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IN VIVO ANTIHYPERLIPIDEMIC, ANTIOXIDATIVE EFFECTS OF *COCCINIA GRANDIS* (L.) VOIGT (CUCURBITACEAE) LEAF EXTRACT: AN APPROACH TO SCRUTINIZE THE THERAPEUTIC BENEFITS OF TRADITIONAL SRI LANKAN MEDICINES AGAINST DIABETIC COMPLICATIONS

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Keywords:

Antihyperlipidemic effect; Antioxidative effects; *Coccinia grandis*; Streptozotocin induced diabetic rats

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ABSTRACT: *Coccinia grandis* (Linn.) Voigt (Cucurbitaceae) has been widely used for the management of diabetes mellitus in Sri Lankan traditional medicine. The aim of the present study was to investigate antihyperlipidemic, antioxidative effects of the leaf extract of *C. grandis* in sterptozotocin induced diabetic rats and to standardize the leaf extract *of C. grandis* by standard analytical methods. The results showed that the concentration of serum total cholesterol, triglycerides and low density lipoprotein cholesterol were significantly lower in C. grandis (0.75 g/kg) treated rats than in diabetic control rats ($P < 0.05$). In addition, an improvement in the antioxidant potential is also showed in C. grandis treated diabetic rats. $P < 0.05$). The polyphenolic compounds, alkaloids flavonoids, saponins and sterols/tritepenoids were present in the C. grandis extract. The results revealed that the aqueous leaf extract of C. grandis (0.75 g/kg) exerted antihyperlipidemic and antioxidative effects in diabetic rats, corroborating the therapeutic use of *C. grandis* leaf extract in the management of diabetic complications in traditional medicine.

INTRODUCTION: Diabetes mellitus has been considered as one of the emerging health problems worldwide because of its high prevalence, adverse clinical outcomes, marked reduction in the quality of life of patients and high healthcare costs [1, 2, 3]. Accordingly, it has become an adverse public health crisis in most of the South Asian countries including Sri Lanka with a prevalence of 8.5% in the general population.

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The total number of deaths caused by diabetes mellitus and its associated complications reached to 16,318 of Sri Lankan population in 2015 [4].

Over the past few decades, there has been a rapid discovery of diverse therapeutic agents for the management of diabetes mellitus. Present pharmacological agents as insulin and oral hypoglycemic drugs aim at correcting/overcoming hyperglycemia and disturbances in the metabolism of carbohydrate, fat and protein resulting from absolute or relative lack of secretion of insulin [5]. Despite the use of different types of oral hypoglycemic agents as insulin releasers, insulin sensitizers and glucosidase inhibitors etc. for the treatment of diabetes mellitus, studies have consistently demonstrated that patients' adherence

to present therapeutic regimes have not been satisfactory. Regime complexity, occurrence of hypoglycemia, associated side effects, lack of confidence in immediate or future benefits and education/beliefs of people are among the common reasons limiting compliance [6]. Although, the present drugs are effective in reducing hyperglycemia, inadequacies in current pharmaceutical agents have resulted in developing new antidiabetic drugs with increased efficacy, lack of toxicity and side effects for the treatment of diabetes and its complications. Thus, despite some encouraging findings on the discovery of novel antidiabetic agents from medicinal plant extracts, extensive research is required for the discovery of antidiabetic drugs/lead compounds from medicinal plant extracts which act against hyperglycemia, dyslipidemia and oxidative stress [7, 8].

Medicinal plants in particular, have formed the basis of sophisticated traditional medicinal systems. Ethnobotanical studies have identified more than 1,200 species of plants with antidiabetic activity [9]. Accordingly, a number of medicinal plant extracts in the form of decoctions have been used by Sri Lankan Ayurvedic practitioners for the management of diabetes mellitus and associated conditions of dyslipidemia and oxidative stress induced microvascular complications [10]. However, this traditional knowledge derived empirically has to be further supported by scientific testing.

