WORLD JOURNAL OF PHARMACY AND PHARMACEUTICAL SCIENCES
SJIF Impact Factor 7.421
Volume 7, Issue 12, XXX-XXX Research Article ISSN 2278 – 4357

BIOCHEMICAL STUDY ON THE EFFECT *MURRAYA KOENIGII (CURRY)* AND *MORINGA OLEIFERA* IN CARDIAC DYSFUNCTION IN DIABETIC RATS

Yakout A. El Senosi*[1], Omayma A. R. Abou Zaid[2], Afaf Desouky A.elmaged[3], EL Saeed E. Ahmed[4] and Heba S. EL. Din Ebrahim[5]*

[1,2,3,4,5]Department of Biochemistry, Faculty of Vet., Medicine Benha University, Egypt.

Article Received on
16 October 2018,

Revised on 06 Nov. 2018,
Accepted on 27 Nov. 2018,

DOI: 10.20959/wjpps201812-12841

***Corresponding Author**
Dr. Yakout A. El Senosi
Department of Biochemistry, Faculty of Vet., Medicine Benha University, Egypt.
hebasaladin98@gmail.com

ABSTRACT

In the present study, the biochemical effect of ***Murraya Koenigii (curry)*** and ***Moringa oleifera*** aquous extract on cardiac dysfunction in experimentally induced diabetes mellitus in rats by Streptozotocin (STZ) was evaluated. Sixty male albino rats **weighting 150-200 g** and divided into four equal groups of 15 rats each. Group 1 :(Control group): received normal diet only. Group 2 :(Diabetic rats group): rats received a single dose of Streptozotocin (STZ) (50- mg/kg-b.wt i.p). Group 3 :(Diabetic rats + *Murraya Koenigii (curry)* aquous extract of leaves at dose of 200mg/kg/ twice daily, orally for four weeks after diabetes induction. Group 4:(Diabetic rats+ *Moringa oleifera* aquous extract of leaves at dose of 200 mg/kg/ twice daily orally for four weeks after diabetes induction. Blood sample and tissues of heart, pancreas and liver were collected at the end of experiment for histopathological examination. The obtained results showed that, STZ-induced diabetic rats exhibited a significant increase in serum glucose, totalcholesterol, triglyceride (TG), LDL cholesterol, creatine kinase (CK), lactate dehydrogenase (LDH) and interleukin 6 (IL6) with marked decrease in insulin and HDL-cholesterol. Treatment with *Murraya Koenigii (curry)* and *Moringa oleifera* were able to mitigate diabetic abnormalities through decreasing serum glucose, totalcholesterol, triglyceride (TG), LDL cholesterol, creatine kinase (CK), lactate dehydrogenase (LDH) and interleukin 6 (IL6) and increasing insulin and HDL-cholesterol. These results suggest that *Murraya Koenigii (curry)* and *Moringa oleifera* have hypoglycemic effect, hypolipidemic and increasing insulin secretion. so, decreasing complication and protect heart in diabetic rats by improving biochemical blood parameters.

KEYWORDS: *Murraya Koenigii (curry)*, *Moringa oleifera*, STZ, Diabetes, glucose, insulin, lipid profile, CK, LDH, IL6.

INTRODUCTION

Diabetes Mellitus is a class of metabolic disorders that comes from insulin defective actions, insulin secretions and often characterized by hyperglycemia. It has become one of most aggressively spreading lifestyle diseases. Diabetic patients are rest increase risks of peripheral arterial, atherosclerosis cardiovascular and cerebrovascular disease. Abnormal lipoprotein metabolism and hypertension are more common in the diabetic patients. Diabetes mellitus is an endocrine and metabolic disorder characterized by high glucose levels in the blood as a result of defects in the secretion and or action of insulin. Insulin is a hormone needed for the utilisation and storage of glucose in the body (**Mahan and Escott-Stump, 2008**). According to the World Health Organization (**WHO, 2015**), diabetes mellitus caused 1.5 million deaths in 2012 and more than 80% of diabetes mellitus deaths occur in low- and middle-income countries. Diabetes mellitus is also projected to be the leading cause of death in 2030.

Majority of diabetic cases comes under two broad and major pathogenic cases. First is **type 1** diabetes with destruction of beta cells that lead to absolute deficiency of insulin A and also called as immune-mediated diabetes and the second is **type 2** diabetes with the combination of insulin resistance action and an improper compensatory response of the insulin secretion **(Joseph *et al.*, 2011).**

Herbal and natural products with antioxidant capacity have been used for centuries in every culture throughout the world. Also, used to lower blood sugar level and for treatment of insulin resistance in diabetic patients **(Babu *etal*., 2012).**

So, in this study we used *Murraya koenijii* (Curry) and *moringa oleifera* to evaluate their biochemical effects on diabetic rats**.**

Murraya koenijii (Curry) leaves: Curry leaves essential oil corrects pathological and hyperglycemia abnormalities in the diabetic rats due to its antioxidant effect and restorative potential of redox homeostasis mechanism, excellent recovery of pancreatic tissue can be explained by positive effect of curry leaves oil on the production of insulin because of regenerative effect of the exocrine pancreatic cells **(Amin *et al*., 2013).** The major constituent responsible for the aroma and flavor has been reported as pinene, sabinene, caryophyllene,

cadinol and cadinene. The leaves have a slightly pungent, bitter and feebly acidic taste, **(Kale *et al*., 2014).**

Moringa oleifera can also be used to stabilize sugar levels and can stabilize arterial tension **(Bukar *et al*., 2010; Kasolo *et al*., 2010).** The leaves have also been found to possess antitumour, antipyretic, antiepileptic, antinflammatory, antiulcer, antispasmodic, diuretic, antihypertensive and antioxidant properties **(Bukar *et.* al., 2010*).***

MATERIALS AND METHODS

Experimental animals

Sixty white male albino rats weighting 150-200 gm were used in the experimental investigation of this study. The rats were obtained from the Laboratory Animals Research Center, Faculty of Veterinary Medicine, Benha University. Rats Animals were housed in separate metal cages, exposed to good ventilation, humidity and to a 12 hr light/dark cycle. Fresh and clean drinking water was supplied ad-libtium. Constant supplies of standard pellet diet, fresh and clean drinking water were supplied ad-libitum.The animals were left for 15 days for acclimatization prior to the beginning of the experiment, and kept at constant environmental and nutritional conditions throughout the period of the experiment.

Chemicals

- **STZ :(Streptozotozin)** was purchased from Sigma Aldrish.
- ***Murraya koenigii (curry)*** was purchased from Saudi Arabia.
- ***Moringa Oleifera*** was obtained from school of agriculture in Mansoura.

Diabetes induction

Rats were fasted for 18 hrs. The experimental induction of diabetes in male rats was induced by a single intraperetinoel (i.p) injection of **50** mg/kg body weight of Streptozotocin (STZ) freshly dissolved in citrate buffer, pH 4.5. Serum glucose estimations were undertaken periodically (days 0, 3, and 7). After week, STZ-treated rats were fasted for 12 h and blood samples were collected from the tail vein for glucose determination. Only those rats in diabetic group with blood glucose level higher than **250 mg/dl** were considered diabetic **(Ramanathan et al., 1999).**

NOTE: You must avoid light during preparation of STZ and you should make fresh preparation of STZ for each ten rats to keep its effect.

Aquous extract of curry leaves

For each leaf extract, 100 gm of shade dried leaves were ground in an electrical grinder and dissolved in 500 ml distilled water at 40-60^{o} C for 48 h. The mixture was left for 24 hrs with a magnetic stirrer at room temperature. The next day the mixture was strained out in a fine sieve and the crude extract was air evaporated for 3 days. The concentrated leaf extract of plant was then orally administered to the rats in 200 mg/kg body weight) using a syringe according to **(Gohil *et al*., 2010**, **Maha *etal*, 2013)**.

