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Assessment of The Therapeutic Role of *Mangifera indica* Leaves Extract in Diabetic Albino Rats

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ABSTRACT

Because synthetic antidiabetic medications have undesirable side effects and are expensive, various studies on medicinal plants with hypoglycemic properties have continued looking for alternative ways to prevent and manage type 2 diabetes (T2D) that may be more efficient, cost-effective and secure. Efficient bioactive substances such as flavonoids, tannins, alkaloids, terpenoids, anthraquinones, saponins, cardiac glycosides, and steroids were discovered in *Mangifera indica* leaves (MIL). The study's goal was to evaluate the MIL's therapeutic potential in diabetic rats treated with streptozotocin (STZ). In the experiment, forty male adult albino rats were employed. The animals were separated into four groups, each with ten rats, and evaluated as follows: group 1 (healthy control, HC); group 2 (untreated diabetic, UD); group 3 (mango treated diabetic, MTD); and group 4 (mango treated, MT). After 8 weeks of MIL extract administration, blood samples were taken to assess several biochemical and hematological markers. The treatment of MIL extract in diabetic rats resulted in significant reductions in blood glucose, glycated hemoglobin (HbA-1c), and amylase enzyme activity. The levels of insulin, vitamin C, and vitamin E were significantly increased with MIL extract treatment. Further, the extract showed significant antihyperlipidemic activity and renoprotective and hepatoprotective effects in diabetic rats. These findings suggested a beneficial effect of MIL extract in the treatment of T2D associated with hyperlipidemia, and liver and kidney complications.

INTRODUCTION

T2D is becoming more common in both developing and industrialized countries, and it will be the world's seventh-largest cause of death by 2030. One of the most prevalent chronic diseases worldwide is diabetes mellitus (DM), according to the International Diabetes Federation (IDF). In 2017, 451 million people were diagnosed with diabetes, and 5 million people died as a result of it (Cho *et al*., 2018). DM is a term used to describe chronic hyperglycemia brought on by impaired insulin production and/or insulin resistance

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in peripheral organs (Goyal & Jialal, 2022). Many variables, including genetic, environmental, and lifestyle (diet and exercise) factors might promote or prevent the development of DM. Furthermore, oxidative stress has been closely linked to DM.

Because of their intrinsic richness of antioxidants, plants and their extracts are interesting candidates for diabetes treatment. Recent research has found that vegetable byproducts (e.g., flowers, roots, seeds, bark, peel, leaves, etc.) may contain bioactive substances with antioxidant and anti-diabetic activities comparable to those of currently marketed drugs. (Naveen & Baskaran, 2018). As many types of hypoglycemic agents create a variety of adverse effects, the demand for natural therapies is increasing (Al-Qattan *et al.*, 2008). Mango leaves contain a lot of nourishing and bioactive compounds. (Palafox-Carlos *et al.*, 2012). Antioxidants and anti-inflammatory properties are abundant in mango leaves (Martin & He, 2009; Velderrain-Rodríguez *et al.*, 2018). The aim of this study is to evaluate the ability of mango leaf extract to optimize the blood glucose level, study the efficacy of mango leaves to reduce the oxidative stress accompanied by diabetes and investigate the potential of mango leaves to normalize some complications due to diabetes such as disturbances in lipid metabolism and kidney function.

MATERIALS AND METHODS

1. Plant Material:
The leaves of *Mangifera indica* (Mango) were collected freshly from a local farm in Jizan region, Saudi Arabia.

2. Chemicals:
Streptozotocin was purchased from Sigma (St. Louis, MO, U.S.A.), Sigma No. S0130. Streptozotocin is soluble in water. Solutions should be prepared just before use since the product is unstable. The drug should be stored frozen and protected from moisture and air. Double-distilled H_2O was used for preparation of mango leaf extract.

3. Animals:
Forty male Wistar albino rats (*Rattus norvegicus*), weighing between 150–200 gm, were used as animal models for this study. Rats were purchased from the animal house unit, Deanship of Scientific Research, University of Jazan, Saudi Arabia. The animals were allowed access to water *ad libitum* and maintained on a balanced normal rodent pellet. The study was conducted in the animal house (Deanship of Scientific Research, University of Jazan). Prior to the beginning and throughout the experiment, the rats were housed at 24˚C room temperature and 12h light: 12h dark cycle. All animal experiments comply with NIH guidelines for the animal care & use of laboratory animals (National Institutes of Health Publications No. 8023, revised 1985).

4. Mango Leaves Extract Preparation:
According to Nwinuka *et al.* (2008), an aqueous extract of *Mangifera indica* leaves was prepared. The mango leaves were freshly harvested from a nearby farm. The leaves were well-cleaned with tap water, diced into little pieces, shade dried in air at room temperature, then ground into a fine powder using an electric grinder. Ten grams of the powdered sample were added to 100 ml of distilled water that was already boiling, stirred for 15 minutes, and then left overnight. The extract was then filtered, and the filtrate was immediately applied.

5. Induction of Diabetes:
With exception of the healthy control group and the group that received MIL extract, the animals were fed a high-fat diet (HFD) for four weeks (Reed *et al.* 2000). The high-fat diet consisting of 40% fat, 41% carbohydrate, and 18% protein. After 4 weeks of HFD, the animals were intraperitoneally injected with a single dose of STZ (40 mg/ kg

b.w., dissolved in 0.9% NaCl) (Parveen *et al*. 2011). After three days from STZ injection, blood samples were drawn from the tail vein, and the glucose concentration was assessed using a commercial glucose meter (ACCU-CHEKR Instant). Rats with blood glucose ≥200 mg/dL were identified as diabetic and used in the study.

6. Animal Groups and Experimental Design:

Rats were randomly divided into four groups: ***Group I:*** the healthy control group (HC, n = 10), provided a standard diet throughout the test, and received saline only. ***Group II:*** untreated diabetic group (UD, n = 10), fed high-fat diet (HFD) for 4 weeks followed by intraperitoneal (ip) injection of streptozotocin (40 mg/kg). **Group III**: mango-treated diabetic group (MTD, n = 10), diabetic rats orally received mango leaves aqueous extract. This MTD group was subdivided into two subgroups, one with a dose of 1 gm/kg/day (MTD1), and the other (MTD2) with a dose of 2gm/kg/day. ***Group IV***: mango-treated group (MT, n = 10), normal rats were given an aqueous extract of mango leaves orally. This group was subdivided into two subgroups, one with a dose of 1 gm/kg/day (MT1), and the other (MT2) with a dose of 2gm/kg/day. The animals in the MTD group and MT groups were orally administered the extract by once daily gastric intubation with a syringe for about 8 weeks.

7. Biochemical and Hematological Analysis:

At the end of the experiment, each animal was anesthetized by being placed in an anesthetic box with diethyl ether vapor, which was periodically maintained by drizzling liquid ether on cotton wool at the bottom of the box. After the animal was anesthetized, the animal was sacrificed through the external jugular vein using a sharp blade. The blood was collected in two tubes, one is EDTA tube for CBC analysis and glycated hemoglobin, (HbA1c), and the other was a dry-clean centrifuge tube for serum collection. Glucose, liver enzymes (ALP, ALT, and AST), kidney function (Creatinine, Urea and uric acid), lipid profile (Triglycerides, Total cholesterol, LDL-C, HDL-C), and total protein were assayed by using Dimension RxL Max from Siemens Healthineers, following the manufacturer's instructions. The serum insulin concentrations were measured by an enzyme-linked immunosorbent assay (ELISA) method using ELISA kits following the manufacturer's instructions. Serum Amylase activities were assessed by an automated biochemical analyzer and were expressed as U/L. Vitamins C and E were measured using the Thermo Scientific™ Vanquish™ Flex Duo UHPLC system for Dual LC. The hematological parameters (RBCs, Hb, Hct, WBCs, PLT) and glycated hemoglobin (HbA1c) were estimated using Sysmex XP-300™ Automated Hematology Analyzer.