Coccinia grandis (Linn.) Voigt (Cucurbitaceae) is an edible perennial climber distributed in tropical Asia, commonly found in Sri Lanka, India and Pakistan. Every part of this plant is valuable in medicine and various decoctions have been prepared in traditional Sri Lankan medicine for the treatment of various skin diseases, diabetes mellitus, urinary tract infections, bronchitis, itchy skin eruptions and ulcers [11]. Further, leaf extract of *C. grandis* is widely used by traditional physicians for the management of hyperglycemia and related microvascular complications associated with diabetes mellitus [10]. Thus, extensive research on bioactivity studies of *C. grnadis* has been carried out by our research group based on the highly demanding therapeutic use of *C. grandis* in traditional medicine.

Dose response experiments of hot water leaf extract of *C. grandis* in streptozotocin induced (STZ) diabetic rats revealed that the optimum therapeutic dose of the extract was 0.75 g/kg [12]. Accordingly, this particular dose was used in the present investigation as well. In addition, extensive research carried out by our research group proved that the extract was able to induce β-cell regeneration as one of the antidiabetic mechanisms *in vivo* [13]. Even though the leaf extract of *C. grandis* has been used in the management of dyslipidemia and oxidative stress associated diabetic complications in traditional medicine, the *in vivo* antihyperlipidemic and antioxidative effects of the aqueous leaf extract of *C. grandis* at its optimum therapeutic dose have not beenreported to date.

The aim of the present study was to investigate antihyperlipidemic, antioxidative effects of the leaf extract *C. grandis* (0.75 g/kg) in sterptozotocin induced diabetic rats and to standardize the *C. grandis* by standard analytical methods.

METHODS:

Chemicals and instruments: D-glucose, glibenclamide and streptozotocin were purchased from Sigma-Aldrich Company (St. Louis, MO, USA). A UV visible spectrophotometer (Gallenkamp PLC, UK) and Olympus CX 21(Japan) microscope were used for spectrophotometric measurements and for the assessment of histopathology of the liver tissue respectively. A rotatory evaporator (Buchi, B-480) will be used in the standardization of *C. grandis* extract.

Plant material: Leaves of *C. grandis* were collected during May-June 2013 from the Southern region of Sri Lanka. Botanical identity was determined by the descriptions given by Jayaweera [11] and confirmed by comparing authentic samples at the National Herbarium, Royal Botanical Gardens, Peradeniya, Sri Lanka. A voucher specimen was preserved at the Department of Biochemistry, Faculty of Medicine, University of Ruhuna, Sri Lanka (Attanayake/2011/ 01).

Preparation of the plant extract: The leaves of *C. grandis* were cut into small pieces, dried at 40°C until a constant weight. Powdered plant material

(50.00 g) was dissolved in 400.0 mL of distilled water and refluxed for 4 h. The mixture was strained and the final volume was adjusted to 50.0 mL. The optimum effective therapeutic dose (0.75 g/kg) was prepared for *in vivo* experiments.

Animals: Healthy adult male rats of Wistar strain (200 ± 25 g body weight) were purchased from the Medical Research Institute (MRI), Sri Lanka and were used to carry out the experiments. They were housed in standard environmental conditions at the animal house of Faculty of Medicine, University of Ruhuna, Sri Lanka (Tem 25 ± 2°C, relative humidity 55- 65% and 12 ± 1 h light/ dark cycle). Rats were fed with a standard diet (MRI rat formulae, Sri Lanka) with free access to water before and during the experiment. The rats were randomized into various groups and allowed to acclimatize for a period of seven days under standard environmental conditions before the commencement of the experiments. The animals described as fasting were deprived of food and water for 12 h *ad libitum*.

All protocols used in this study were approved by the Ethical Review Committee of Faculty of Medicine, University of Ruhuna, Sri Lanka guided by the Council for International Organization of Medical Sciences (CIOMS) international guiding principles of biomedical research involving animals.

Development of diabetes mellitus in Wistar rats: Streptozotocin dissolved in citrate buffer (0.1M, pH 4.4) at a dose of 65 mg/kg was administered intraperitonially to rats fasted for 8 h. Thereafter, rats were maintained on 5% D-glucose solution for the next 24 h. Rats were allowed to stabilize for three days and on the 4th day, blood samples were drawn from tail vein to determine the blood glucose concentration to confirm the development of diabetes mellitus. Rats with fasting blood glucose concentration of 12.0mmol/L or above were considered as hyperglycemic and on the 4th day onwards the hyperglycemic rats were used in the experiments [14].