Aquous extract of moringa leaves

For each leaf extract, 100 gm of shade dried leaves were ground in an electrical grinder and dissolved in 500 ml distilled water at 40-60^{o} C for 48 h. The mixture was left for 24 hrs with a magnetic stirrer at room temperature. The next day the mixture was strained out in a fine sieve and the crude extract was air evaporated for 3 days. The concentrated leaf extract of plant was then orally administered to the rats in 200 mg/kg body weight) using a syringe according to **(Dolly *etal*, 2013)**.

***NOTE*:** We prepare the aquous extract of curry and moringa every week during four weeks to be fresh and keep its effects.

Experimental groups

Group 1 (Normal Control)

Composed of 15 male rats, rats were administered distilled water orally using a feeding cannula for study period. 0.01 M citrate buffer, pH 4.5, was injected intraperitoneally to mimic the STZ injections.

Group 2: (Diabetic Control)

Composed of 15 male rats. Diabetes was induced by a single STZ injection (50 mg/kg body wt, i.p., dissolved in 0.01 M citrate buffer, pH 4.5).

Group 3: (diabetic treated with curry)

Composed of 15 male rats, received aqueous extract of curry leaves at a dose **of 200 mg/kg/twice daily. for four weeks.**

Group 4: (diabetic treated with Moringa)

composed of 15 male rats, received aqueous extract of *Moringa oleifera* leaves at a dose of 200 mg/kg/ twice daily for four weeks.

SAMPLING

Blood sampling

Blood samples were collected after overnight fasting from all animal groups. Blood was collected from retero-orbital plexus of eyes in clean, dry screw capped tubes, the blood sample was allowed to coagulate at room temp for 30 mins, and centrifuged at 3000 r.p.m for 15 mins. The clean clear serum was aspirated by pasture pipette and received in dry aterile sample tube. Processed directly for glucose determination, then kept in deep freezing at – 20^{o}C until used for another biochemical analyses. All sera were analysed for the following parameters (glucose, insulin, lipid profile, CK, LDH and IL6).

Tissue specimens

Specimens of rats heart, pancreas and liver from scarifying animals will be histopathologically examined.

Biochemical analysis

Serum glucose was determined enzymatically according to the method described by **Trinder (1969).** Plasma insulin was determined according to **Findlay and Dillard (2007).** Total cholesterol in serum was determined according to the method described by **Allain et al., (1974).** Serum triglycerides were determined by enzymatic method which was described by **Fossati and Prencipe (1982).** Serum LDL was determined using the following equation : LDL-cholesterol = total cholesterol – triacylglycerol/5 – HDL-cholesterol according to **Falholt et al., (1973)**. Serum HDL was determined according to the method of **Burnstein et al., (1970).** CK was determined by ELISA kit of my biosource company catalog number **MBS722021.** LDH was determined by colorimetric assay kit of abcam company catalog number ab102526. Serum IL6 concentration was determined according to the method described by **(Chan and. Perlstein, 1987).**

Statistical analysis

All the data were expressed as means ±S.E. The statistical significance was evaluated by one-way analysis of variance (ANOVA) using SPSS, 18.0 software, 2011 and the individual comparisons were obtained by Duncan's multiple range test (DMRT). Values were considered statistically significant when $p<0.05$.

RESULTS

Effect of treatment with curry and moringa on some serum parameters in STZ-induced diabetic rats

The obtained results in table (1, 2 and 3) revealed that, a significant increase in serum glucose, total cholesterol, triglyceride, LDL, creatine kinase (CK), Lactate dehydrogenase(LDH) and interleukin6 (IL6) with marked decrease in insulin and HDL in diabetic control rats. Treatment with curry and moringa were able to mitigate diabetic abnormalities through decreasing serum glucose, total cholesterol, triglyceride, LDL, creatine kinase (CK), Lactate dehydrogenase(LDH) and interleukin6 (IL6) but higher than normal control and increasing insulin and HDL but less than normal control. These results revealed that treatment with curry and moringa act as hypoglycemic, effective in increasing insulin sensitivity and secretion, hypolipidemic and decreasing inflammation in diabetic rats by improving biochemical blood parameters.

Table (1): Effect of Curry and Moringa on the serum level of glucose and insulin in in STZ-induced diabetic rats (Mean±SE) after 4 weeks.

Groups	Glucose (mg/dl)	Insulin (ng/ml)
G1	85.33± 10.68 ^{d}	5.13 ± 0.26 ^{a}
G2	312.67± 17.14 ^{a}	0.84± 0.06 ^{d}
G3	126.67± 6.33 ^{c}	2.70 ± 0.12 ^{b}
G4	179.00± 10.02 ^{b}	1.80 ± 0.12 ^{c}

G1=normal control, G2=Diabetic control, G3=Diabetic+curry, G4=Diabetic+Moringa.

Means within the same column carrying different superscript letters are significantly different (P≤ 0.05).

Table (2): Effect of Curry and Moringa on the serum level of lipid profile in diabetic rats induced by STZ (Mean±SE) after 4 weeks.

Groups	Total cholesterol (mg/dl)	TG (mg/dl)	LDL (mg/dl)	HDL (mg/dl)
G1	**94.67± 1.45 ^{d}**	**42.67± 0.88 ^{d}**	**45.33± 0.88 ^{d}**	**45.00± 1.15 ^{a}**
G2	**125.67± 2.40 a**	**109.33± 1.45 ^{a}**	**73.27± 1.51 ^{a}**	**30.00± 0.87 ^{d}**
G3	**107.03± 1.32 ^{c}**	**62.67± 0.88 ^{c}**	**52.93± 1.16 ^{c}**	**39.57± 0.58 b**
G4	**114.00± 1.53 ^{b}**	**76.33± 0.88 ^{b}**	**62.33± 1.20 ^{b}**	**35.17± 0.73 c**

G1=normal control, G2=Diabetic control, G3=Diabetic+curry, G4=Diabetic+Moringa.

Means within the same column carrying different superscript letters are significantly different (P≤ 0.05).

Table (3): Effect of Curry and Moringa on the serum level of CK, LDH, IL6 in diabetic rats induced by STZ (Mean±SE) after 4 weeks.

Groups	CK (ng/ml)	LDH (mg/dl)	IL6 (pg/ml)
G1	26.82± 0.85 [d]	53.52± 0.65 [d]	175.00± 8.39 [d]
G2	121.19± 0.75 [a]	129.70 ±1.21[a]	490.67± 18.22 [a]
G3	57.31± 0.88 [c]	76.67± 0.88 [c]	316.00± 11.37 [b]
G4	81.25± 1.06 [b]	91.44± 0.95 [b]	381.00± 12.42 [c]

G1=normal control, G2=Diabetic control, G3=Diabetic+curry, G4=Diabetic+Moringa.

Means within the same column carrying different superscript letters are significantly different ($P \leq 0.05$).