8. Statistical Analysis:

Statistical analysis was performed using SPSS PASW Statistics 18-Portable. The results are expressed as mean ± SEM. One-way analysis of variance (ANOVA) was used to assess the data, and then Tukey's multiple comparisons process. Any P value ≤ 0.05 was considered significant.

RESULTS

1. Antihyperglycemic Effects of Mango Leaf Extract:

The results illustrated in Figure 1, show the effect of oral administration of aqueous extract of *Mangifera indica* leaves (1 gm/kg and 2 gm/kg) on the level of blood glucose, HbA1c%, insulin level, and amylase activity in all studied groups. As shown in figure 1a, the diabetic group (UD) showed a marked high glucose level, this elevation was significantly higher than the healthy control group (HC). Oral administration of the MIL extract to diabetic rats at doses of 1 gm/kg and 2 gm/kg produced a significant decrease in blood glucose levels in the two groups (MTD1 and MTD2) compared to the diabetic group.

Rats treated with mango extract only (MT1 and MT2) did not produce any changes in the normal glucose level. Similarly, figure 1b revealed a marked significant increase of HbA1c% ($p < 0.05$) in the UD group compared to HC rats. Mango treatment in diabetic rats caused a decrease in HbA1c%, this decrease was more pronounced and significant after the higher dose (2 gm/kg), it reached about $4.2 \pm 0.03\%$ compared to $6.8 \pm 0.6 \%$ before the treatment. MT1 and MT2 groups did not show any significant difference compared to healthy control rats. On the other hand, figure 1c showed a marked decrease in insulin levels after STZ administration (UD). Oral administration of the aqueous MIL raised the level of insulin in both the MTD1 and MTD2 groups, this increase was significant and pronounced with the highest dose (2 gm/kg). The insulin level in the MT1 and MT2 groups did not differ significantly from its level in the HC group. In the UD group, the amylase activity was significantly increased. Oral administration with 1 or 2 mg/kg of the aqueous mango extracts showed a significant decrease in amylase activity in STZ diabetic rats ($P < 0.05$) as compared to the control group. The groups treated with the extract only did not show any significant difference as compared to the HC group.
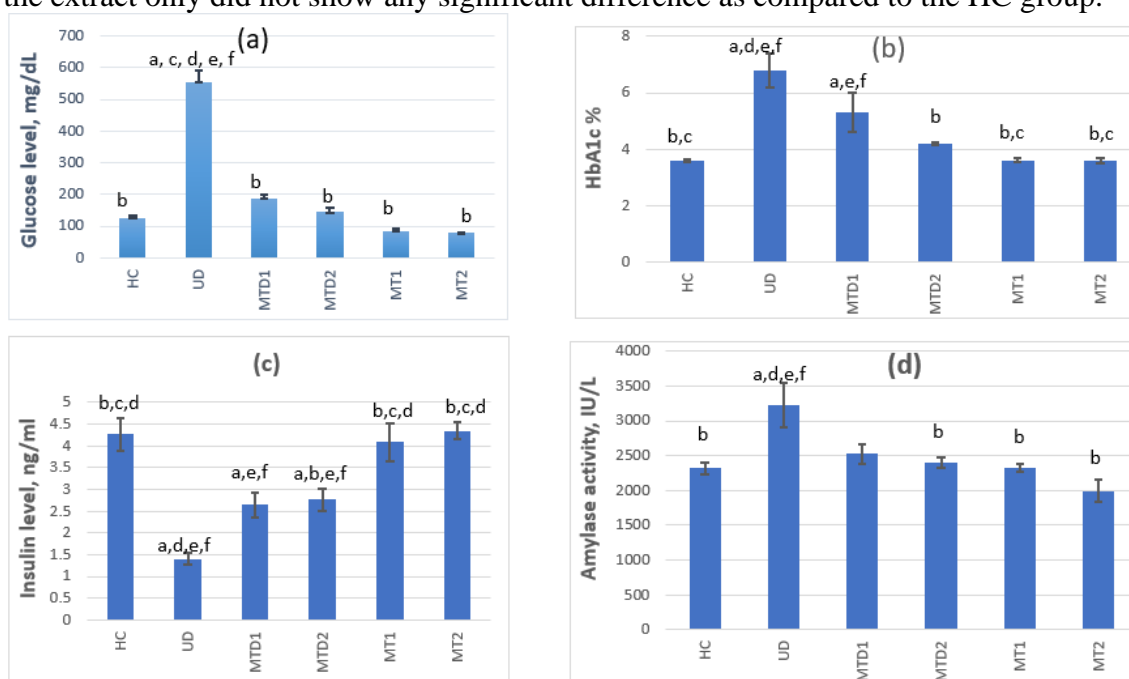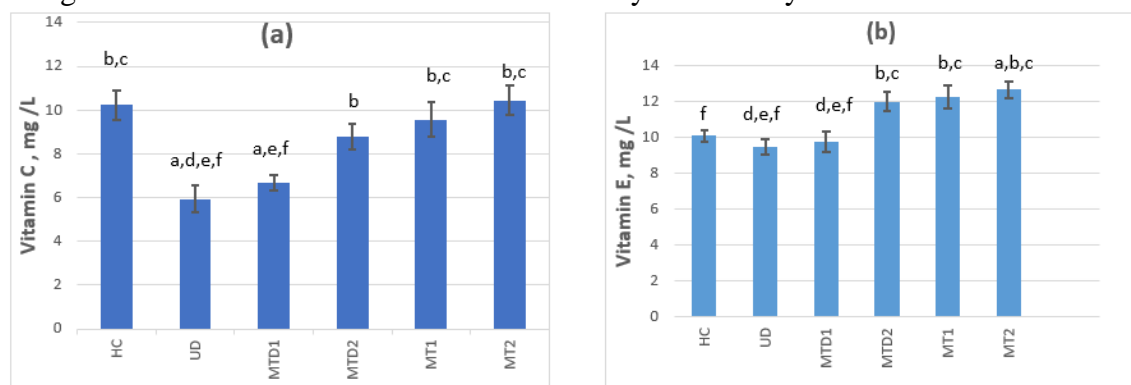

Fig. 1. Effect of 8 weeks of oral administration of MIL aqueous extracts (1 gm/kg and 2 gm/kg body weight) on (a) blood glucose level, (b) HbA1c %, (c) insulin level, and (d) amylase activity in STZ- diabetic rats. Results expressed as mean $\pm$ SEM (n= 10). [a] $P < 0.05$, significant against HC group; [b] $P < 0.05$, significant against UD group; [c] $P < 0.05$, significant against MTD1 group; [d] $P < 0.05$, significant against MTD2 group; [e] $P < 0.05$, significant against MT1 group; [f] $P < 0.05$, significant against MT2 group.

2. Effect of Mango Leaf Extract on Vitamin C and Vitamin E:

The results illustrated in Figure 2, show the effect of mango leaf extract on vitamin C and vitamin E in all experimental groups. As shown in figure 2c, a significant decrease in vitamin C levels in the UD group as compared to the HC group. The dose of 1 gm/kg of mango extract raised vitamin C levels slightly, but the increase was not significant. The higher dose (2 gm/kg) produced a significant increase in vitamin C as compared to UD. The groups treated with the extract only recorded a significant elevation as compared to UD and MTD1 groups. The level of vitamin E did not change significantly after STZ injection (UD group) compared to the HC group (figure 2b). Treatment of diabetic rats with the highest dose (2 gm/kg) of mango leaf extract resulted in a significant increase (P

< 0.05) in the level of vitamin E compared with the diabetic group. As shown in the figure, the vitamin E level was increased in the groups treated with the extract only, the increase was significant as compared to HC and UD groups. These findings support the role of mango leaves to elevate the antioxidant efficiency of the body.

