Antidiabetic, antihyperlipidemic and histopathological analysis of *Panax quinquefolium* extract on alloxan induced diabetic rats

M.A.Nagy
Chemistry Department, Faculty of Science, Beni-Suef University, (EGYPT)
E-mail : nagy_bio@yahoo.com

ABSTRACT

Objective: To investigate antidiabetic, hypolipidemic histopathological analysis of *Panax quinquefolium* aqueous (AGE) extract in alloxan induced diabetic rat by administering intraperitoneal dose (100 mg/kg body weight / day). **Methods**: Optimal cutoff level of each of the four plasma glucose values of oral glucose tolerance test in alloxan diabetic rat was done. Other parameters as liver profile, renal profile and total lipid levels were determined in normal and alloxan induced diabetic rats after intraperitoneal administration of the extract for 30 days. Histopathological changes in diabetic rat organs (pancreas, liver and kidney were also observed after extract treatment. **Results**: Daily intraperitoneal administration AGE (100 mg/kg body weight) and glimebride (10 microg/g body wt) showed beneficial effects on blood glucose level (P<0.001) as well as improving kidney, liver functions and hyperlipedemia due to diabetes. The extract treatment also showed enhanced serum insulin level and body weight of diabetic rats as compared to diabetic control group. Furthermore, the extract has a favorable effect on the histopathological changes of the pancreas, liver and kidney in alloxan induced diabetes. **Conclusion**: *Panax quinquefolium* posses antidiabetic property as well as improve body weight, liver profile, renal profile and total lipid levels. AGE has also favorable effect to inhibit the histopathological changes of the pancreas and kidney in alloxan induced diabetes.   © 2015 Trade Science Inc. - INDIA

INTRODUCTION

Diabetes mellitus (DM) is a very commonly occurring metabolic disorder characterized by hyperglycemia and altered metabolism of carbohydrates, lipids and proteins. DM occurs due to absolute or relative deficiency of insulin or insulin resistance[9].

This metabolic disorder affects approximately 4% of the population worldwide and is expected to be increased by 5.4% in 2025. Because DM is associated with oxidative stresss, it alters the cellular microenvironment in many different types of tissues causing myriad untoward effects, collectively referred to as 'diabetic complications'. Two cellular processes affected by diabetes are inflammation and apoptosis[24].

Glimepiride is a medium-to-long acting sulfonylurea anti-diabetic drug. sometimes classified as the first third-generation sulfonylurea and sometimes classified as sec-

ond-generation. Glimepiride acts as an insulin secreta-gogue. It lowers blood sugar by stimulating the release of insulin by pancreatic beta cells and by inducing increased activity of intracellular insulin receptors[28].

Synthetic drugs usually come with considerable side effects, such as hypoglycemia, drug-resistance, dropsy, and weight gain In recent years due to the adverse effects of synthetic hypoglycemic drugs, interests in alternate therapeutic approach have become very popular Nowadays, herbal drugs are gaining popularity in the treatment of diabetes and its complications due to their efficacy, low incidence of side effects and low Cost.

The medicinal plants provide a useful source of oral hypoglycemic compounds for the development of new pharmaceutical leads. Some of the plants which are being used for the treatment of diabetes have received scientific or medicinal scrutiny and even the WHO expert committee on diabetes recommends that this area warrant further attention[27].

(Panax quinque folius L.) American ginseng is slow-growing perennial plants with fleshy roots, belonging to the *Panax* genus in the family Arali-aceae. Ginseng is one of the most widely used herbal medicines and is reported that root extract of ginseng has a wide range of therapeutic and pharmacological applications for antioxidant, antidiabetic and vasorelaxation effects[1].

MATERIALS AND METHODS

Glimepiride

The drug was purchased from (Delta Pharma for Pharmaceuticals, Egypt). Oral dose: Oral administration of (10 microg/g body wt) daily in accordance with[15].

Plant material

Mature whole *Panax quinque folius* plants were collected and authenticated as *Panax quinque folius* by Dr. H. Ezzat, Department of Pharmacognacy, Faculty of pharmacy, Minia university.

Intraperitoneal dose: 100 mg/kg of body weight per day in accordance with[10].