Histopathological examination

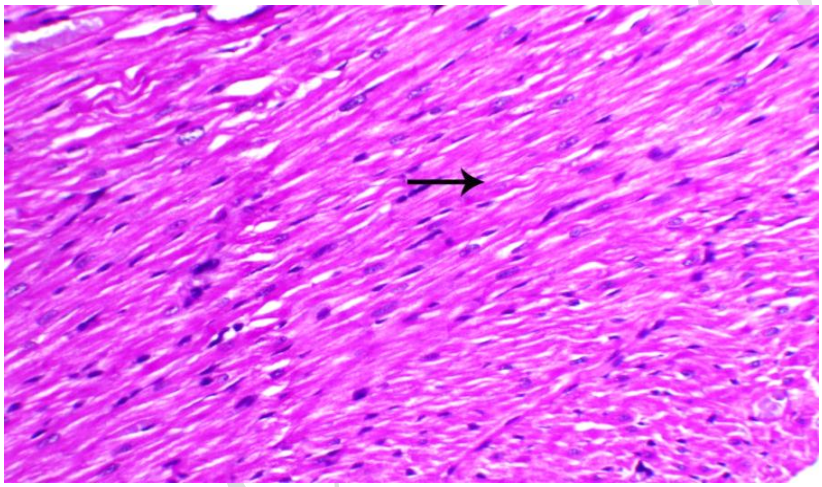

Fig. 1: Heart of normal control animal showing normal myocardial fibers (arrow), H&E, X200.

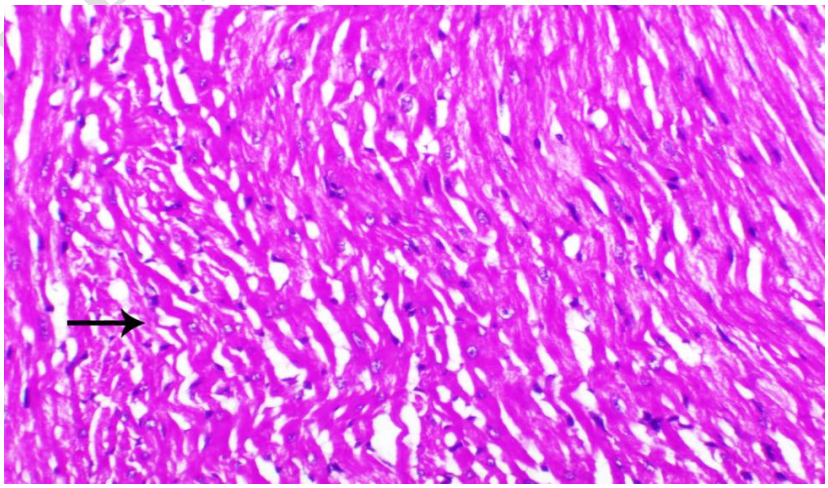

Fig. 2: Heart of diabetic control animal showing disorganization of myocardial fibers associated with myolysis (arrow), H&E, X200.

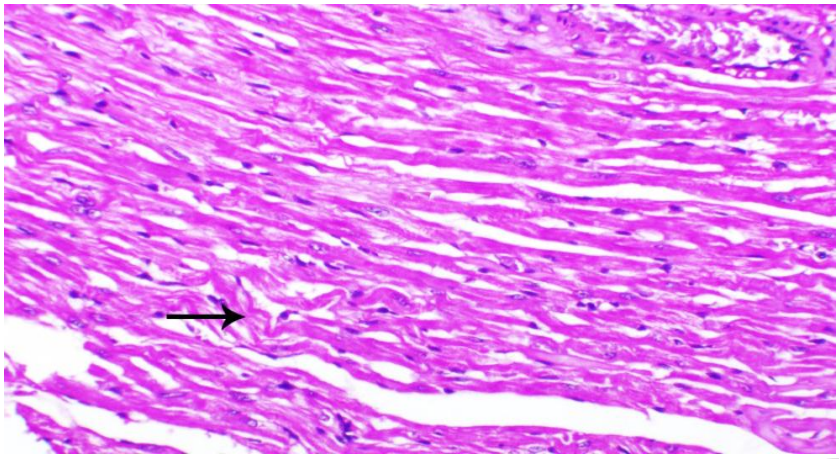

Fig. 3: Heart of diseased animal treated with curry showing mild degree of myolysis (arrow), H & E, X200.

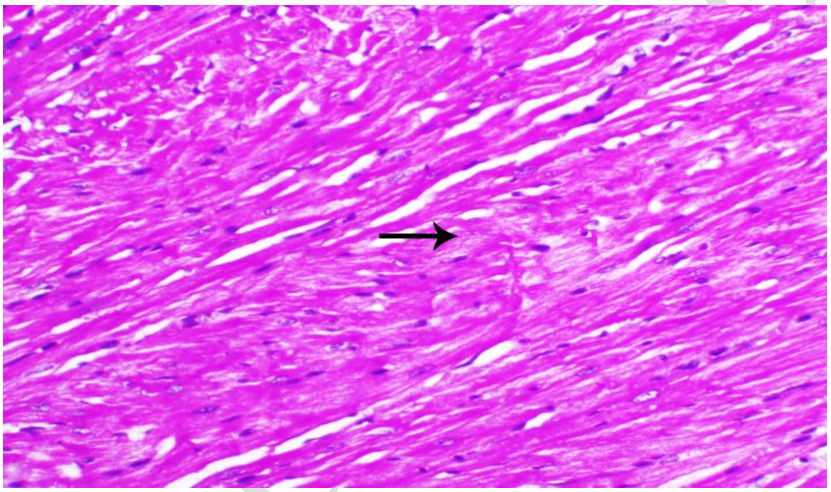

Fig. 4: Heart of diseased animal treated with moringa showing mild degree of myolysis (arrow), H&E, X200.

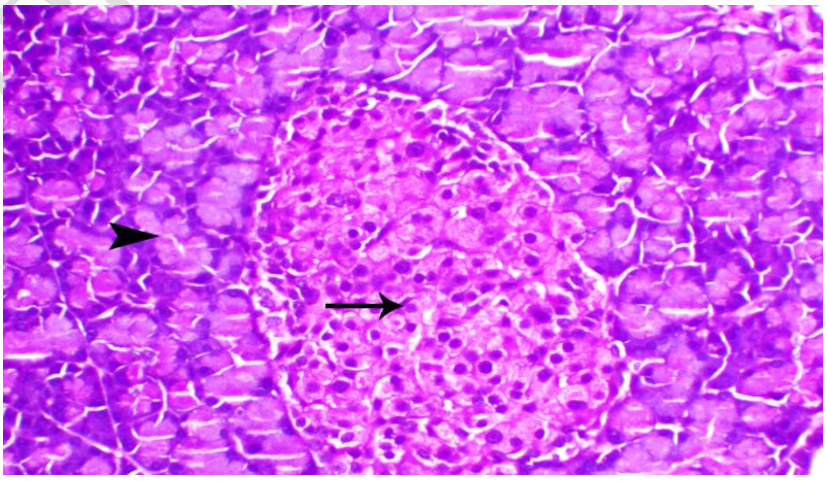

Fig. 5: Pancreas of normal control animal showing normal pancreatic acini (arrowhead) and islets of Langerhans (arrow), H&E, X200.

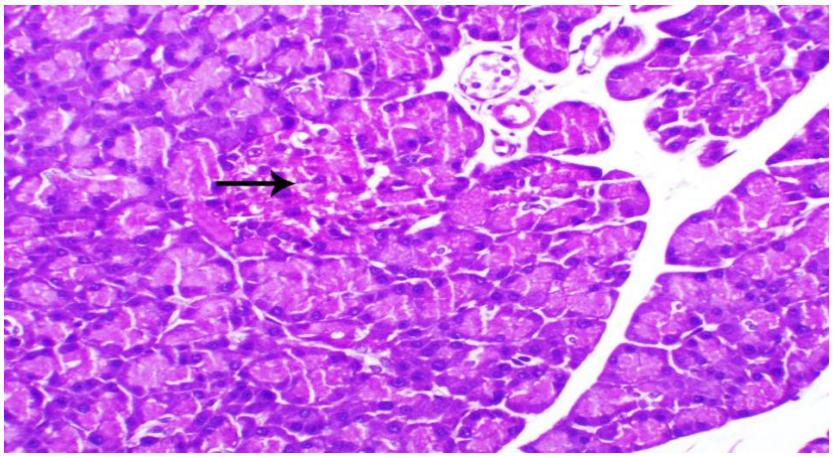

Fig. 6: Pancreas of diabetic control animal showing marked decrease of size and cellular components of the endocrine portion (islets of Langerhans, arrow), H&E,X200.