Fig. 2. Effect of 8 weeks of oral administration of MIL aqueous extracts (1 gm/kg and 2 gm/kg body weight) on (a) vitamin C level, and (b) vitamin E level in STZ- diabetic rats. Results expressed as mean ± SEM (n= 10). [a] P < 0.05, significant against HC group; [b] P < 0.05, significant against UD group; [c] P < 0.05, significant against MTD1 group; [d] P < 0.05, significant against MTD2 group; [e] P < 0.05, significant against MT1 group; [f] P < 0.05, significant against MT2 group.

3.Effect of Mango Leaf Extract on A Renal Function Profile:

Figure 3, shows the effect of oral administration of an aqueous extract of mango leaves at doses of 1 gm and 2 gm/kg on blood urea nitrogen, creatinine and uric acid in the all-animal groups. As shown in Figure 3a, blood urea in the UD group was significantly elevated as compared to all other groups (HC, MTD1, MTD2, MT1, and MT2). Both doses of the mango extract produced a significant improvement and a decrease in urea levels compared to diabetic rats. The urea level was unaffected in the mango-treated groups (MT1 and MT2), and there was no significant difference between them and the control group. Similarly, figure 2b revealed that creatinine level was significantly increased after STZ injection (UD group), this increase was significantly higher than in HC, MTD1, MTD2, MT1 and MT2 groups. Mango leaves extract administration succeeded to reduce creatinine in MTD1 and MTD2 groups, and this reduction was significantly different from that UD group. 8 weeks of the extract administration was safe and did not cause any increase in creatinine as indicated by its level in MT1 and MT2 which did not change significantly from the HC group. Uric acid was increased in the UD group but this increase was insignificant. Statistically, there was no significant difference in uric acid levels between the different studied groups. Total protein in all studied groups did not show any significant difference.
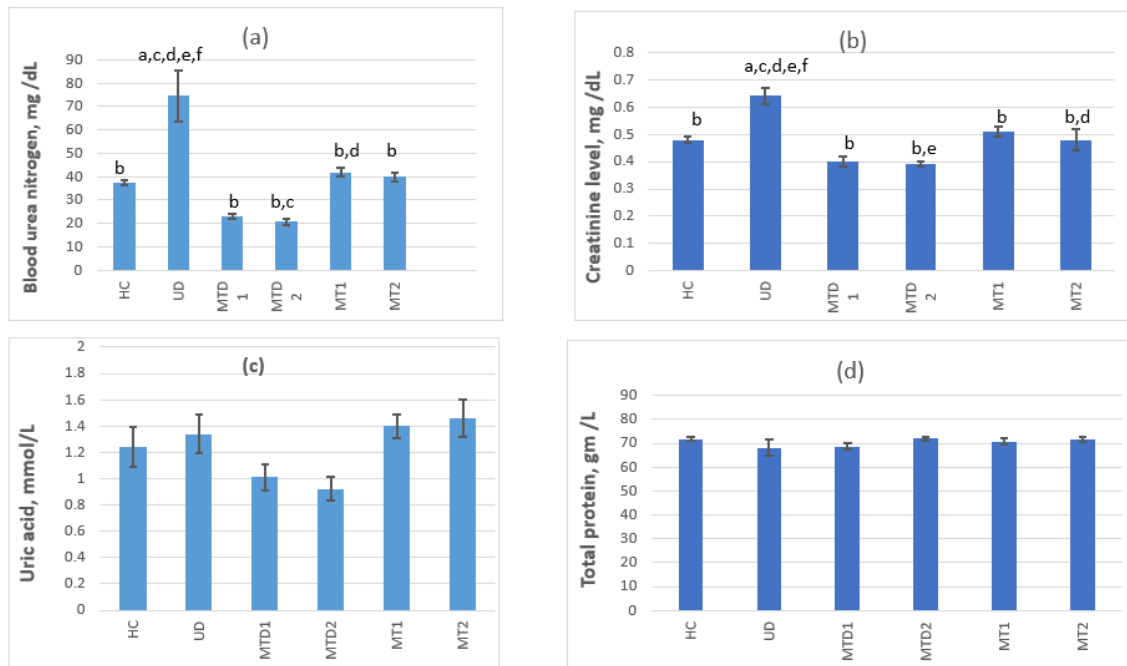
Fig. 3. Effect of 8 weeks of oral administration of MIL aqueous extracts (1 gm/kg and 2 gm/kg body weight) on (a) blood urea nitrogen, (b) creatinine, (c) uric acid, (d) total protein in STZ- diabetic rats. Results expressed as mean ± SEM (n= 10). [a] $P < 0.05$, significant against HC group; [b] $P < 0.05$, significant against UD group; [c] $P < 0.05$, significant against MTD1 group; [d] $P < 0.05$, significant against MTD2 group; [e] $P < 0.05$, significant against MT1 group; [f] $P < 0.05$, significant against MT2 group.