Experimental group design: Group one and two served as the healthy and diabetic control rats and received distilled water. Group three and four were diabetic rats, received the optimum effective therapeutic dose of the extract *C. garndis* (0.75 g/kg) and glibenclamide (0.50 mg/kg) daily for 30 days respectively. At the end of the study (on the 30th day), blood was collected by cardiac puncture and the liver was excised from sacrificed rats. Serum was separated from blood of all rats for the estimation of biochemical parameters. Liver tissue was excised for the assessment of histopathology.

Blood/serum glycemic parameters: Oral glucose tolerance test was performed in all groups on the 1st, 7th, 14th, 21st, 28th and 30th day. Blood glucose concentration was determined by using a spectrophotometric enzyme assay kit [15]. The effect was evaluated over a four hour period using area under the oral glucose tolerance curve [16].

Serum lipid parameters in diabetic rats: The concentrations of serum total cholesterol (TC), high density lipoprotein cholesterol (HDL-C), triglyceride (TG) were estimated in all rats using spectrophotometric enzyme assay kits [17, 18, 19]. The concentrations of serum low density lipoprotein cholesterol (LDL-C), very low density lipoprotein cholesterol (VLDL-C) were calculated using the Friedewald formulae [20].

Antioxidant markers in diabetic rats: Fasting serum activities of liver enzymes; alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP) were estimated using spectrophotometric enzyme assay kits [21, 22]. The estimation of reduced glutathione (GSH), activities of glutathione reductase (GR, EC 1.6.4.2), glutathione peroxidase (GPx, EC 1.11.1.9) and glutathione S-transferase (GST, EC2.5.1.18) in the liver homogenates were done using reported protocols [23, 24, 25]. Further the extent of lipid peroxidation and total protein were estimated in liver homogenates by the formation of malondialdehyde (MDA) using thiobarbituric acid and Lowry methods respectively [26, 27].

Qualitative and quantitative analysis of chemical constituents:

Phytochemical screening: Preliminary qualitative phytochemical screening for the presence of alkaloids, cardenolide glycosides, phenols, flavonoids, phytosterols, saponins, tannins, reducing sugars, and proteins was carried out by the reported protocol [28].

Total polyphenol content: Total polyphenol content was measured using Folin-Ciocalteu colorimetric method [29]. Plant extract (1. 0 mL of 0.05 g/mL) was mixed with 1.0 mL of 95% ethanol, 5.0 mL of distilled water and 0.5 mL of 50% Folin-Ciocalteu reagent. The mixture was allowed to react for 5 min and 1.0 mL of 5% sodium carbonate was added. Thereafter, it was thoroughly mixed and placed in dark at room temperature (27°C) for one hour and the absorbance was measured spectrophotometrically at 725 nm. Quantification was done with respect to the standard curve of gallic acid (0-50 µg/mL). The results were expressed in gallic acid equivalents mgGAE/g of the dry weight.

Total flavonoid content: The total flavonoid content of plant extract was determined using aluminum chloride colorimetric method of Koksal and Gulcin [30] based on the method of Chang et al. [31]. The plant extract (0.50 mL) was mixed with ethanol 95% (1.5 mL) followed by aluminium chloride 10% (0.10 mL), potassium acetate 1M (0.10 mL) and distilled water (2.8 mL). The resultant mixture was incubated at 27°C for 30 minutes. The absorbance of the reaction mixture was measured spectrophotometrically at 415 nm. The flavonoid content was calculated using standard calibration of quercetin solution in range of 0-50 µg. The results are expressed as micrograms of quercetin equivalent (QE)/g of the dry weight. cyclohexane in a ratio of 0.1:1:1 (v/v/v). Spots were observed under UV (both 254 nm and 366 nm) light and they were visualized using vanillin sulphuric acid.