Extract preparation of *Panax quinque folius* (American ginseng)

The plant roots were soaked in cold water for 2 h,

and then cut into small pieces less than 2mm in diameter. These pieces were mixed with hot water about 95°C for 1 h, and then filtered. The filtrates were evaporated and lyophilized. The dried powders were suspended in bath solution. The suspension was centrifuged for 10 min at 3000rpm and the supernatant was used for the experiment.

Chemicals

Alloxan was purchased from Loba chemie Pvt. Ltd. Mumbai, India. Total cholesterol (TC), serum high-density lipoprotein (HDL), serum Creatinine (SC), serum urea (SU), alanine transaminase (ALT), serum aspartate transaminase (AST) and triglyceride (TG) standard kits were obtained from Erba diagnostics Mannheim Gambh, Germany. Blood glucose level was measured using Elegance glucose meter (CT-X10) of Convergent Technologies, Germany. All reagents used in study were analytical grade.

Animals

40White male albino rats weighting about 190 ± 10 g were used as experimental animals in the present investigation. The animals were housed in standard polypropylene cages and maintained under controlled room temperature (22 ± 2 æ%C) and humidity ($55\pm5\%$) with 12 h light and 12 h dark cycle and were fed a standard diet of known composition, and water ad libitum. The chow was purchased from El-Gomhoria Company, Cairo, Egypt. They were housed for two weeks for accommodation. Our work was carried out in accordance with the guidelines of El Minia University for animal use. These animals were used for induction of *Diabetes mellitus.*

Induction of diabetes

Rats were made diabetic by a single intraperitoneal njection of alloxan monohydrate (Loba Chemie, Bombay; 150 mg/kg i.p.) in sterile saline. Twelve days after Alloxan injection, rats with blood glucose level of >200 mg/dL were separated and used for the study. Blood glucose levels were measured using blood glucose test strips with elegance glucometer (Frankenberg, Germany) at weekly intervals till the end of study (i.e. 3 weeks). Blood glucose estimation was done on 0, 1, 2 and 3 hours after administration of extract orally at the 30 th day of starting experiment.

TABLE 1 : Effect of AGE on optimal cutoff level of each of the four plasma glucose values of oral glucose tolerance test in alloxan induced diabetic rats (A-D)(n=10)

Group	Optimal cutoff level			
	Fasting glucose level	1. h glucose level	2. h glucose level	3. h glucose level
Normal (N)	69.12±2.3	112.3±9.8	89.3±7.2	74.9±6.8
Diabetic Control (DC)	142±12.3 ***[a]	465±41.8 ***[a]	435±30.25 ***[a]	412±38.5 ***[a]
Diabetic +Glimepride (G)	176±16.2 [a]	484±40.28 [a]	401±29.8 [b]	314±27.1 [c]
Diabetic+ AGE (AGE)	109±9.3 [c]	405±39.65 [c]	381±32.0[c]	374±25.36 [b]

* Values significantly different compared to normal P***‹0.001; * Values are expressed as means ± SE. Means not sharing common letter are significantly different (p‹0.05) based on one –way ANOVA with Tukey's post –hoc test.

Experimental design

Overnight fasted rats were divided into four groups and for each group 10 animals and treated orally once a day for 30 days as follows:

- Group I**:**Normal healthy control: given only vehicle (Tween)
- Group II: Diabetic control: given only vehicle (Tween 80, 5% 80, 1% v/v) v/v)
- Group III**:** Diabetic rats given Glimepiride (10 microg/g body wt.)
- Group IV**:** Diabetic rats given intraperitoneal dose: 100 mg/kg of body weight per day in accordance with[10].

Biochemical parameters

Optimal cutoff level of each of the four plasma glucose values of oral glucose tolerance test in alloxan diabetic was measured with elegance glucometer (Frankenberg, Germany) at hour intervals i.e. 0, 1, 2 and 3 hours after daily administration of extract intraperitoneally. After blood glucose estimation on day 30, whole blood was collected by cardiac puncture under mild ether anesthesia from rats. Serum cholesterol, triglycerides, creatinine, urea, and HDL cholesterol levels were also evaluated in normal and alloxan induces diabetic rats. Serum alanine transaminase (ALT) and serum aspartate transaminase (AST) were measured by autoanalyser (Erba Chem 7, Mannheim, Germany) using Erba diagnostic kits. Serum insulin levels were determined using insulin ELISA kit.