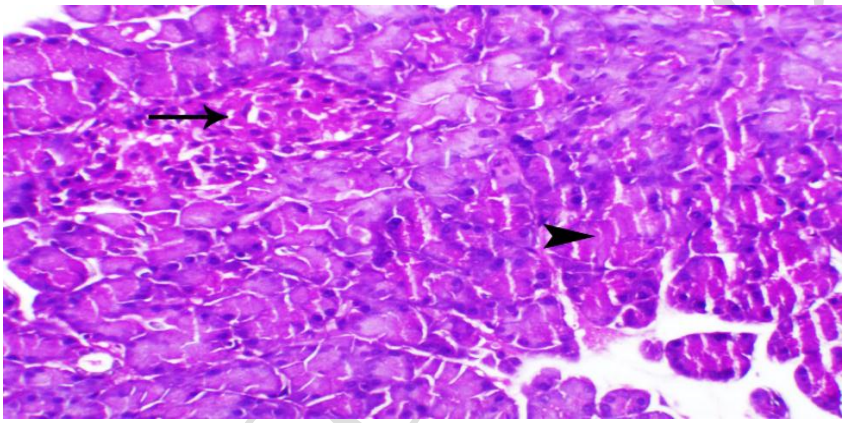

Fig. 7: Pancreas of diseased animal treated with curry showing mild degenerative changes within the exocrine portion (acini, arrowhead) and the endocrine portion (arrow), H&E,X200.

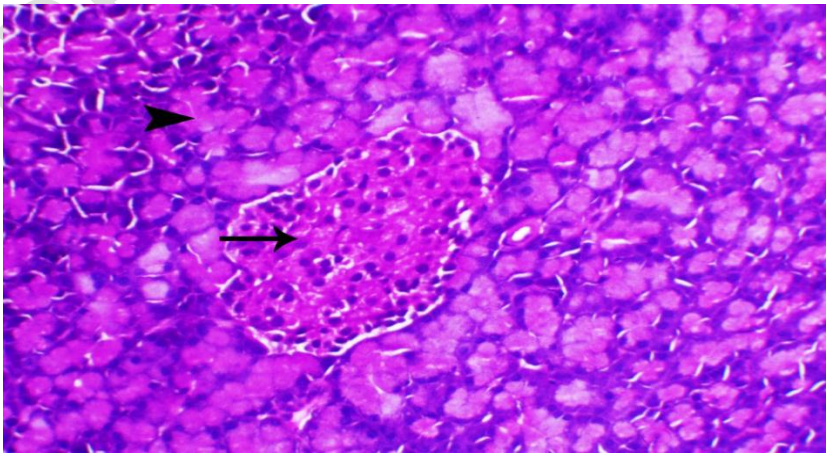

Fig. 8: Pancreas of diseased animal treated with moringa showing mild degenerative changes within the exocrine portion (acini, arrowhead) and the endocrine portion (arrow), H&E,X200.

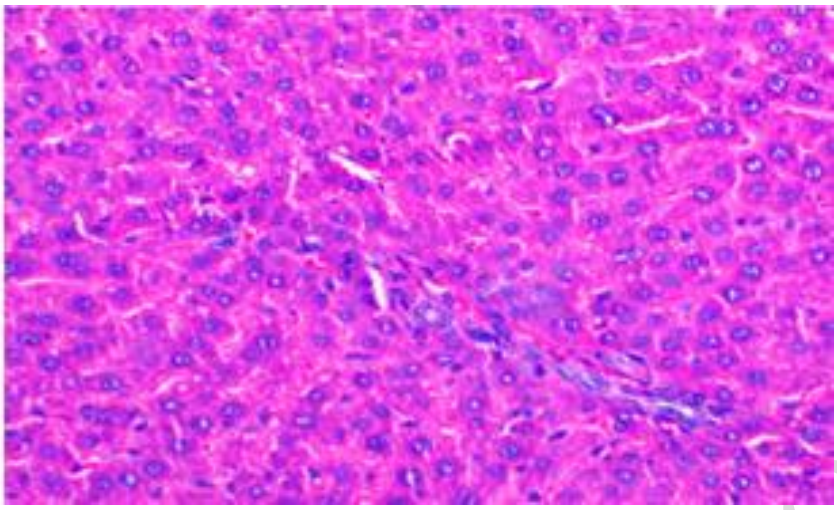

Fig. 9. Liver of normal control animal showing normal liver structures, H&E, X200.

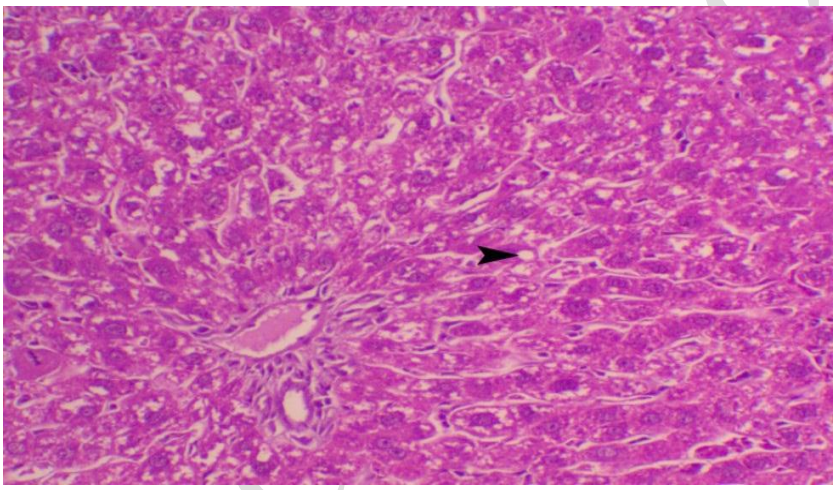

Fig. 10. Liver of diabetic control showing marked vacuolation of hepatocytes within the periportal area mostly of fatty degeneration type (arrowhead), H&E, X200.

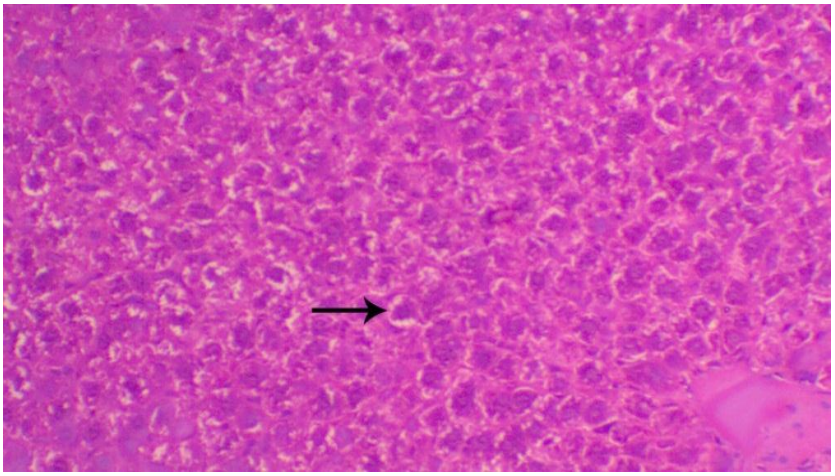

Fig. 11. Liver of STZ rats treated by curry showing mild vacuolation of hepatocytes (arrow), H&E, X200.