4. Effect of Mango Leaf Extract on A Liver Function Profile:

Figure 4, shows the effect of oral administration of aqueous extract of mango leaves extract at doses of 1 gm/kg and 2 gm/kg on blood ALP, ALT, and AST activities in all experimental groups. As shown in Figure 4a, the UD group showed the highest ALP activity, which was significantly higher than HC, MT1 and MT2 groups. Mango leaves extract administration to diabetic rats (MTD1 and MTD2) diminished ALP levels but the reduction was not significant. MT1 and MT2 groups did not differ significantly as compared to HC group, which reflects the safety of mango leaf administration on the liver enzyme ALP. Similarly, figure 4b showed a significant increase in ALT activity after STZ treatment as compared to all studied groups (UD, MTD1, MTD2, MT1 and MT2). ALT activity was improved and significantly reduced after the administration of 2 doses of the mango extract. MT1 and MT2 groups did not appear any significant change with the healthy control group. Likewise, As shown in figure 4c, there was an increase in AST activity after STZ injection, but this increase was statistically non-significant compared to HC group. AST was reduced in the MTD1 and MTD2 groups, this reduction was statistically significant only with the lowest dose of mango. Similarly, mango leaves extract administration to normal rats did not produce any significant change compared to HC group which confirms and reflects its effectiveness in improving liver functions without negative effects.
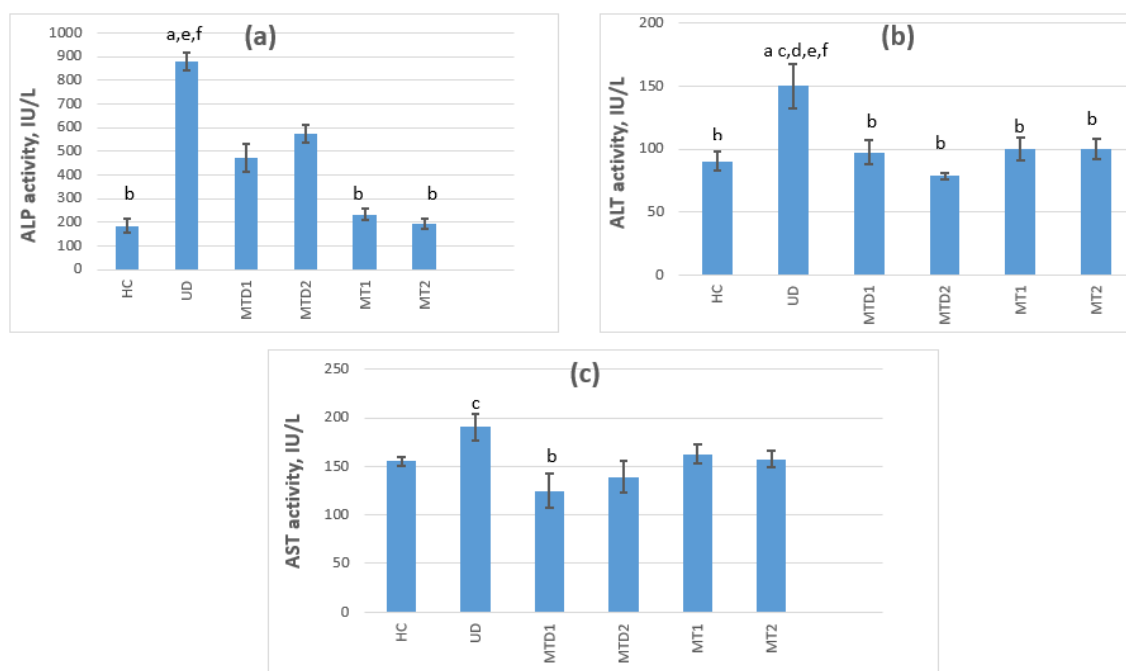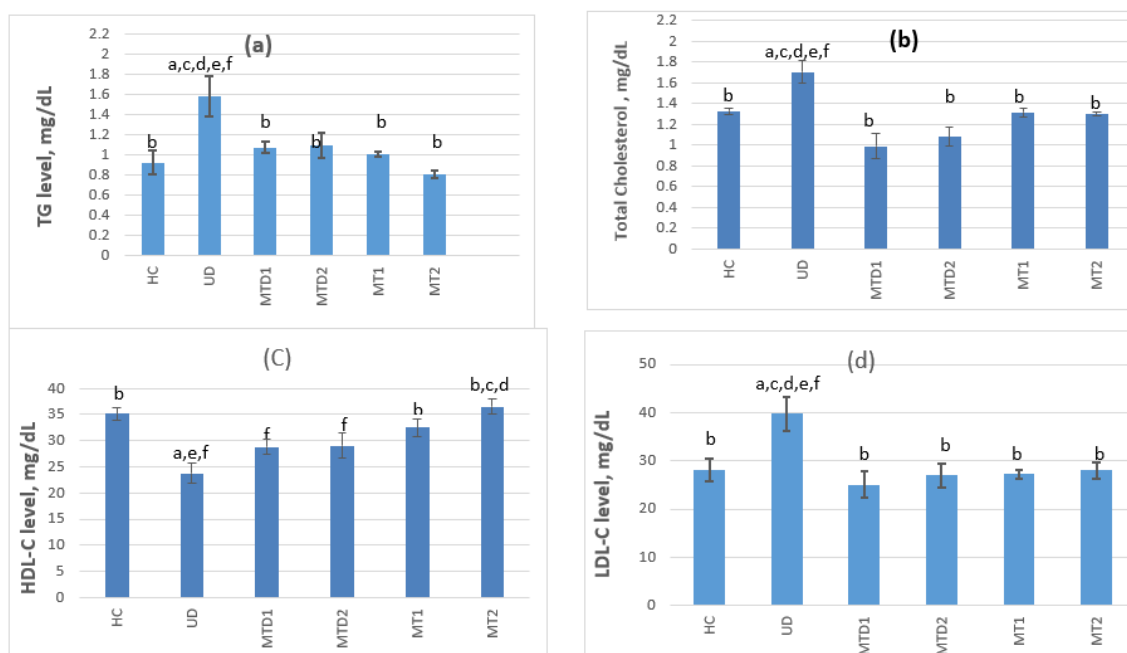
Fig. 4. Effect of 8 weeks of oral administration of MIL aqueous extracts (1 gm/kg and 2 gm/kg body weight) on (a) ALP, (b) ALT, and (c) AST activities in STZ- diabetic rats. Results expressed as mean ± SEM (n= 10). [a] P < 0.05, significant against HC group; [b] P < 0.05, significant against UD group; [c] P < 0.05, significant against MTD1 group; [d] P < 0.05, significant against MTD2 group; [e] P < 0.05, significant against MT1 group; [f] P < 0.05, significant against MT2 group.

5. Effect of Mango Leaf Extract on Lipid Profile:

Figure 5, refers to the effect of the aqueous extract of mango leaves on triglycerides levels, total cholesterol, LDL-C, and HDL-C. As it is apparent from Figure 5a, there was a significant elevation (P < 0.05) in blood TG in the untreated diabetic group when compared to HC group. The doses of 1 gm/kg and 2 gm/kg of mango extract significantly decreased (p <0.05) TG levels of the diabetic rats. The groups MT1 and MT2 showed a non-significant change in TG level compared to HC group but a significant decrease as compared to the UD group. The data in figure 5b revealed a significant increase (P < 0.05) in total cholesterol after STZ injection compared to the HC group. The extract administration had a good effect in lowering the cholesterol with the two doses (MTD1 and MTD2). On the other hand, there was not any significant difference between MT1 and MT2 in the HC group, this means mango leaves extract therapy is safe and does not elevate blood cholesterol levels. Figure 5c showed a significant decrease of HDL-C in UD group as compared to HC, MT1 and MT2 groups. HDL-C was elevated after mango extract treatment in diabetic rats, but the increase was not significant. The level of HDL-C in MT1 and MT2 groups does not differ significantly from HC group. In the same context, LDL-C was increased after STZ administration (UD group), and that elevation was significant compared to HC, MTD1, MTD2, MT1 and MT2 groups. Extract treatment at both doses (MTD1 and MTD2) significantly lowered LDL-C levels compared to UD group.

Fig. 5. Effect of 8 weeks of oral administration of MIL aqueous extracts (1 gm/kg and 2 gm/kg body weight) on (a) total glycerides, (b) total cholesterol, (c) HDL-C, and (d) LDL-C in STZ- diabetic rats. Results expressed as mean ± SEM (n= 10). [a] P < 0.05, significant against HC group; [b] P < 0.05, significant against UD group; [c] P < 0.05, significant against MTD1 group; [d] P < 0.05, significant against MTD2 group; [e] P < 0.05, significant against MT1 group; [f] P < 0.05, significant against MT2 group.

6. **Effect of Mango Leaves Extract on RBCs, Hb, HCT and WBCs:**

Table 1 shows the effect of the aqueous extract of mango leaves on RBCs, HB, HCT and WBCs. Statistically, there were no significant differences in RBCs, HB, and HCT between the different studied groups. The data presented in the table showed an increase in WBCs count after STZ injection (UD group) as compared to the HC group but statistically, the increase was not significant. MIL extract succeeded in significantly (P<0.05) decreasing the WBCs in diabetic rats in MTD1 and MTD2 groups.