Statistical analysis

Statistical analysis was carried out using Graph Pad Instat software (version 3, ISS-Rome, Italy). Groups of data were compared with ANOVA, followed by Tukey-Kramer (TK) multiple comparisons post-test.

TABLE 2 : Effect of AGE on serum insulin level in alloxan induced diabetic rats (A-D)(n=10)

Group	Serum insulin levels (ng/ml)
	After 30 days of treatment
Normal (N)	1.18±0.07
Diabetic control (DC)	0.48±0.07***[a]
Diabetic + Glimepride (+G)	0.62±0.04 [a]
Diabetic+ AGE (AGE)	0.78±0.06 [b]

* Values significantly different compared to normal P***‹0.001; * Values are expressed as means ± SE. Means not sharing common letter are significantly different (p‹0.05) based on one –way ANOVA with Tukey's post –hoc test.

Values of $P < 0.05$ were regarded as significant. Data were expressed as mean ± standard error (SEM).

RESULTS

Antidiabetic activity

Single dose alloxan monohydrate (150 mg/kg) significantly (P<0.01) increases the blood glucose as shown in TABLE 1. After the daily intraperitoneal administration with AGE, for 30 days, significant decreased (P<0.01) in the blood glucose levels was observed in the diabetic rats (TABLE 1. The reduced insulin level in diabetic rats was also significantly improved by treatment of AGE. At the end of experiment (30[th]day) blood glucose level was (374±25.36) mg/dL of the groups treated with AGE (TABLE 2).

Effect on body weight of rats

In diabetic rats, continuous reduction in body weight was observed as shown in TABLE 2. AGE treatment significantly (P<0.05) improved the body weight of diabetic rats.

Effect on lipid profile

In diabetic rats, there was a significant increase of

Regular Paper

TABLE 3 : Effect of AGE on body weight, triglycerides (T.G) and total cholesterol (T.C) in alloxan induced diabetic rats.

Group	Body weight gain	T.G	T.C
	gm	mg/dl	mg/dl
Normal (N)	37±3.21	74.2±6.8	87.2±8.4
Diabetic Control (DC)	-27±3.4[***a]	178.2±10.4[***a]	163.2±7.9[***a]
Diabetic + Glimepride (G)	-21±2.6 [a]	120.3±8.6 [b]	102.3±7.4 [b]
Diabetic+ AGE (AGE)	-23±4.2 [a]	123.3±7.4 [b]	108.2±8.2 [b]

* Values significantly different compared to normal P***‹0.001; * Values are expressed as means ± SE. Means not sharing common letter are significantly different (p‹0.05) based on one –way ANOVA with Tukey's post –hoc test.

TABLE 4 : Effect of AGE on HDL cholesterol and LDL cholesterol in alloxan induced diabetic rats.

Group	HDL.C	LDL.C
	mg/dl	mg/dl
Normal (N)	63.2±5.7	13.5±7.3
Diabetic Control (DC)	39.3±4.5[***a]	53.2±4.8[***a]
Diabetic + Glimepride (G)	45.2±3.4 [b]	35.1±3.3 [b]
Diabetic+ AGE (AGE)	46.7±3.8 [c]	31.2±3.4 [b]

* Values significantly different compared to normal P***‹0.001; * Values are expressed as means ± SE. Means not sharing common letter are significantly different (p‹0.05) based on one –way ANOVA with Tukey's post –hoc test.

serum total cholesterol, triglycerides (TABLE 3) and significant decrease in HDL cholesterol in compared to that of normal control. The standard drugs as well as AGE used in the experimental study significantly decreased (P<0.05) the levels of cholesterol and triglycerides whereas HDL cholesterol significantly increased (TABLE 4).

Effect on liver functions

The effect of AGE on liver functions is represented in the TABLE 5. ALT, AST were significantly elevated in alloxan induced diabetes. The rats treated with AGE showed significant (P<0.01) reduction in the elevated levels of liver enzymes (transaminase) in a dose dependent manner as shown in TABLE 5.

