from pancreatic β cells principally by inhibiting ATP sensitive K_{ATP} channels in the plasma membrane.[4)] Courtois *et al.*[37)] have reported that glibenclamide treated STZ-induced diabetic rats showed decrease in blood glucose level. The previous reports are consistent with our present findings. The decreased level of blood glucose and increased level of plasma insulin were observed in our present study, which indicates that mangiferin stimulates insulin secretion from the remnant β cells or regenerated β cells. The mechanism of the antidiabetic activity of mangiferin may be involved by increasing either the pancreatic secretion of insulin from the remnant Q cells of the islets of Langerhans. Some plants have antidiabetic activity through insulin releasing stimulatory effects.[38)]

The oral glucose tolerance test showed that the mangiferin gave definite lower blood glucose level at the end of 60 min after glucose loaded and even lower level at end of 120 min. The mangiferin enhanced glucose utilization, so the blood glucose level was significantly decreased in glucose-loaded rats.

The excess of glucose is present in the blood during diabetes, which react with hemoglobin and form glycosylated hemoglobin. The various proteins including hemoglobin, albumin, collagen, and low density lipoprotein (LDL)/crystalline proteins under go nonenzymatic glycation in diabetes.[39)] The hemoglobin level was decreased in diabetic rats that may increase the formation of glycosylated hemoglobin. Glycosylated hemoglobin was found to be increased in diabetic mellitus and the amount of increase is directly proportional to that of fasting blood glucose level.[40)] The significant decrease in glycosylated hemoglobin indicated that the efficiency of mangiferin in glycemic control.

The hexokinase involved in the phosphorylation

step of glucose in glycolysis. Hexokinase was significantly reduced in the liver of diabetic rats, may be the reason for decreasing the utilization of glucose and the increased blood glucose level.[41] The mangiferin and glibenclamide treated diabetic rats showed increased activity of hexokinase that may lead to activation of glycolysis and increase the utilization of glucose for energy production.

Pyurvate kinase is regulated at the mRNA levels in insulin dependent diabetes.[42] The mangiferin and glibenclamide treated diabetic rats were increasing the activity of pyruvate kinase that may increase the utilization of glucose. The finding suggested that the mangiferin was improving the glucose metabolism by increase the utilization of glucose.

Lactate dehydrogenase and aldolase are the bifunctional enzymes involved in the glycolytic pathway. The lactate dehydrogenase system reflects the NAD^+/NADH ratio indicated by the lactate/pyruvate ratio of hepatocyte cytosol.[43] The mangiferin and glibenclamide treated diabetic rats were reversible to near normal lactate dehydrogenase activity. This may be regulated by NAD^+/NADH ratio.

Glucose-6-phosphatase and fructose-1,6-bisphosphatase are the important enzymes in regulating of gluconeogenic pathway. The activities of glucose-6-phosphatase and fructose-1,6-bisphosphatase were increased in the liver of diabetic rats.[44] The mangiferin inhibits gluconeogensis by inhibiting the activity of glucose-6-phosphatase and fructose-1,6-bisphosphatase. The mangiferin and glibenclamide treated diabetic rats enhanced glucose utilization by increasing the activity of glucose-6-phosphate dehydrogenase.

The glycogen content is decreased in liver muscle of diabetic rats.[45] But liver glycogen content was increased significantly in mangiferin treated diabetic rats, it may be due to the activation of the glycogen synthase system by the mangiferin. The glycogen phosphorylase and glycogen synthase are reciprocal nature. The activity of glycogen phosphorylase was increased and glycogen synthase activity was decreased in the liver of diabetic mice.[46] The mangiferin reversed the liver glycogen by means of decreasing the activity of glycogen phosphorylase and increasing the activity of glycogen synthase.

Thus our results is in agreement with other medicinal plants such as, aqueous extract of *Mangifera indica* leaves possesses antihyperglycemic activity in glucose-induced hyperglycemic rats and mice.[47] The mangiferin exerts antidiabetic properties by decreasing insulin resistance in non-insulin dependent KK/Ay mice.[19] The chronic administration of mangiferin significantly improved oral glucose tolerance in glucose-loaded normal rats.[48]

In conclusion, the administration of mangiferin is a significant restoration of the blood glucose, glycosylated hemoglobin, and carbohydrate metabolizing enzymes such as hexokinase, pyruvate kinase, glycogen synthase, lactate dehydrogenase, glucose-6-phosphatase, fructose-1,6-bisphosphatase, and glycogen synthase. The present study indicates that a significant antidiabetic effect of mangiferin potentiate the β cells of pancreas. The mangiferin enhance the glycolytic enzymes and controls the glucose metabolism in the liver tissues of STZ-induced diabetic rats that lead to normoglycemic. Therefore, the mangiferin possess antidiabetic activity by stimulating the insulin production from the pancreas, extrapancreatic action and its support to control the diabetes and their complications.

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