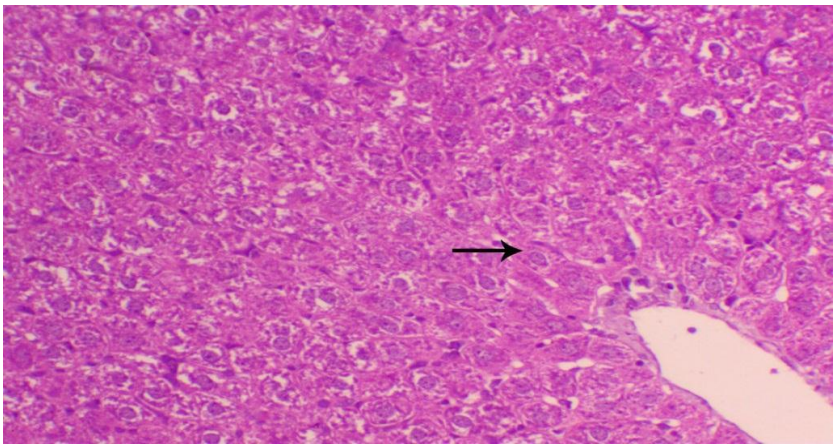

Fig. 12. Liver of STZ rats treated by moringa showing mild degree of swelling and vacuolar degeneration of hepatocytes (arrow), H&E, X200.

DISCUSSION

In this study, rats treated with STZ showed a significant increase in serum glucose and asignificant decrease in serum insulin. It was reported that induction of diabetes using streptozotocin in rats of the positive control group caused severe health problems illustrated in the increase in serum glucose which is consistent with previous findings (**Szkudelski, 2001)**. This is due to streptozotocin are toxic glucose analogues that preferentially accumulate in pancreatic beta cells via the GLUT2 glucose transporter. streptozotocin is split into its glucose and methylnitrosourea moiety. Owing to its alkylating properties, the latter modifies biological macromolecules, fragments DNA and destroys the beta cells, causing a state of insulin-dependent diabetes. The targeting of mitochondrial DNA, thereby impairing the signalling function of beta cell mitochondrial metabolism, also explains how streptozotocin is able to inhibit glucose-induced insulin secretion (**Lenzen, 2008**). streptozotocin-induced depletion of NAD^+ may result in the inhibition of insulin biosynthesis and secretion (**Strandell *etal*, 1988**). Later, inhibition of glucose-induced and amino acid-induced insulin secretion **(Eizirik *etal*, 1988)** as a result of mitochondrial enzyme dysfunction (**Rasschaert *etal*, 1992**) and damage to the mitochondrial genome become apparent **(Eizirik *etal*, 1991).** In our study treatment with curry (MK) in G3 significantly decrease serum glucose level. Similar results was reported by **Sucheta and Kavitha, 2013** who found that curry leaf powder had the property to decrease the blood glucose load and is the dietary adjunct in the management of Type 2 Diabetes. Also, by (**Venuthan *et al*, 2005) and** There was a significant blood glucose lowering effect in diabetic treated rats as compared to normal controls; the maximum fall of 85% for the rats treated with MK-200 mg/kg on the 30th day (**Imad *etal*, 2017).** The mode of action Murraya

koenigii (MK) has been suggested to be either due to increased glycogenesis or decreased glycogenolysis or gluconeogenesis and/or due to insulin secretogogue effect of MK, which causes an increased glucose uptake and its consumption by cells **(Vinuthan *etal,* 2004).** The characterization of the active principle responsible for the antihyperglycemic activity of MK has not yet been elucidated. However, "carbazole alkaloid" which has anti-hyperglycaemic and antihyperlipidaemic activity, has been found in the leaves of MK (**Yankuzo *etal,* 2011). Also,** These results suggest that G4 treated with moringa may be a potential agent in the treatment of diabetes and are agreed with the observations that suggest the beneficial effects of *Moringa oleifera* supplementation on diabetes (**Anthanont *etal,* 2016**). There is evidence to suggest that membrane environment may have some effect on insulin sensitivity and insulin-stimulated glucose uptake **(Storlien *etal,* 1991**) and Na, K-ATPase activity (**Pan *etal,*1994***)*. Membrane environment can also be modified by diet and a number of therapeutic diets are used It was observed Altered lipid environment and protein expression were the most likely explanations for the decrease in Na, K-ATPase activity observed in cardiac membranes (**Gerbi**1 ***etal,*** **1997**). Glucose sensing requires oxidative mitochondrial metabolism, leading to the generation of ATP. This increases the ratio of ATP to adenosine diphosphate (ADP) in the β cell, which then initiates the following chain of events: inhibition of the cell's ATP/ADP-regulated potassium channel (KATP), plasma membrane depolarization, opening of a voltage-gated calcium channel, calcium influx, and secretion of insulin. Although insulin secretion is also modulated by a number of stimuli that operate outside this pathway, it is clear that oxidative mitochondrial metabolism is central to glucose stimulated insulin secretion (**Maechler and Wollheim, 2001**).

In our study, there are asignificant increase in total cholesterol, TG, LDL and asignificant decrease in HDL in rats treated with STZ in G2. Some research articles indicated that diabetes mellitus is closely associated with dyslipidemia in both IDDM and NIDDM. The hypertriglyceridemia may be due to higher rates of production of triglyceride rich VLDL by the liver (**Nikkila and Kekki (1973)** and to decreased removal of TG by peripheral tissues-primarily adipose tissue and muscle. Insulin deficiency leads to high TG production and subsequent high packaging in VLDL. Several studies using radioactive substrates to trace the metabolism of plasma VLDL are consistent with their simultaneous overproduction and reduced clearance as the common etiologic mechanism for hypertriglyceridemia in poorly controlled IDDM **(Nikkila etal, 1977).** LDL production rates are reported to be elevated in IDDM but return to normal after insulin infusion (**Howard, 1987)**. It may be due to increased

synthesis of VLDL or impaired removal of VLDL remnant. Impaired receptor mediated clearance of LDL has also been postulated (**Esko *etal*, 1978)**. Lower HDL-C in diabetes may be due to reduced Lipoprotein Lipase activity. The activity of cholesterol ester transfer protein is increased in IDDM **(Nikkila *etal*, 1977)**. The main 'anti-atherogenic' lipoprotein HDL is involved in the transportation of cholesterol from peripheral tissues into liver and thereby acts as a protective factor against coronary heart disease. STZ decreases HDL-Cholesterol level and are major factor for coronary heart disease which is one of complication of diabetes (**Ananthan *etal*, 2003).** It is mandatory to treat this dyslipidemia to prevent adverse lipemic status and long term complications to ensure a healthy and happy life inspite of diabetic dyslipidemia (**Dr. Bhagyashree, 2017). G3, G4 treated with curry and moringa showed** antihyper lipidaemic activity. lines of evidence showed that The characterization of the active principle responsible for the antihyperglycemic activity of *Murraya koenigii* (curry) has not yet been elucidated. However, "carbazole alkaloid" which has anti-hyperglycaemic and antihyperlipidaemic activity, has been found in the leaves of MK (curry) (**Yankuzo *etal*, 2011). Also, it was revealed by astudy that** *MK (*possess a potent antihyperglycemic and hypolipidemic effect, which may be due to the presence of antioxidants such as carbazole alkaloids and polyphenols (**Maha *etal*, 2013).** Decrease in LDL-C was observed in diabetic animals with the administration of moringa leaf extract. Insulin is responsible to increase receptor-mediated removal of LDL-C from the body and hence decreased activity of insulin during diabetes leads to increased level of serum LDL-C and consequently hypercholesterolemia. Therefore, significant control in the serum lipid level in MO treated diabetic rats might be due to the insulinotropic action upon MO administration (**Kim and Kim, 2012)**. It was indicated, the increase in the HDL concentration in moringa leaf extract, which contains cardio protective potential against STZ induced diabetic rats (**Nandave etal, 2009)**. The biological activity of *Moringa oleifera* is not limited to the antioxidant capacity. In fact, other important biological activities such as hypolipidaemic, antiatherosclerotic, and anticarcinogenic activities of *M. oleifera* leaves and seeds have been reported (**Al-Malki and El Rabey, 2015).**