Effect on kidney functions

Kidney function markers like urea and creatinine were elevated in the alloxan induced diabetic rats when compared with the normal rats. AGE reduced both the levels in dose dependent manner (TABLE 6).

Histology of pancreas

Histology of pancreas (Figure 1) showed normal acini, and normal cellular in the islets of langerhans in the pancreas of normal control (1.a). In diabetic animals treated extensive damage to islets of langerhans and reduced dimensions of islets were observed in diabetic rats (1.b). On other hand, AGE treatment showing vacuolization of B cells of islets of langerhans (1.d).

Histology of liver

Photomicrographs of liver (Figure 2) showed normal hepatic cells with well preserved cytoplasm, nucleus, nucleolus and central vein (2.a). In case of group II diabetic rats, the normal lobular structure was preserved. The central vein was prominent and prominently congested. Focal areas of hemorrhage were also seen. Fatty change was evident.

The portal tracts appeared normal (2.b). In group IV [diabetic rats + AGE mg/kg], showing kupffer cells activation (2.d).

Histology of kidney

TABLE 5 : Effect of AGE on liver parameters in normal and diabetic rats

Group	sALT	sAST
	U/l	U/l
Normal (N)	19.51±0.84	23.17±0.75
Diabetic Control (DC)	62.39±0.87***[a]	84.29±0.94***[a]
Diabetic + Glimepride (G)	59.18±0.91[a]	70.06±0.89[a]
Diabetic+ AGE (AGE)	43.28±0.92[b]	45.17±0.74[b]

* Values significantly different compared to normal P***‹0.001; * Values are expressed as means ± SE. Means not sharing common letter are significantly different (p‹0.05) based on one –way ANOVA with Tukey's post –hoc test.

TABLE 6 : Effect of AGE on kidney parameters in normal and diabetic rats (n=10).

Group	Urea	Creatinine
	(mg/dl)	(mg/dl)
Normal (N)	25.41±2.14	0.65±0.07
Diabetic Control (DC)	66.14±2.14***[a]	1.31±0.01***[a]
Diabetic + Glimepride (G)	29.26±2.65 [c]	0.98±0.03 [b]
Diabetic+ AGE (AGE)	41.21±2.33 [b]	1.09±0.04 [b]

* Values significantly different compared to normal P***‹0.001; * Values are expressed as means ± SE. Means not sharing common letter are significantly different (p‹0.05) based on one –way ANOVA with Tukey's post –hoc test.

1.a

1.b

1.c

1.d
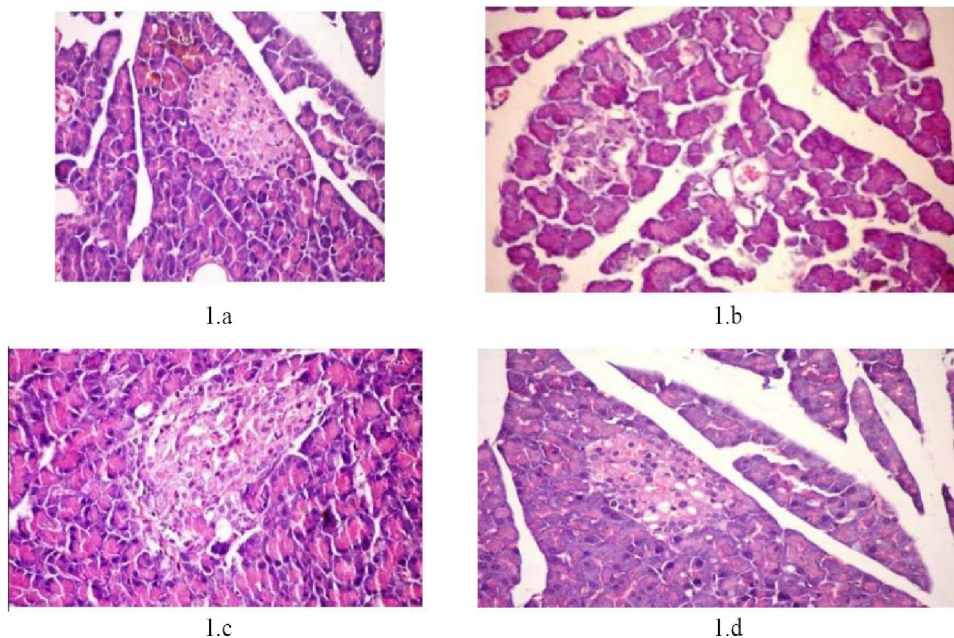
Figure 1 : Effect of AGE on pancreas of alloxan induced diabetic rats