Table 1. Effect of 8 weeks of oral administration of aqueous extract of *Mangifera indica* leaves (1 gm/kg and 2 gm/kg body weight) on RBCs count (x 10^6/µL), hemoglobin level (g/dL), hematocrit value (HCT) and WBCs count in adult male albino rats

Groups	RBCs count (x 10^6/µL)	Hb (g/dL)	HCT (%)	WBCs count (x 10^3/µL)
HC (healthy control)	8.7 ± 0.13	14.5 ± 0.09	41.6 ± 0.31	10.82 ± 2.04
UD (untreated diabetic)	8.2 ± 0.21	14.8 ± 0.3	42.3 ± 0.62	15.48 ± 0.53 [c,d]
MTD1 (mango-treated diabetic 1)	8.2 ± 0.21	14.1 ± 0.3	42.8 ± 1.02	9.06 ± 1.21 [b]
MTD2 (mango treated diabetic 2)	8.4 ± 0.19	13.9 ± 0.2	43.5 ± 0.76	8.52 ± 0.77 [b]
MT1 (mango treated 1)	8.2 ± 0.14	14.5 ± 0.2	41.1 ±1.04	12.92 ± 1.6
MT2 (mango treated 2)	8.3 ± 0.12	14.2 ± 0.3	40.8 ±1.04	13.06 ± 0.47

DISCUSSION

The prevalence of T2D has been rising quickly; by 2030, it will rank as the seventh greatest cause of death worldwide (WHO, 2016). Research on less-toxic and less expensive alternatives to synthetic anti-diabetic medications has increased in light of their negative side effects and high cost. Herbal dietary therapy provides a secure and affordable alternative to the existing pharmacological approaches (Hanhineva *et al*., 2010; Chang *et al*., 2013; Cao *et al*., 2019; Storz and Kuster, 2019). *Mangifera indica leaves (MIL)* have been found to be a rich source of phenolic acids and flavonoids (Batool *et al*., 2018; Atanu *et al*., 2022). In previous studies, rats were induced to develop T2D using a high-fat diet and the drug streptozotocin (HFD/STZ). (Parveen *et al*., 2011; Guo *et al*., 2021). In this model, the HFD causes hyperinsulinemia, insulin resistance, and/or glucose intolerance, and the STZ injection thereafter causes a significant decrease in the mass of functional β -cells (Szkudelski, 2001; Marianna *et al*., 2006; Lenzen, 2008; Mazo *et al*., 2016).

The present study revealed a significant increase in serum glucose levels in the HFD-STZ rats (UD group) as compared with the normal control rats (HC) group. These results are in accordance with the findings of several authors (Abou El-Soud *et al.,* 2007 and Bhowmik *et al*., 2009; Sankaranarayanan and Kalaivani, 2020). On the other hand, the results obtained for serum insulin of STZ-diabetic rats showed low levels as compared with normal healthy rats. Such results agree with that of Sellamuthu *et al.* (2009) and Zhang *et al*., (2020) and may be explained by the diabetogenic effect of STZ which leads to the destruction of ß-cells and decreased number of insulin-containing secretory granules. Treatment of diabetic rats with water extract of MIL at 1 gm/kg and 2 gm/kg showed a significant decrease in the blood glucose level. That improvement in blood glucose was consistent with the significant increases in insulin and this correlates well with the observations of Aderibigbe *et al.,* (2001), Bhowmik *et al*., (2009) and Villas-Boas *et al*., (2021). The extract significantly increased dose-dependent insulin sensitivity in diabetic animals, as well as the plasma insulin level. Muruganandan *et al*. (2005) suggested that both pancreatic and extrapancreatic mechanisms might be involved in its antidiabetic. However, the extrapancreatic actions could consist of (i) stimulation of peripheral glucose utilization, (ii) an enhancement of glycolytic and glycogen processes, and/or (iii) a glycemia reduction through the inhibition of glucose intake. Erato *et al.* (2005) and Saleem *et al*., (2019) clarified that MIL extract may be helpful in controlling diabetes and associated side effects, including weight reduction and lipid profile changes.

On the other hand, glycosylated hemoglobin (HbA1c) can be identified as a biomarker for detecting diabetes (Kundu *et al*., 2013). The increase in hemoglobin glycation under the hyperglycemic condition may be responsible for the rise in HbA1c % seen in the UD group. This is consistent with other researchers' reports (Bernadette *et al*., 2008; Surbakti *et al*., 2019; Kalidhindi *et al*., 2020). However, consumption of the MIL extract resulted in a decrease in the diabetic rats' HbA1c%. A significant increase in amylase activity was reported after HFD-STZ treatment. The results coincided with many previous findings such as Rocha *et al*., (2019) and Bashary *et al*., (2020). This elevation in amylase activity was antagonized by MIL treatment. The decrease in amylase activity causes a delay in the breakdown of carbohydrates and lengthens the overall time required for carbohydrate digestion, which lowers the rate of glucose absorption and slows the rise in postprandial plasma glucose. The inhibitory effects of *M. indica* leaf extract on -glucosidase and -amylase have been studied in a number of investigations (Andrew *et al*., 2013, Bhuvaneshwari *et al*., 2014; Ganogpichayagrai *et al*., 2017; Aswal *et al.,* 2020). The active phenolic compounds found in the plant may be responsible for the useful effects of the extract on the hyperglycemic conditions of the diabetic rats seen in this study (Boas *et*

al., 2020). The inhibition of enzymes that hydrolyze carbohydrates is one of the known methods by which these phenolic compounds exert their anti-diabetic effects (Iwai *et al.*, 2006; Bhowmik *et al.*, 2009; Ojo *et al.*, 2018).

Hyperglycemia is associated with the development of auto-oxidation of glucose (Bajaj & Khan, 2012; Herder *et al.*, 2013; Goycheva *et al.*, 2019). Ascorbic acid (Vitamin C) and vitamin E are powerful antioxidants that directly combat oxidizing radicals and protect cells from reactive oxygen species (Rai *et al.*, 2009). The present results showed that diabetic rats have much lower vitamin C levels than normal control rats. These results are in agreement with the findings of Naziroglu and Butterworth, (2005) and Ramos *et al.*, (2015). This vitamin reduction in diabetics, perhaps due to a greater need to control the excessive oxidative stress brought on by anomalies in glucose metabolism. An aqueous extract of MIL effectively increased vitamin C and E levels by a significant amount. This improvement can be ascribed to the antioxidant properties of the extract, which function to protect cells from harmful free radical damage (Viswanatha *et al.*, 2013; Ediriweera *et al.*, 2017).

The present results indicated that STZ administration (UD group) significantly reduced renal function through the increase in serum creatinine and urea concentrations as compared to healthy control rats. These results strongly suggest impairment of kidney function in diabetic rats. This increment in serum creatinine and urea in UD group may be a result of vascular and tubular damage induced by oxidative stress. Similar results have already been reported by Alarcon *et al.* (2005), Jaya *et al.* (2010), Manikandaselvi *et al.* (2012) and Liu *et al.* (2020). When administered to diabetic rats, MIL extract effectively reduced serum urea and creatinine levels and brought them back to normal. Renal function may improve as a result of MIL extract's antioxidant properties. These findings suggest the renoprotective properties of mango leaves. The results are consistent with Sahu *et al.* (2019) that suggested the renoprotective effects of mangiferin are due to its anti-apoptotic, anti-inflammatory, and antioxidant properties. Similar to this, Li *et al.* (2020) found that mangiferin dramatically slowed the evolution of diabetic nephropathy and enhanced renal function.

In the present study, ALT, AST, and ALP enzyme activities were increased in diabetic rats which is consistent with the findings of Nwanjo, (2007); Idris *et al.*, (2011) and Alanazi *et al.*, (2020). Some theories contend that sustained hyperglycemia will result in oxidative stress, the generation of free radicals, lipid peroxidation, and oxidative necrosis. (Mansour *et al.*, 2002; Idris *et al.*, 2011; Alanazi *et al.*, 2020). Due to the lack of intracellular glucose in DM, the hepatic cells become damaged and necrotic, which triggers an apoptotic response (Frances *et al.*, 2013). Additionally, a number of hepatic inflammatory reactions to DM have been documented (Lee and Lim, 2019). The activity of ALT, AST, and ALP in the diabetic rats was reduced after MIL treatment, this reduction suggests that the MIL may have a hepatoprotective effect. The favorable effects that have been seen are probably a result of the anti-inflammatory and antioxidant properties of the active compounds present in the mango leaf extract. The results agreed with those of Nadella and Kumar, (2016); Gazwi and Mahmoud, (2019); and Toledo *et al.*, (2019).