Statistical analysis: The replicates of each sample were used for statistical analysis and the values were expressed as mean ± standard deviation in the *in vitro* study. The data were analyzed using analysis of variance (ANOVA) and the mean values for each group were compared by Dunnett's multiple comparison tests in the *in vivo* study. The level of significance was set at $P < 0.05$.

RESULTS:

Blood/serum glycemic parameters: Effect of the extract of *C. grandis* (0.75 g/kg) on fasting blood glucose concentration in diabetic rats is shown in **Fig.1.** The fasting blood glucose concentration of *C. grandis* treated diabetic rats was reduced significantly on the 14th day on wards for the period of 30 days ($P < 0.05$). The reduction in fasting blood glucose concentration with the administration of *C. grandis* and glibenclamide was 48% and 61% in streptozotocin induced diabetic rats on the 30th day respectively ($P < 0.05$). The healthy animals were normoglycemic throughout the experimental period. The total area under the curve values of plant extract treated diabetic rats showed a statistically significant improvement of 54% on the 30th day ($P < 0.05$, **Fig.2**).

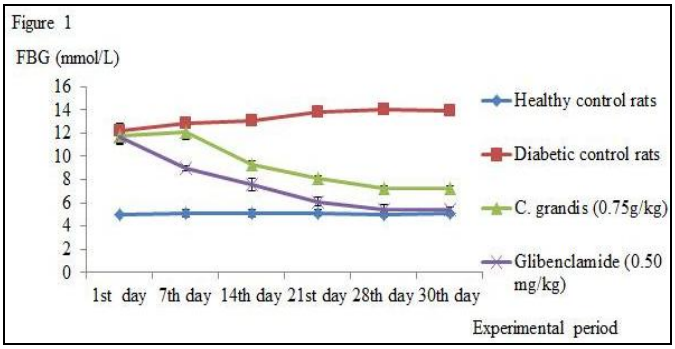


FIG. 1: EFFECT OF *C. GRANDIS* EXTRACT ON FASTING BLOOD GLUCOSE CONCENTRATION IN DIABETIC RATS AT SPECIFIC INTERVALS FOR 30 DAYS Each line represents mean ± SEM (*n*=6/group) FBG: Fasting blood glucose concentration.

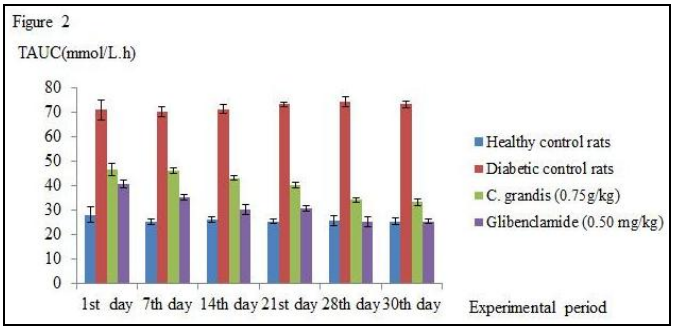


FIG. 2: TOTAL AREA UNDER THE ORAL GLUCOSE TOLERANCE CURVE VALUES OF *C. GRANDIS* EXTRACT TREATED DIABETIC RATS AT SPECIFIC INTERVALS FOR 30 DAYS

Each column represents mean ± SEM (n=6/group). TAUC: Total area under the curve from the 21st day onwards, total area under the curve value of *C. grandis* (0.75 g/kg) treated rats is significantly different from the total area under the curve values of diabetic untreated rats ($P < 0.05$).

Serum lipid parameters: The concentrations of serum TC, HDL-C, LDL-C, VLDL-C, TG in streptozotocin induced diabetic rats followed by the plant treatment is shown in **Fig. 3**. The streptozotocin induced diabetic control rats had a significant elevation in the concentration of serum TC (57%), LDL-C (93%), VLDL-C (95%), TG

(94%) and a reduction in HDL-C (12%) as compared with the untreated healthy control rats. As shown in **Fig.3,** the extract of *C. grandis* treated streptozotocin induced diabetic rats showed a significant reduction in the concentration of serum TC (25%), LDL-C (33%), VLDL-C (21%), TG (21%) and an elevation in HDL-C (4%) on the 30th day of the study ($P < 0.05$). The concentration of serum TC, LDL-C, VLDL-C, TG were reduced by 32%, 38%, 49% and 48% in glibenclamide treated diabetic rats. In contrast, there was no significant change in the concentration of serum HDL-C with the glibenclamide treatment in diabetic rats ($P > 0.05$).