2.a

2.b

2.c

2.d
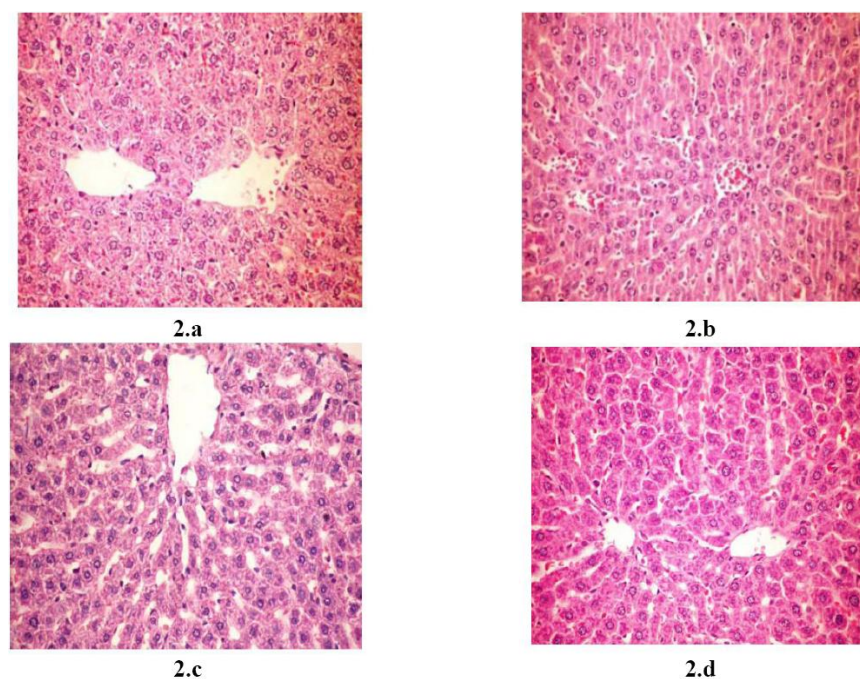
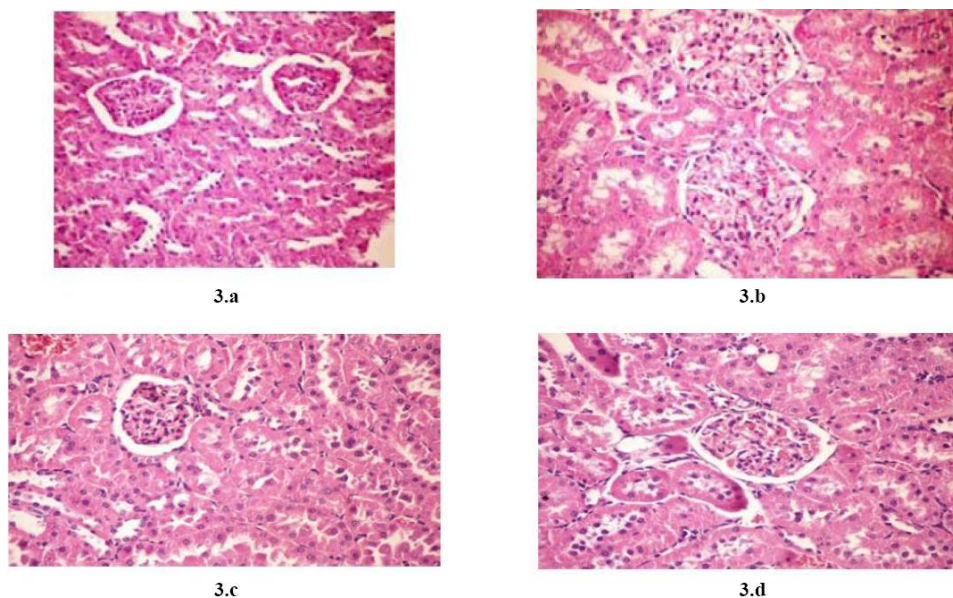**Figure 2 : Effect of AGE on liver of alloxan induced diabetic rats**