In this present study, there are asignificant increase in creatine kinase in diabetic rats treated with STZ in G2. Similar results demonstrate that the activity of the enzyme creatine kinase is significantly higher in patients with diagnosis of diabetes mellitus type I and diabetes mellitus type II, when compared to activity of the enzyme observed in control population. (**Adlija *etal*, 2006).** In diabetes in general, the alterations in glucose, lipid and

protein metabolism are present and especially evident at the level of muscle cells (miocytes). Due do the disease, the utilization of glucose is decreased, phosphorylation of glucose is altered, synthesis of glycogen reduced, glycolysis suppressed. Due to smaller capacity of glycolysis, concentrations of oxal acetate and pyruvate are diminished also. Capacity of Krebs cycle is reduced as well as capacity of oxidative phosphorylation and respiratory chain. ATP in muscle cells are diminished. Through stimulated glycolysis and further beta oxidation of fatty acids, organism tries to fight these processes. Due to the shortage of ATP, synthesis of creatine phosphate is decreased and the possibility of resynthesis of ATP from ADP is diminished. Sometimes, lack of synthesis of ATP can lead to total inhibition of creatine kinase activity. Therefore, it can be assumed that one of the potential mechanisms explaining observed small increase in creatine kinase activity in diabetics could be related to energy shortage (metabolic pathways leading to energy production are supressed, creatine phosphate is missing and due to potential inhibition of creatine kinase activated by AMP activated protein kinase), all of these factors being necessary for normal functioning of muscle cells. Due to these events, enzyme is leaking out of cytoplasm into blood. Evident positive correlation between concentration of glucose and the activity of creatine kinase can be easily attributed for. With higher concentration of glucose, disease is obviously not well controlled, uptake of glucose by the cells is lower, and the degree of metabolic disturbance higher leading to lower energy supply and therefore to a high degree of cell destruction. Since in this case one is speaking of autoimmune disease, which is accompanied by the appearance of macro CK type 1 activity, this can not be neglected as a cause of increased activity of creatine kinase in patients with this disease(**Adlija *etal*, 2006)**.

Also, there are asignificant increase in lactate dehydrogenase (LDH) in diabetic rats treated with STZ in G2. It was indicated that plasma LDH levels in rabbits with diabetes mellitus were significantly higher when compared with the controls(**Ismail and Esref, 2002)**. Diabetes is accepted to be the commonest endocrine disease, which are multi-systemic disorders resulting from deficiency in the secretion or action of the pancreatic hormone insulin, which in turn produces profound abnormalities of metabolism (**Leninger, 1982**). Since muscle and liver dysfunction is frequently associated with diabetes mellitus, many clinical reports have indicated that serum enzyme activities derived from the muscle and liver such as creatine phosphokinase (CPK), LDH and GOT are elevated. Increased serum or plasma enzyme levels were considered important evidence supporting the diagnosis of diabetes mellitus (**Stewart, 1991).** DM induce hepatic malfunctioning i.e liver was necrotized

in diabetic patients due to which percentage of LDH-C enzyme increases in plasma because of the spillage of these enzymes into the blood stream from liver cytosol, which indicates the hepatotoxic impact of STZ. Administration of MO for 30 days might restore the action of LDH-C enzyme to their normal state and inhibits the liver damage induced by STZ (**Mansour *etal*, 2002).** There isn't any previous study reveal the effect of curry extract on lactate dehydrogease**.** It was observed also by **Muhammad *etal*, 2017** the significant decrease in LDH in group treated with moringa.

In our study, there are asignificant increase in interleukin6 (IL6) in diabetic rats treated with STZ in G2. IL-6 has been identified as an independent predictor of T2DM and associated cardiovascular events (**Spranger *et al*., 2003; Lowe *et al*., 2014**). Adipocytes and macrophages residing in adipose tissue are the major sources for the elevated plasma IL-6 concentration up to 2–3 pg·mL−1 in patients with obesity and T2DM (**Spranger *et al*., 2003**). Nevertheless, the existing evidence is not enough to establish a causal association between IL-6 levels and the progression to metabolic and cardiovascular disorders. Due to its pleiotropic actions in various tissues and organs, the exact role of IL-6 in the pathogenesis of diabetes must be examined carefully in a cell- and tissue-specific manner, but allowing for the possibility of crosstalk between affected tissues and organs. IL-6 is a pleiotropic cytokine that participates in normal functions of the immune system, haematopoiesis, metabolism, as well as in the pathogenesis of metabolic and cardiovascular diseases. The role of IL-6 is not restricted to the immune system, as it is also involved in neuronal differentiation and regeneration, liver regeneration and regulation of metabolic process. Although IL-6 is mostly regarded as a pro-inflammatory cytokine that promotes inflammation under various pathological conditions, its anti-inflammatory and regenerative properties have been increasingly recognized (**Scheller *et al*., 2011**). On the contrary, a significant elevation of plasma IL-6 level has been characterized as a marker for metabolic disorder and cardiovascular disease. IL-6 is normally not expressed constitutively, but its expression is extensively induced by a spectrum of stimuli such as viral and bacterial infection, pro-inflammatory cytokines (TNF-α and IL-1), angiotensin II, oxidative stress and physical exercise. In addition, the anti-inflammatory and immunosuppressive role of IL-6 in both local and systemic inflammatory responses has been widely accepted (**Kristiansen and Mandrup-Poulsen, 2005).** Moreover, IL-6 is elevated in concert with a number of anti-inflammatory cytokines such as IL-10, and may regulate the level of pro-inflammatory cytokines (**Xing *et al*., 1998).** It was shown that the anti inflammatory effect of *Murraya Koenigii by*

(Yedukondalu etal, 2016). It was shown that Hydroethanolic extract of *M. oleifera* flower significantly suppressed the secretion and expression of interleukin- (IL-) 6, (**Woan *etal*, 2015).** In our study, we observe in G3 treated with curry and G4 treated with moringa asignificant decrease in creatine kinase and lactate dehydrogenase and interleukin 6. So, it can be concluded that curry and moringa extract possesses significant cardioprotective effect, which may be attributed to its antioxidant, antiperoxidative, and myocardial preservative properties. This is assured by histopathological investigation, the G3 treated with curry and G4 treated with moringa can ameliorate the bad effect that caused by STZ induced diabetes mellitus in heart, pancreas and liver tissue. Liver of diabetic rats treated with Murraya (curry) (50 ml/kg/bw) extract showed a recovery from the diabetic condition **(Vijayanand, 2015).** Curry leaves protein antioxidant is an effective antioxidant in preventing membrane damage and associated functions mediated by reactive oxygen species. It can be further developed as an effective bioprotective antioxidant agent to cellular components (**Mylarappa *etal*, 2016)**. It was shown that aqueous extract of *Moringa oleifera* leaf protects pancreas against ROS mediated damage by enhancing cellular antioxidant defenses and minimizing hyperglycemia in STZ-induced diabetes, which might be due to the glucose uptake enhancement in skeletal muscle, insulin secretion stimulation, and alpha amylase and alpha-glucosidase inhibition. **(Khan *etal*, 2017).**

CONCLUSION

The present study demonstrated that aquous extract of leaves of curry and moringa have hypoglycemic, hypolipedemic and anti-inflammatory effects and improve insulin secretion in STZ-induced diabetic rats and ameliorate the bad effects caused by oxidative stress due to antioxidant properties.