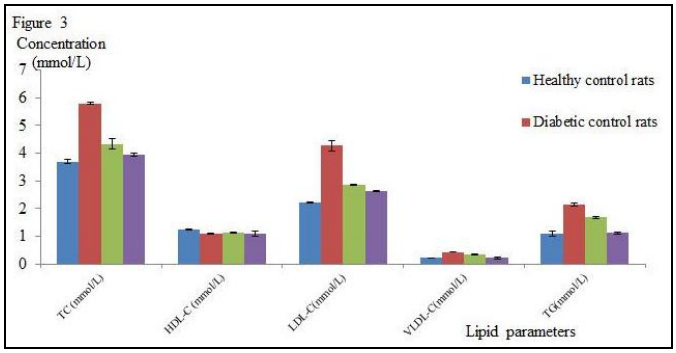


FIG. 3: EFFECT OF *C. GRANDIS* EXTRACT ON SERUM LIPID PARAMETERS IN DIABETIC RATS AFTER 30 DAYS OF TREATMENT

Each column represents mean ± SEM (n=6/group). TC: Total cholesterol, HDL-C: High density lipoprotein cholesterol, LDL-C: Low density lipoprotein cholesterol, VLDL-C: Very low density lipoprotein cholesterol, TG: Triglyceride. The concentrations of TC, HDL-C, LDL-C, VLDL-C and TG of *C. grandis* (0.75 g/kg) treated rats is significantly different from the total area under the curve values of diabetic untreated rats ($P < 0.05$).

Serum hepatic enzymes and oxidative stress markers: The effect of the *C. grandis* extract on liver enzymes and hepatic oxidative stress markers in STZ-diabetic rats is shown in **Table 1** and **2** respectively. There was an elevation in the activities of ALT, AST, ALP and the concentration of MDA in streptozotocin induced diabetic rats when compared to the untreated healthy rats. On the other hand, there was a reduction in the concentration of GSH, GR, GPx and GST in the same treatment groups. Treatment of diabetic rats with the extract and glibenclamide decreased the activities of ALT (by 18%, 16%), AST (by 16%, 17%) and ALP (by 40%, 5%) ($P<0.05$). The protective effect of the extract on lipid peroxidation was also demonstrated; a significant reduction in the concentration of MDA by 14% when compared to untreated diabetic rats ($P<0.05$). The administration of the plant extract restored the concentration of GSH, activities of GR, GPx, GST to near normalcy (by 41%, 49%, 40%, 57%) and it was more effective than the attainment of above biochemical parameters by glibenclamide (by 35%, 20%, 38%, 12%).

As shown in **Fig. 4**, liver histology was normal in untreated healthy rats. In contrast, untreated diabetic rats showed very early microvesicular fatty change in centrilobular areas of the liver, mild congestion, moderate lymphocytic infiltrates mostly around the portal tract, increased fibrosis with paranchymal infiltrates and focal necrosis. The light microscopic appearance of the liver tissue in *C. grandis* treated rats are in line with biochemical results, with a reduction in microvesicular fatty change, mild lymphocytic infiltrates and no necrosis. Further, reduction in microvesicular fatty change, moderate lymphocytic infiltrates were also observed in glibenclamide treated diabetic rats.