Because of primarily insulin resistance and insulin insufficiency, dyslipidemia is a typical biochemical characteristic T2D (Chahil and Ginsberg 2006; Chehade *et al.*, 2013; Poolsup *et al.*, 2019). Diabetes alters the plasma lipid profile due to abnormalities in lipid metabolism brought on by an increase in a free fatty acid release from insulin-resistant fat cells. (Chehade *et al.,* 2013). The diabetic rats' altered plasma lipid profiles observed in this study (more triglycerides, total cholesterol, LDL-C, and lower HDL-C) are consistent with findings from previous researchers' studies. (Zhang *et al.*, 2010; Kaga *et al.,* 2018). Following MIL treatment, it was shown that plasma levels of TG, TC, and

LDL-C decreased, while HDL-C levels increased. This improvement could be explained by the increase in insulin secretion and the increase in free fatty acid metabolism from peripheral fat depots (Pari and Latha, 2002; Saleh *et al.,* 2014). The lipid profile results demonstrated that MIL extract treatment of diabetic rats was beneficial as an antihyperlipidemic.

DM is associated with increased oxidative stress, production of free radicals, and a decline in antioxidant capability, as was previously addressed. White blood cells (WBCs) are the most significant inflammatory cells that aid in body defense. WBCs rise in oxidative conditions. This might explain why the UD group had more WBCs than the healthy control group. Additionally, the rise in liver enzymes (ALP, ALT, and AST) in UD rats can be used to explain the rise in WBC count (El-Metwaly *et al.,* 2019). In MTD groups, MIL extract reduced the number of WBCs. The extract's effectiveness as an anti-inflammatory and antioxidant can be used to explain this decline.

Conclusion

- The current study's findings suggested that *Mangifera indica* leaves aqueous extract had anti-diabetic effects in T2D rats and may be a promising nutritional therapy for the treatment of T2D and its related diabetic complications.
- Mango leaf extract is regarded as a safe medication because it did not have any negative effects on the liver, kidney, or body fat functions when consumed.

REFERENCES

Abou El-Soud, N. H., Khalil, M. Y., Hussein, J. S., Oraby, F. S. H. and Hussein, F. A. R. (2007): Antidiabetic effects of fenugreek alkaloid extract in streptozotocin induced hyperglycémie rats. *Journal of Applied Sciences Research,* 3(10): 1073-1083.

Aderibigbe, A. O., Emudianughe, T. S. and Lawal, B. A. (2001): Evaluation of the antidiabetic action of Mangifera indica in mice. *Phytotherapy Research,* 15(5): 456-458

Alanazi, A. Z., Alqahtani, F., Mothana, R. A. A., Mohany, M., Abuohashish, H. M., Ahmed, M. M. and Al-Rejaie, S. S. (2020): Protective Role of Loranthus regularis against Liver Dysfunction, Inflammation, and Oxidative Stress in Streptozotocin Diabetic Rat Model. *Evidence-Based Complementary and Alternative Medicine,* Article ID 5027986, 8 pages, https://doi.org/10.1155/2020/5027986

Alarcon, A. F. J., Calzada, B. F., Hernandez, G. E., Ruiz, A. C. and Roman, R. R. (2005): Acute and chronic hypoglycaemic effect of Ibervilles Sonorae root extracts. *Journal of Ethnopharmacology,* 97:447.

Al-Qattan, K., Thomson, M., and Ali, M. (2008): Garlic (Allium sativum) and ginger (Zingiber officinal) attenuate structural nephropathy progression in streptozotocin-induced diabetic rats. *The European e-Journal of Clinical Nutrition and Metabolism,* 3, e62–e71.

Andrew, O., Yusuf, S., Jangabe, L. M., Lawal, B. S., and Adamu, A. A. (2013): α-glucosidase inhibitory potential of selected anti-diabetic plants used in north-western Nigeria. *Journal of Medicinal Plants Research,* 7(27), 2010-2018.

Aswal, S., Kumar, A., Chauhan, A., Semwal, R.B., Kumar, A. and Semwal, D.K. (2020): A Molecular Approach on the Protective Effects of Mangiferin Against Diabetes and Diabetes-related Complications. *Current Diabetes Reviews,* 16(7):690-698.

Atanu, F.O, Ikeojukwu. A., Owolabi, P.A., and Avwioroko, O.J. (2022): Evaluation of chemical composition, in vitro antioxidant, and antidiabetic activities of solvent extracts of Irvingia gabonensis leaves. *Heliyon*, 8(7): e09922.

Bajaj, S., and Khan, A. (2012): Antioxidants and diabetes. *Indian Journal of Endocrinology and Metabolism,* 16(Supplement 2), S267–S71.

Bashary, R., Vyas, M., Nayak, S. K., Suttee, A., Verma, S., Narang, R., and Khatik, G. L. (2020): An Insight of Alpha-amylase Inhibitors as a Valuable Tool in the Management of Type 2 Diabetes Mellitus. Review, *Current Diabetes Reviews,* 16(2):117-136

Batool, N., Ilyas, N., Shabir, S., Saeed, M., and Mazhar, R. (2018): Mini-Review- A mini-review of therapeutic potential of *Mangifera indica* L. *Pakistan Journal of Pharmaceutical Sciences,* 31(4):1441-1448.

Bernadette, B. A. C. Sam, J., Chong, C. U., Culing, W., Clinton, R., Tatjana, S.V., Mitchell, *et al.* (2008): Diabetes, fasting glucose levels and risk ischaemic and vascular events. *Diabetes Care,* 31:1132–1137.

Bhowmik, A., Khan, L. A., Akhter, M. and Rokeya, B. (2009): Studies on the antidiabetic effects of *Mangifera índica* stem-barks and leaves on nondiabetic, typeI and type2 diabetic model rats. *Bangladesh Journal of Pharmacology,* 4: 1 IO-l 14

Bhuvaneshwari, J., Khanam, S., and Devi, K. (2014): p In-vitro enzyme inhibition studies for antidiabetic activity of mature and tender leaves of *Mangifera indica* var. *Totapuri. Journal of Microbiology and Biotechnology,* 3(3), 36-41

Boas, G. R., Lemos, J. M. R., de Oliveira, M. W., Dos Santos, R. C., da Silveira, A. P. S., Bacha, F. B., Ito, C. N. A., Cornelius, E. B., Lima, F. B., Rodrigues, A. M. S., Costa, N. B., Bittencourt, F. F., de Lima, F. F., Paes, M. M., Gubert, P., Oesterreich, S. A. (2020): Aqueous extract from Mangifera indica Linn. (Anacardiaceae) leaves exerts long-term hypoglycemic effect, increases insulin sensitivity and plasma insulin levels on diabetic Wistar rats. *PLoS One,* 8;15(1): e0227105. doi: 10.1371/journal.pone.0227105. eCollection.

Cao, N., Alexandre, E. C., Gotoh, D., Kurobe, M., Mizoguchi, S., Gu, B., and Yoshimura, N., (2020): Urethral dysfunction and alterations of nitric oxide mechanisms in streptozotocin-induced diabetic rats with or without low-dose insulin treatment. *Life Science,* 249:117537

Chahil, T. J., and Ginsberg, H. N. (2006): Diabetic dyslipidaemia. *Endocrinology & Metabolism Clinics of North America,* 35:491–510.

Chang, C. L. Y., Lin, A. P., Bartolome, Y. C., Chen, S. C., and Yang, W. C. (2013): Herbal therapies for type-2 diabetes mellitus: Chemistry, biology, and potential application of selected plants and compounds. *Journal of Evidence-Based Complementary and Alternative Medicine,* 2013:1–33.