ACKNOWLEDGEMENTS

The author thanks Drs. Yakout ELsenosi, Omayma Ragab, Afaf ELdesoky for their critical review of the manu script. The work was supported by grants from Faculty of Vet., Medicine Benha University, Egypt.

REFERENCES

1. Adlija JIvrić-Čaušević, Maja Malenica, Tanja Dujić. Creatine Kinase activity in Patients with Diabetes Mellitus Type I and Type II.(Bosnian journal of basic medical sciences, 2006; 6(3): 5-9.
2. Allain CC, Poon LS, Chan CS, Richmond W, Fu PC. Enzymatic determination of total serum cholesterol. Clin Chem., 1974; 20(4): 470-5.
3. Al-Malki, A. L. and. El Rabey, H. A Antidiabetic effect of low doses of *Moringa oleifera* Lam. seeds on streptozotocin induced diabetes and diabetic nephropathy in male rats," *BioMed Research International*, 2015, Article ID 381040, 13 pages.
4. Amin, M.E., P. Virk, M.A.R. Elobeid, Z.M. Almarhoon, Z.K. Hassan, S.A. Omer, N.M. Merghani, M.H. Daghestani and E.M.A. Olayan. Anti-diabetic effect of *Murraya Koenijii* (L) and Olea europaea (L) leaf extracts on streptozoticin induced diabetic rats. Pak. J. Pharma. Sci., 2013; 26: 359-365.
5. Ananthan R, Latha M, Ramkumar K, Pari L, Baskar C, Bai V. *et al.* Effect of *Gymnema montanum* leaves on serum and tissue lipids in alloxan diabetic rats. Experimental Diabetes Research., 2003; 4: 183-189.
6. Anthanont, P; Lumlerdkij, N; Akarasereenont, P; Vannasaeng, S and Sriwijitkamol, A. *Moringa oleifera* leaf increases insulin secretion after single dose administration: a preliminary study in healthy subjects," *Journal of the Medical Association of Jailand*, 2016; 99(3): 308–313.
7. Babu, B.V., V.K. Chaturvedi and R.M. Uppu. Herbs in the management of hyperglycemia in diabetes. Importance of screening methods in the identification of phyto anti-hyperglycemic principles. J. Diabetic Metabol, 2012; 3: 7-16.
8. Bukar, A. Uba and T.I. Oyeyi, Antimicrobial profile of *Moringa oleifera* Lam. Extracts against some food-borne microorganisms, *Bayero Journal of Pure Applied Sciences*, 2010; 3(1): 43-48.
9. Burstein M, Scholnick HR, Morfin R. Rapid method for the isolation of lipoproteins from human serum by precipitation with polyanions. J Lipid Res., 1970; 583-95.
10. Chan, D and. Perlstein, M. Immunoassay: A Practical Guide Paperback. Clin Chem. 1987; (10): 2077-80.
11. Dolly Jaiswal, Prashant Kumar Rai, Shikha Mehta, Sanjukta Chatterji, Surekha Shukla, Devendra Kumar Rai, Gaurav Sharma, Bechan Sharma, Shahidul khair, Geeta Watal. Role of Moringa oleifera in regulation of diabetes-induced oxidative stress. Asian Pacific Journal of Tropical Medicine, 2013; 426-432.

12. Dr. Bhagyashree K Bhuyar. Lipid profile in Diabetes Mellitus. International Journal of Biotechnology and Biochemistry, 2017; 123-131.
13. Eizirik DL, Sandler S, Ahnström G, Welsh M. Exposure of pancreatic islets to different alkylating agents decreases mitochondrial DNA content but only streptozotocin induces long-lasting functional impairment of B-cells. Biochem Pharmacol, 1991; 42: 2275–2282.
14. Eizirik DL, Sandler S, Sener A, Malaisse WJ. Defective catabolism of d-glucose and l-glutamine in mouse pancreatic islets maintained in culture after streptozotocin exposure. Endocrinology, 1988; 123: 1001–1007.
15. Esko, A Nikkila etal. Serum lipids and lipoproteins in insulin treated diabetes, Nov-1978; 27: 1078-86.
16. Falholt K, Lund B, Falholt W. An easy colorimetric micromethod for routine determination of free fatty acids in plasma. (Clin Chim Acta, Jun 28, 1973; 46(2): 105-11.
17. Findlay, J and. Dillard, R (2007). Appropriate Calibration Curve Fitting in Ligand Binding Assays.
18. Fossati P, Prencipe L. (1982). Serum triglycerides determined colorimetrically with an enzyme that produces hydrogen peroxide.
19. Gerbi1, A; Barbey, O; Raccah1, D; Coste, T; Jamme, I; Nouvelot, A. Ouafik, L;.Le´vy, S; Vague, P; Maixent, J.-M. Alteration of Na, K-ATPase isoenzymes in diabetic cardiomyopathy: effect of dietary supplementation with fish oil (n-3 fatty acids) in rats. Diabetologia, 1997; 40: 496–505.
20. Gohil T, PathakN, N Jivani, Devmurari V and Patel J. Treatment with extracts of *Eugenia jambolana*seed and *Aegle marmelos* leaf extracts prevents hyperglycemia and hyperlipidemia in alloxan induced diabetic rats. *AJPP*, 2010; 4: 270-275.
21. Howard. BV. lipoprotein metabolism in diabetes msllitus. J Lipid Res., 1987; 28: 613-28.
22. Imad. M, Rahajoe. I, Muhammad. H, Anil. K, Khalid. S. The Antidiabetic Activity of Curry Leaves "*Murraya Koenigii"* on the Glucose Levels, Kidneys, and Islets of Langerhans of Rats with Streptozotocin Induced Diabetes. Makara J. Health Res., 2017; 21(2): 54-60.
23. Ismail. Celik, Esref Yegin. Effect of Experimental Diabetes Mellitus on Plasma Lactate Dehydrogenase and Glutamic Oxaloacetic Transaminase Levels in Rabbits. Turk J Biol., 2002; 26-151-154.
24. Joseph, B., and D. Jini. Insight into the hypoglycemic effect of traditional Indian herbs used in the treatment of diabetes. Res. J. Med. Plant, 2011; 5: 352-376.