Figure 3 : Effect of AGE on kidney of alloxan induced diabetic rats

Histology of kidney (Figure 3) in normal animals showed normal structure (3.a). In diabetic rats, mild thickening of the basement membrane of the arterioles of glomeruli along with mild change of density of mesangial mesangium were observed. No other significant changes were seen (3.b). After AGE 100 mg/kg treatment, these changes were improved towards normal condition (3.d).

DISCUSSION

Glucose is the key physiological regulator of insulin secretion; indeed, short-term exposure of β-cells to increasing glucose concentrations induces proliferation in a concentration-dependent manner. In addition to its effect on β-cell turnover, hyperglycemia also impairs β-cell secretary function. This glucotoxic effect is evident before apoptosis leads to a significant decrease in β-cell mass[6].

The present investigation indicated that a single dose of alloxan (150 mg/kg) intraperitoneally to adult male albino rats (210-220g) was suitable to induce histological changes of the islets of Langerhans characterized appearance, hypoinsulinemia and hyperglycemic state. The present dose as well as the observed histopathological and biochemical manifestations agree with the literature of[11].

A gradual loss of β-cells due to apoptosis significantly hinders insulin production and inhibits cell viabil-

ity. During apoptosis, cells shrink; chromatin condenses; DNA is cleaved into pieces at inter nucleosomal regions. A proactive way to increase β-cell viability is to decrease apoptosis level in order to retain the cell population and increase insulin production.

Oral administration of glimepiride causes significant decrease in levels of blood glucose in accordance with who revealed that glimepiride exerts its insulin-releasing effect mainly by inhibiting ATP-sensitive potassium channels. In the pancreatic β-cell this action induces depolarization of the cell membrane, allowing an influx of calcium in the cell. This in turn induces insulin release into the blood.

Pfützner et al[20] revealed that the increase in the number of β-cells in the islets of Langerhan's in glimepiride-treated diabetic rats in comparison to alloxan induced diabetic rats can be attributed to the fact that glimepiride affect the activation of the redox sensitive transcription factor NF (Kappa) β in alloxan induced diabetic rats. Although the mechanism of β-cell neoformation is not clear but there is strong evidence that islet stem cells may exist in the pancreatic duct and that these ductal epithelial cells may be switched into a proliferative /regenerative phase leading to nesideoblastosis (neogenesis of islets)

Antidiabetic effect of glimepiride was further examined by histological observations made on the pancreatic tissue of glimepride treated rats that show vacuolization and necrosis of some β cells of islets of Langer-

hans.

Intraperitoneal administration of AGE causes significant decrease in levels of blood glucose accordance with[26]. Also, intraperitoneal administration of AGE causes significant increase in level of serum insulin.

Theantidiabetic effect of AGE may due to antioxidant activity of ginsenosides was involved in the anti-apoptosis activity as production of free radicals, such as NO and ROS was reduced by ginsenosides[14].

Antidiabetic effect of AGE was further evidenced by histological observations made on the pancreatic tissue of AGE treated rats that revealed few vacuolizations and slightly divided β-cells in the islets of Langerhans.

The characteristic features of diabetic dyslipidemia are a high plasma triglyceride concentration, low HDL-cholesterol concentration and increased concentration of small dense LDL-cholesterol particles. The lipid changes associated with *diabetes mellitus* are attributed to increased flux of free fatty acids into the liver secondary to insulin deficiency/ resistance. This results in excess fatty acid accumulation in the liver, which is converted to triglycerides. The impaired ability of insulin to inhibit free fatty-acid release leads to elevated hepatic VLDL-cholesterol production. The increased VLDL-cholesterol and triglyceride levels decrease the level of HDL-cholesterol and increase the concentration of small dense LDL-cholesterol particles by activation of lipoprotein lipase and lecithin acyl-cholesterol transferase[19].