TABLE 1: EFFECT OF *C. GRANDIS* EXTRACT ON SERUM HEPATIC ENZYMES IN DIABETIC RATS AFTER 30 DAYS OF TREATMENT

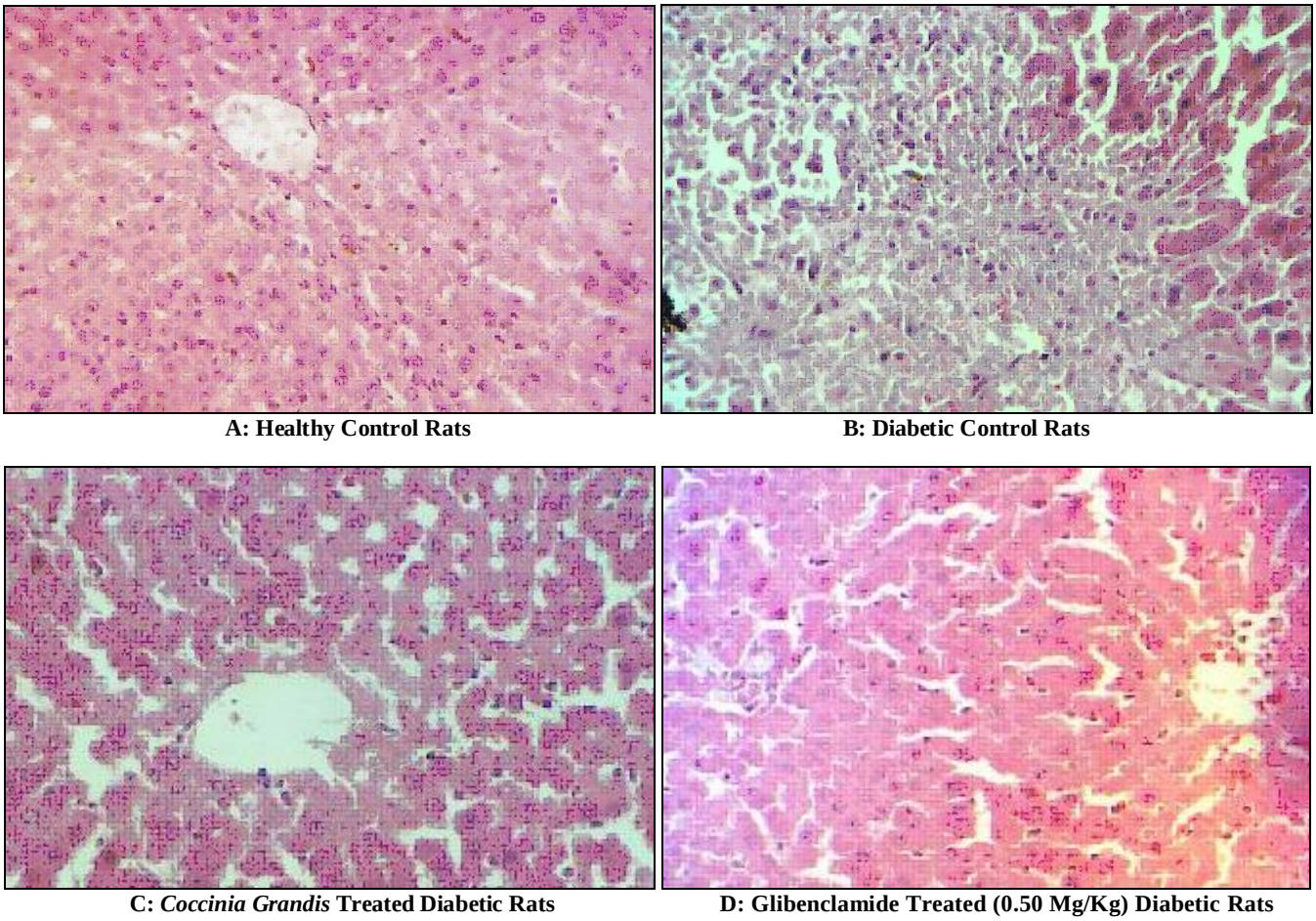
Treatment	ALT(U/L)	AST(U/L)	ALP(U/L)
Healthy control rats	12.4±0.1	44.2±1.7	61.4±1.0
Diabetic control rats	36.6± 2.2	90.1±0.1	165.6±2.5
C. grandis (0.75 g/kg)	29.9±0.3*	76.0±0.9*	100.0±2.8*
Glibenclamide (0.50 mg/kg)	30.6±1.3*	74.9±3.0*	56.6±2.3*

The values are expressed as mean ± SEM (*n*=6/group). *Statistically significant from streptozotocin induced diabetic control rats at $P < 0.05$ (ANOVA followed by Dunnett's test). ALT: Alanine aminotransferase, AST: Aspartate aminotransferase, ALP: Alkaline phosphatase.