25. Kale, N.V. and G.S. More. Biochemical, antimicrobial and organoleptic analysis of curry leaf. Plant Sciences Feed, 2014; 4: 6-9.
26. Kasolo, J.N. Bimenya, G.S,. Ojok, J. Ochieng and. Ogwal-Okeng, J.W. Phytochemicals and uses of *Moringa oleifera* Leaves in Ugandan rural communities, *Journal of Medicinal Plant Research*, 2010; 4(9): 753-757.
27. Khan, W, Parveen, K. Chester, S. Parveen, and Ahmad, S. Hypoglycemic potential of aqueous extract of *Moringa oleifera* leaf and *in vivo* GC-MS metabolomics," *Frontiers in Pharmacology*, 2017; 8: 577.
28. Kim MJ, Kim HK. Insulinotrophic and hypolipidemic effects of *Ecklonia cava* in streptozotocin-induced diabetic mice. Asian Pac. J. of Trop. Med., 2012; 374-379.
29. Kristiansen OP, Mandrup-Poulsen T. Interleukin-6 and diabetes: the good, the bad, or the indifferent? Diabetes, 2005; 54(2): S114–S124.
30. Leninger, A.L. Principles of Biochemistry, New York, Worth Publishers Inc., 1982; 712-14.
31. Lenzen, S. The mechanisms of alloxan- and streptozotocin-induced diabetes. springer link, 2008; 216–226.
32. Lowe G, Woodward M, Hillis G, Rumley A, Li Q, Harrap S *et al*. Circulating Inflammatory markers and the risk of vascular complications and mortality in people with type 2 diabetes and cardiovascular disease or risk factors: the ADVANCE study. Diabetes, 2014; 63: 1115–1123.
33. Maechler, P; Wollheim, C. B. Nature, 2001; 414: 807.
34. Maha El Amin, Promy Virk, Mai Abdel Rahman Elobeid, Zainab Mohammed Almarhoon, Zeinab Korany Hassan, Sawsan Ali Omer, Nada Mohammed Merghani, Maha Hassan Daghestani and Ebtisam Mohammed Al Olayan. Anti-diabetic effect of *Murraya koenigii* (L) and *Olea europaea*(L) leaf extracts on streptozotocin induced diabetic rats. Pak. J. Pharm. Sci., 2013; 359-365.
35. Maha El Amin., Promy Virk, Mai AbdelRahman Elobeid, Zainab Mohammed Almarhoon, Zeinab Korany Hassan, Sawsan Ali Omer, Nada Mohammed Merghani, Maha Hassan Daghestani and Ebtisam Mohammed Al Olayan. 2013 Anti-diabetic effect of *Murraya koenigii* (L) and *Olea europaea* (L) leaf extracts on streptozotocin induced diabetic rats.
36. Mahan KL & Escott-Stump SR. Krause's Food and Nutrition Therapy (12th ed.). Saunders Elsevier, Missouri, 2008; 766-769.

37. Mansour HA, Newairy AA, Yousef MI, Sheweita SA. Biochemical study on the effects of some Egyptian herbs in alloxan-induced diabetic rats. *Toxicology.*, 2002; 170: 221-228.
38. Muhammad N, Asif A, Ahmad A, Khawar M. Effect of different concentration of *Moringa oleifera* leaves on the serum profile and organs of the induced diabetic rats. International Journal of Advanced Educational Research. Page No. 24-30. *Murraya koenigii* on Alloxan Induced Diabetic rats. ISSN: 0975-9492.
39. Mylarappa B Ningappa, Dinesha Ramadas1, Dinesha Nanjegowda, Kiruthika Balasubramanian1, Khusdeep Chahal, Sachin Patil and Leela Srinivas. Cytoprotective Properties of Antioxidant Protein from Curry leaves (Murraya koenigii L.) against Oxidative Stress Induced Damage in Human Erythrocytes. Med chem., 2016. ISSN: 2161-0444.
40. Nandave M, Ojha SK, Joshi S, Kumari S, Arya DS. "*Moringa oleifera* leaf extract prevents isoproterenol-induced myocardial damage in rats: Evidence for an antioxidant, antiperoxidative, and cardioprotective intervention," J. Med. Food, 2009; 12(1): 47-55.
41. Nikkila and Kekki. Plasma triglyceride transport kinetics in diabetes mellitus. Metabolism., 1973; 22: 1-22.
42. Nikkila et al. Diabetes., 1977; 26: 11-24.
43. Pan DA, Hulbert AJ, Storlien LH. Dietary fats, membrane phospholipids and obesity. J Nutr., 1994; 124: 1555–1565.
44. Ramanathan, M.; Jaiswal, A.K. and Bhattacharya, S.K. Superoxide dismutase, catalase jand glutahiones peroxidase activities in the brain of streptozotocin induced diabetic rats. Indian. J. Exp. Biol., 1999; 37: 182-183.
45. Rasschaert J, Eizirik DL, Malaisse WJ. Long term in vitro effects of streptozotocin, interleukin-1, and high glucose concentration on the activity of mitochondrial dehydrogenases and the secretion of insulin in pancreatic islets. Endocrinology, 1992; 130: 3522–3528.
46. Scheller J, Chalaris A, Schmidt-Arras D, Rose-John S. The pro- and anti-inflammatory properties of the cytokine interleukin-6. Biochim Biophys Acta, 2011; 1813: 878–888.
47. Spranger J, Kroke A, Mohlig M, Hoffmann K, Bergmann MM, Ristow M *et al*. Inflammatory cytokines and the risk to develop type 2 diabetes: results of the prospective population-based European Prospective Investigation into Cancer and Nutrition (EPIC)-Potsdam Study. Diabetes, 2003; 52: 812–817.
48. Stewart, M. Animal Physiology, Kent UK, Holder and Stough Ltd., 1991; 313-7.

49. Storlien LH, Jenkins AB, Chisholm DJ et al. Influence of dietary fat composition on development of insulin resistance in rats. Diabetes, 1991; 40: 280–289.
50. Strandell E, Eizirik DL, Korsgren O, Sandler S. Functional characteristics of cultured mouse pancreatic islets following exposure to different streptozotocin concentrations. Mol Cell Endocrinol, 1988; 59: 83–91.
51. Sucheta, L and Kavitha, R, Hypoglycemic effect of *murraya koenigii* (curry leaf) in type 2 diabetes mellitus. International journal of food and nutritional sciences, 2013. *Issn* 2320 –7876.
52. Szkudelski, T. The mechanism of alloxan and streptozotocin action in B cells of the rat pancreas," *Physiological Research*, 2001; 50(6): 537–546.
53. Trinder, P. An. Clin. Biochem, 1969; 6: 24.
54. Venuthan, M K, Girish Kumar V, Ravindra J P, Jayaprakash Narayana K. Ind. J. Physio. Pharmacol, 2005; 49(2): 241-242.
55. Vijayanand, S. Evaluation of Antidiabetic activity of *Murraya koenigii* on Alloxan Induced Diabetic rats, 2015. ISSN: 0975-9492.
56. Vinuthan MK, Girish KV, Ravindra JP, Jayaprakash, Narayana K. Effect of extracts of *murraya koenigii* leaves on the levels of blood glucose and plasma insulin in alloxan-induced diabetic rats. *Indian J Physiol Pharmacol*., 2004; 48: 348-52.
57. Woan Sean Tan, Palanisamy Arulselvan, Govindarajan Karthivashan, and Sharida Fakurazi. *Moringa oleifera* Flower Extract Suppresses the Activation of Inflammatory Mediators in Lipopolysaccharide-Stimulated RAW 264.7 Macrophages via NF-*κ*B Pathway., Article ID 720171, 2015; 2015; 11.
58. World Health Organization [WHO](2015). Diabetes. From http*://www.who.int/mediacentre/factsheets/fs312/en/* [Retrieved 4 October.
59. Xing Z, Gauldie J, Cox G, Baumann H, Jordana M, Lei XF *et al*. IL-6 is an antiinflammatory cytokine required for controlling local or systemic acute inflammatory responses. J Clin Invest, 1998; 101: 311–320.
60. Yankuzo MH, Ahmed QU, Santosa RI, Akter SF, Talib NA. Beneficial effect of the leaves of *Murraya koenigii* (Linn.) Spreng (Rutaceae) on diabetes-induced renal damage *in vivo*. *J Ethnopharmacology*., 2011; 35: 88-94.
61. Yankuzo MH, Ahmed QU, Santosa RI, Akter SF, Talib NA. Beneficial effect of the leaves of *Murraya koenigii* (Linn.) Spreng (Rutaceae) on diabetes-induced renal damage *in vivo*. *J Ethnopharmacology*., 2011; 35: 88-94.

62. Yedukondalu Nalli, Vidushi Khajuria, Shilpa Gupta, Palak Arora, Syed Riyaz-Ul-Hassan, Zabeer Ahmeda, and Asif Ali. Four new carbazole alkaloids from Murraya koenigii that display anti-inflammatory and anti-microbial activities. Cite this: Org. Biomol, 2016; 14: 3322.