Chehade, J. M., Gladysz, M. and Mooradian, A. D. (2013): Dyslipidaemia in type 2 diabetes: prevalence, pathophysiology, and management. *Drugs,* 73:327–339.

Cho, N. H., Shaw, J. E., Karuranga, S., Huang, Y., da Rocha Fernandes, J. D., Ohlrogge, A. W., and Malanda, B. (2018): IDF Diabetes Atlas: Global estimates of diabetes prevalence for 2017 and projections for 2045. *Diabetes Research and Clinical Practice,* 138, 271–281.

Ediriweera, M. K., Tennekoon, K. H., and Samarakoon, S. R. (2017): A review on ethnopharmacological applications, pharmacological activities, and bioactive compounds of *Mangifera indica* (Mango). *Evidence-Based Complementary and Alternative Medicine,* eCAM, Article ID: 6949835.

El-Metwaly, S., El-Senduny, F. F., El-Demerdash, R. S., and Abdel-Aziz, A. F. (2019): Mesenchymal stem cells alleviate hydrochloric acid-induced lung injury through suppression of inflammation, oxidative stress and apoptosis in comparison to moxifloxacin and sildenafil. *Heliyon*, 5(12): e02710.

Erato, P., Adebola, P. O., Grierson, D. S. and Afolayan, A. J. (2005): An ethno botanical study of plants used for the treatment of diabetes in the Eastem Cape Province, South Añica. *African Journal of Biotechnology,* 4: 1458-1460.

Frances, D. E. A., Ingaramo, P. I. and Mayoral, R. et al., (2013): "Cyclooxygenase - 2 over-expression inhibits liver apoptosis induced by hyperglycemia," *Journal of Cellular Biochemistry,* vol. 114, no. 3, pp. 669–680.

Ganogpichayagrai, A., Palanuvej, C., and Ruangrungsi, N. (2017): Antidiabetic and anticancer activities of Mangifera indica cv. Okrong leaves. *Journal of Advanced Pharmaceutical Technology and Research,* 8(1), 19-24.

Gazwi, H. S. S. and Mahmoud, M. E. (2019): Restorative activity of aqueous extract Mangifera indica leaves against CCl4 induced hepatic damage in rats *Journal of Pharmaceutical and Biomedical Analysis,* 164: 112–118

Goyal, R. and Jialal, I. (2022): Diabetes mellitus, type 2. Second ed., *StatPearls Publishing.*

Goycheva, P., Nikolova, G., Ivanova, M., Kundurdzhiev, T., and Gadjeva, V. (2019): Predictive value of some pro-oxidants in type 2 diabetes mellitus with vascular complications. *Bioscience Trends,* 13(2):168-175.

Guo, J., Li, J., Wei, H., and Liang Z. (2021): Maackiain Protects the Kidneys of Type 2 Diabetic Rats via Modulating the Nrf2/HO-1 and TLR4/NF-κB/Caspase-3 Pathways. *Drug Design Development and Therapy*, 14 (15):4339-4358.

Hanhineva, K. R., Törrönen, I., Bondia-Pons, J., Pekkinen, M., Kolehmainen, *et al.* (2010): Impact of dietary polyphenols on carbohydrate metabolism. *International Journal of Molecular Sciences,* 11:1365–1402.

Herder, C., Carstensen, M. and Ouwens, D.M. (2013): Anti-inflammatory cytokines and risk of type 2 diabetes. *Diabetes, Obesity and Metabolism, 15:39–50.*

Idris, A. S., Mekky, K. F. H., Abdalla, B. E. E. and Ali, K. A. (2011): Liver function tests in type 2 Sudanese diabetic patients. *International Journal of Nutrition and Metabolism,* 3:17–21.

Iwai, K., Kim, M. Y., Onodera, A. and Matsue, H. (2006): Alpha-glucosidase inhibitory and antihyperglycemic effects of polyphenols in the fruit of Viburnum dilatatum Thunb. *Journal of Agricultural and Food Chemistry,* 54:4588–4592.

Jaya, A., Shanthi, P., and Panchanadham, S. (2010): Cytoprotective effect of *Semecarpus anacardium* against toxicity induced by Streptozotocin in rats. *Journal of Experimental Pharmacology,* 2 :135-143.

Kaga, A. K., Barbanera, P. O., do Carmo, N. O., Rosa, L. R., and Fernandes, A. H. (2018): Effect of N-Acetylcysteine on Dyslipidemia and Carbohydrate Metabolism in STZ-Induced Diabetic Rats. *International Journal of Vascular Medicine,* 28;2018:6428630

Kalidhindi, S., Uddandrao, V. V. S., Sasikumar, V., Raveendran, N., and Ganapathy, S. (2020): Mitigating Perspectives of Asiatic Acid in the Renal Derangements of Streptozotocin-Nicotinamide Induced Diabetic Rats. *Cardiovascular & Hematological Agents in Medicinal Chemistry,* 18(1):37-44

Kundu, D., Roy, A., Mandal, T., Bandyopadhyay, U., Ghosh, E., and Ray, D. (2013): Relation of microalbuminuria to glycosylated haemoglobin and duration of type 2 diabetes. *Nigerian Journal of Clinical Practice,* 16:216–220.

Lee, H and Lim, Y. (2019): "Gamma-tocopherol ameliorates hyperglycemia- induced hepatic inflammation associated with NLRP3 inflammasome in alloxan-induced diabetic mice*," Nutrition Research and Practice, vol. 13, no. 5, pp. 377–383.*

Lenzen, S. (2008): The mechanisms of alloxan-and streptozotocin-induced diabetes. *Diabetology,* 51:216–226.

Li, X., Yan, Z., Carlström, M., Tian, J., Zhang, X., Zhang, W., Wu, S., and Ye, F. (2020): Mangiferin ameliorates hyperuricemic nephropathy which is associated with downregulation of AQP2 and increased urinary uric acid excretion. *Frontiers in Pharmacology,* 7;11:49.

Liu, Y., Kou, D., Chu, N., and Ding, G. (2020): Cathelicidin-BF attenuate kidney injury through inhibiting oxidative stress, inflammation and fibrosis in streptozotocin-induced diabetic rats. *Life Science,* 15;257:117918.

Manikandaselvi, S. Subalakshmi, R., Thinagarbabu, R. and Ravikumar, R. (2012): Evaluation of antidiabetic potential of ethanolic extract of leaves of ficus bengalensis line. *International Journal of Pharma and Bio Sciences,* 3(2): B108-B113.

Marianna, M., Chaonan, D., QingPing, H., Yanling, L. and Ping, L. (2006): Streptozotocin-induced diabetes causes astrocyte death after ischemia and reperfusion injury. *Diabetes,* 55:349–355.

Martin, M., and He, Q. (2009): Mango bioactive compounds and related nutraceutical properties-A review. *Food Reviews International, 25, 346–370.*

Mazo, V. K., Sidorova, Y. S., Zorin, S. N. and Kochetkova, A. A., (2016): Streptozotocin induced diabetes rat models. *Voprosy pitaniia,* 85(4):14-21.

Muruganandan, S., Srinivasan, K., Gupta, S., Gupta, P. K, and Lal, J. (2005): Effect of mangiferin on hyperglycemia and atherogenicity in streptozotocin diabetic rats. *Journal of Ethnopharmacology,* 97(3), 497- 501.

Nadella, S.K. and Kumar, N. R. (2016): *Mangifera indica* leaves extract effect on liver function in experimental animal studies. *Biomedical and Pharmacology Journal, 9 (3):* 1009-1012.

Naveen, J., and Baskaran, V. (2018): Antidiabetic plant-derived nutraceuticals: A critical review. *European Journal of Nutrition,* 57(4), 1275–1299.