TABLE 2: EFFECT OF *C. GRANDIS* EXTRACT ON HEPATIC ANTIOXIDATIVE STRESS MARKERS IN DIABETIC RATS AFTER 30 DAYS OF TREATMENT

Treatment	MDA (nmol/mg protein)	GSH (µg/g liver)	GR (nmol/min/mg of protein)	GPx (nmol/min/mg of protein)	GST (nmol/min/ mg of protein)
Healthy control rats	12.1±0.3	719.8±7.4	9.1±1.1	10.5±0.9	10.9±1.4
Diabetic control rats	36.3±1.1	419.6±8.7	5.3±0.5	4.6±1.0	4.7±0.1
C. grandis (0.75 g/kg)	31.1±0.3*	590.0±22.6*	7.9±0.7*	7.7±0.2*	7.4±0.6*
Glibenclamide (0.50 mg/kg)	22.1±0.2*	564.7±19.8*	6.3±0.9*	6.3±0.6*	7.3±0.2*

The values are expressed as mean ± SEM (*n*=6/group). *Statistically significant from streptozotocin induced diabetic control rats at P < 0.05 (ANOVA followed by Dunnett's test). MDA: Malondialdehyde, GSH: Reduced glutathione, GR: Glutathione reductase, GPx: Glutathione peroxidase, GST: Glutathione S-transferase



A: Healthy Control Rats **B: Diabetic Control Rats**

C: *Coccinia Grandis* Treated Diabetic Rats **D: Glibenclamide Treated (0.50 Mg/Kg) Diabetic Rats**

FIG. 4(A-D): PHOTOMICROGRAPHS OF HISTOPATHOLOGY OF THE LIVER TISSUES, STAINED WITH HEMATOXYLIN AND EOSIN (X 400)

(A) Healthy control rats, liver tissue is within normal histological limits
(B) Diabetic control rats, focal necrosis associated with inflammation
(C) *Coccinia grandis* (0.75 g/kg) treated diabetic rats, restoration of hepatocytes
(D) Glibenclamide treated (0.50 mg/kg) diabetic rats, restoration of hepatocytes

DISCUSSION: In the present study, the antihyperlipidemic and antioxidant activities of the leaf extract of *C. grandis* were evaluated in streptozotocin induced diabetic rat model. Streptozotocin is a pancreatic toxin that induces diabetes mellitus by destroying pancreatic β- cells resulting insulin deficiency. The cytotoxic effects of streptozotocin are dependent on DNA alkylation by site-specific action with DNA bases and by excessive formation of free radicals [32]. The area under the oral glucose tolerance curve of diabetic rats showed a highly significant elevation at fasting state 1h, 2h, 3h and 4h after glucose loading as compared to healthy (normoglycemic) animals.

However, the *C. grandis* extract appeared more effective at all tested points after oral glucose loading throughout the study period. In addition, from the observations made in **Fig. 1**, it is further confirmed that the leaf extract of *C. grandis* possesses significant antihyperglycemic activity as it could produce a fall of 48% in fasting blood glucose in diabetic rats after the plant treatment for 30 days.

Lipid abnormalities are postulated to be major causes for complications associated with diabetes mellitus. Indeed, diabetes is associated with profound alterations in the serum concentration of TG, lipoprotein profile and with an increased risk of coronary heart disease [33]. The hyperlipidemia associated with diabetes mellitus may result from an accelerated hepatic biosynthesis of triglycerides and a release of VLDL without an increase in its rate of clearance from the blood by lipoprotein lipase, which is dependent on the insulin/glucagon ratio. Furthermore, the alterations in lipid profile may be a consequence of increased breakdown of lipids and mobilization of free fatty acids from peripheral deposits [34]. Accordingly, an increase in the concentration of serum TC, TG and a decrease in the concentration of HDL-C were demonstrated in diabetic control rats in the present study. This is similarly mentioned by other authors who also observed alterations in the serum lipid parameters in diabetic rats [35, 36]. High serum concentration of TC and importantly, LDL-C are the predictors of atherosclerosis [37]. Therefore, the ideal treatment for diabetes mellitus should have favorable effects on serum lipid parameters in addition to the glycemic control.

Treatment of diabetic rats with *C. grandis* extract reversed dyslipidemia, as evidenced by a significant decrease in the serum concentration of TC, LDL-C and TG coupled to an increase in the serum concentration of HDL-C ($P < 0.05$). The increase in the concentration of HDL- C by the plant treatment is of therapeutic advantage as it improves the glycemic control as well as reduces the cardiovascular risk associated with diabetes mellitus. The strong antihyperlipidemic effect by the administration of *C. grandis* in diabetic rats could be through its' control on hyperglycemia or/and direct actions on the absorption of intestinal cholesterol and biosynthesis of cholesterol. Several authors have reported the antihyperlipidemic activities of medicinal plants in experimental diabetic animals which are similar to our results [38, 39].

Preliminary phytochemical screening revealed the presence of polyphenolic compounds, alkaloids flavonoids, saponins and sterols/tritepenoids in the extract of *C. garndis*. Saponins possess antihyperlipidemic activity and this has been reported to increase the lipoprotein lipase activity, which helps to remove free fatty acids from circulation, causing decrease in the concentration of serum cholesterol [40]. Flavonoids and other polyphenols may also contribute to the antihyperlipidemic activity by increasing the cholesterol metabolism and by modulating the enzymes involved in cholesterol metabolism, such as HMG CoA reductase, cholesterol 7α-hydroxylase and acyl-CoA:cholesterol acyltransferase [41, 42].

Liver is the vital organ of metabolism, detoxification, storage and excretion of xenobiotics and their metabolites [43].

Thus far, ALT, AST and ALP are reliable markers of liver function. Therefore, an increase in the activities of above liver enzymes in the serum of diabetic rats might be mainly due to the leakage of these enzymes from the liver cytosol into the blood stream which gives an indication of the hepatotoxic effect of streptozotocin [44]. Treatment of the diabetic rats with the leaf extract of *C. grandis* caused a reduction in the activity of these enzymes in serum compared to the diabetic untreated group. Further, the liver histopathology matches with biochemical results. These results are in agreement with those obtained by Kasetti et al. [45] and Eliza et al. [46] in diabetic rats.

The experimental data from the present study indicated that the concentration of malonaldehyde, an end-product of lipid peroxidation was elevated in the liver homogenates of diabetic rats. In contrast, the activities of hepatic antioxidant enzymes as GR, GPx and GST were decreased in diabetic rats. This may be due to the rise in generation of reactive oxygen species results in hyperglycemia and/or excessive formation of superoxide and hydroxyl radicals by the induction

of streptozotocin [47]. Indeed, the free radicals may inactivate the activities of these enzymes [48]. This may also be responsible for the insufficiency of antioxidant defences in mitigating reactive oxygen species mediated damage [48]. However, the administration of leaf extract of *C. grandis* was able to improve the activities of these antioxidant enzymes, increase the concentration of reduced glutathione and decrease the concentration of malonaldehyde in diabetic rats. The *in vivo* antioxidative activity of *C. grandis* may be due to the high content of polyphenolic compounds. Indeed, natural polyphenols are capable of scavenging free radicals, chelate metal catalysts, activate antioxidant enzymes, reduce α-tocopherol radicals and inhibit oxidases [48].

CONCLUSIONS: The results revealed that the leaf extract of C. grandis exert antihyperglycemic and antihyperlipidemic effects in streptozotocin induced diabetic rats, corroborating the therapeutic use of *C. grandis* leaf extract in the management of diabetic complications in traditional Sri Lankan medicine. Furthermore, the data obtained from the study, confirmed the safe consumption of *C. grandis* as a dietary adjunct and in the development of pharmaceutical products and neutarceuticals for the management of diabetes mellitus and its' associated complications.

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