Naziroglu, M. and Butterworth, P. (2005): Protective effects of moderate exercise with dietary vitamin C and E on blood antioxidative defense mechanism in rats with streptozotocin-induced diabetes. *Canadian Journal of Applied Physiology ;30(2):172–85.*

Nwanjo, H. U. (2007): Studies on the effect of aqueous extract of Phyllanthus niruri on plasma glucose level and some hepatospecific markers in diabetic Wistar rats. *International Journal of Laboratory Medicine,* 2:1–18.

Nwinuka, N. M., Monanu, M. O. and Nwiloh, B. (2008): Effects of Aqueous Extract of *Mangifera indica L.* (Mango) Stem Bark on Haematological Parameters of Normal Albino Rats. *Pakistan Journal of Nutrition,* 7(5):663-666.

Ojo, O.A., Afon, A. A., Ojo, A. B., Ajiboye, B. O., Oyinloye, B. E. and Kappo, AP. (2018): Inhibitory Effects of Solvent-Partitioned Fractions of Two Nigerian Herbs (Spondias mombin Linn. and Mangifera indica L.) on α-Amylase and α-Glucosidase. *Antioxidants (Basel)*, 7(6):73.

Palafox-Carlos, H., Yahia, E. M., and González-Aguilar, G. A. (2012): Identification and quantification of major phenolic compounds from mango (*Mangifera indica*, cv. Ataulfo) fruit by HPLC–DAD–MS/MS-ESI and their individual contribution to the antioxidant activity during ripening. *Food Chemistry,* 135(1), 105–111

Pari, L., and Latha, M. (2002): Effect of Cassia auriculata flowers on blood sugar levels, serum and tissue lipids in streptozotocin diabetic rats. *Singapore Medical Journal,* 43(12): 617–621.

Parveen, K., Khan, R. and Siddiqui, W. A. (2011): Antidiabetic effects afforded by Terminalia arjuna in high fat-fed and streptozotocin-induced type 2 diabetic rats. *International Journal of Diabetes and Metabolism,* 19:23–33.

Poolsup, N., Suksomboon, N., Kurnianta, P. D. M., and Deawjaroen, K. (2019): Effects of curcumin on glycemic control and lipid profile in prediabetes and type 2 diabetes mellitus: A systematic review and meta-analysis. *PLoS One,* 23;14(4): e0215840

Rai, D. K., Rai, P. K., Rizvi, S. I. Watal, G., and Sharma, B. (2009): Carbofuran-induced toxicity in rats: protective role of vitamin C.*Exp. Toxicologic Pathology,* 61(6):531–5.

Ramos, R. V., Guadarrama-López, A. L., Martínez-Carrillo, B. E., and Benítez-Arciniega, A. D. (2015): Vitamins and type 2 diabetes mellitus. *Endocrine, Metabolic & Immune Disorders-Drug Targets,* 15(1):54-63.

Rocha, S., Sousa, A., Ribeiro, D., Correia, C. M., Silva, V. L. M., Santos, C. M. M., Silva, A. M. S., Araújo, A. N., Fernandes, E., and Freitas, M. (2019): A study towards drug discovery for the management of type 2 diabetes mellitus through inhibition of the carbohydrate-hydrolyzing enzymes α-amylase and α-glucosidase by chalcone derivatives. *Food and function journal,* 1;10(9):5510-5520

Sahu, A. K., Verma, V. K., Mutneja, E., Malik, S., Nag, T. C., Dinda, A. K. Dharamvir Singh Arya, D. S. and Bhatia, J. (2019): Mangiferin attenuates cisplatin-induced acute kidney injury in rats mediating modulation of MAPK pathway. *Molecular and Cellular Biochemistry,* 452:141–152.

Saleem, M., Tanvir, M., Akhtar, M. F., Iqbal, M., and Saleem, A. (2019): Antidiabetic Potential of *Mangifera indica* L. cv. Anwar Ratol Leaves: Medicinal Application of Food Wastes. *Medicina,* 9;55(7):353

Saleh, S., El-Maraghy, N., Reda, E., and Barakat, w. (2014): Modulation of diabetes and dyslipidemia in diabetic insulin-resistant rats by mangiferin: role of adiponectin and TNF-α. *Anais da Academia Brasileira de Ciencias,* 86(4):1935-48

Sankaranarayanan, C., and Kalaivani, K. (2020): Isopulegol Mitigates Hyperglycemia Mediated Oxidative and Endoplasmic Reticulum Stress in HFD/STZ Induced Diabetic Rats. *Archives of Medical Research,* 51(3):204-214.

Sellamuthu, P. S., Muniappan, B. P., Perumal, S. M. and Kandasamy, M. (2009): Antihyperglycemic effect of mangiferin in streptozotocin induced diabetic rats. *Journal of Health Sciences,* 55(2): 206-214.

Storz, M. A., and Küster, O. (2019): Hypocaloric, plant-based oatmeal interventions in the treatment of poorly-controlled type 2 diabetes: A review. *Nutrition and Health,* 25(4):281-290.

Surbakti, C., Sitorus, P., Rosidah, Satria, D., (2019): Effect of Saurauia vulcani Korth. Leaves on Superoxide Dismutase, HbA1c Levels and Insulin Expression in Hyperglycemic Rats. *Macedonian Journal of Medical Sciences,* 14;7(22):3741-3744

Szkudelski, T. (2001): The mechanism of alloxan and streptozotocin action in β-cells of the rat pancreas. *Physiological Research*, 50:537–546.

Toledo, R. C. L., Brito, L. F., Caetano, M. M. M., Nakajima, V. M., da Silva, B. P., de Freitas Soares, F. E., Martino, H. S. D., and de Queiroz, J. H. (2019): Acute treatment with Mangifera indica L. leaf extract attenuates liver inflammation in rats fed a cafeteria diet. *Food and function journal*, 1;10(8):4861-4867

Velderrain-Rodríguez, G., Torres-Moreno, H., Villegas-Ochoa, M., Ayala-Zavala, J., Robles-Zepeda, R., Wall-Medrano, A., and González-Aguilar, G. 2018: Gallic acid content and an antioxidant mechanism are responsible for the antiproliferative activity of 'Ataulfo'mango peel on LS180 cells. *Molecules,* 23(3), 695.

Villas-Boas G. R., Paes M. M., Gubert, P., Oesterreich, S. A. (2021): Evaluation of the toxic potential of the aqueous extract from *Mangifera indica* Linn. (Anacardiaceae) in rats submitted to experimental models of acute and subacute oral toxicity. *Journal of Ethnopharmacology*, 275:114100.

Viswanatha, G. L, Shylaja, H., and Mohan, C. G. (2013): Alleviation of transient global ischemia/reperfusion induced brain injury in rats with 1,2,3,4,6-penta-Ogalloyl-β-D-glucopyranose isolated from Mangifera indica. *European Journal of Pharmacology,* 720(1- 3), 286-293.

World Health Organization. (2016): Diabetes Fact Sheet. *http://www.who.int/mediacentre/factsheets/fs312/en, accessed 29 September 2016.*

Zhang, W., Zhao, J., Wang, X., Pang, X., Zhuang, X., Zhu, *et al.* (2010): Hypoglycemic effect of aqueous extract of sea buckthorn (*Hippophae rhamnoides* L.) seed residues in streptozotocin-induced diabetic rats. *Phytotherapy Research,* 24:228–232.

Zhang, Y., Zhou, G., Peng, Y., Wang, M., and Li, X. (2020): Anti-hyperglycemic and anti-hyperlipidemic effects of a special fraction of Luohanguo extract on obese T2DM rats. *Journal of Ethnopharmacology,* 30;247:11227