In our study, elevated levels of serum TC, TG, LDL and VLDL-cholesterol and decreased HDL -cholesterol concentration in alloxan-induced diabetic rats are in accordance with[29].

On other hand, Induction of diabetes by alloxan resulted in loss of body weight in the diabetic control rats in accordance with that may due to catabolic effect on protein metabolism by retarding protein synthesis and stimulating protein degradation.

Oral administration of glimepiride causes significant decrease in the serum levels of triglycerides, total cholesterol and LDL-cholesterol in contrast to significant elevation in HDI.-cholesterol and body weight in accordance with[8].

Motoyama et al[18] revealed that glimepiride improved HDL-c level via improvement of plasma adiponectin level as adiponectin could increase HDL-c

levels directly via increased lipoprotein lipase and decreased hepatic lipase activity. On other hand, the antilipidemic action of glimepiride may reside in their ability to stimulate insulin secretion and action.

Intraperitoneal administration of AGE causes significant decrease in the serum levels of triglycerides, total cholesterol and LDL-cholesterol in contrast to significant elevation in HDI.-cholesterol and body weight in accordance with[2].

Yeo et al[30] revealed that AGE improves the lipid profile by regulation of the expression of genes associated with abnormal physiology associated DM, insulin, and adiponectin, which carry out critical functions in energy and lipid metabolism.

DM causes a disturbance in the uptake of glucose, as well as glucose metabolism. The liver plays an important role in the maintenance of blood glucose levels by regulating its metabolism.

The present investigation indicated that, a single dose of alloxan (150 mg/kg) intraperitoneally to adult male albino rats (210-220g) was suitable to induce histological changes in the liver of alloxan induced diabetic rats with characterized appearance, enlarged and swollen hepatocytes.

Alloxan causes significant increase in activity of sAST and sALT, in accordance with that is directly related to changes in metabolism in which the enzymes are involved. The increased activities of transaminases, which are active in the absence of insulin due to the availability of amino acids in the blood of DM and are also responsible for the increased gluconeogenesis and ketogenesis[3].

DM induces the growth of HSCs via MAP kinase pathways, which are activated by ROS produced by the NADPH oxidase system under the regulation of protein kinase C. On other hand, hepatic oxidative stress induces proinflammatory cytokines, such as TNF-α, transforming growth factor-β (TGF-β), interleukin-1β, and interleukin-6, which are critical for HSC activation and perpetuation[25].

Intraperitoneal administration of AGE causes significant decrease in level of AST and ALT in accordance with[12] who attributed the antioxidant effect of ginsenosides in AGE. Hepatic protective effect AGE of was further evidenced by histological observations made on the hepatic tissue of AGE treated rats that revealed

Regular Paper

no histological changes.

Renal dysfunction is a slowly progressive process that is postulated to be accelerated by intervening diseases, such as diabetes, due in part to the addition of excessive stress and inflammation[16].

Alloxan led to a significant increase in serum urea and serum creatinine A similar effect was recorded by[4]. [23]revealed that enhanced protein catabolism and accelerated amino acid deamination for gluconeogenesis is probably an acceptable postulate to interpret the elevated levels of urea and creatinine. Furthermore, Alloxan increased the productions of reactive oxygen species, enhanced lipid peroxidation and protein carbonylation in association with decreased intracellular antioxidant defense in the kidney tissue

Cao et al[5] suggests that development of diabetic renal dysfunction may due to activation of endoplasmic reticulum stress that can mediate progressive endothelial damage through growth and migration of vascular smooth muscle and inflammatory cells, alteration of extracellular matrix, apoptosis of endothelial cells, overexpression of inflammatory cytokines.

Oral administration of glimepiride causes significant decrease in level of seum urea and creatinine in accordance with[22].

In the present study the improvement in blood urea, serum creatinine and subsequent amelioration of histomorphological changes in kidneys of glimepiride treated rats can be attributed to the recovery of renal function which is explained by the regenerative capability of the renal tubules as good metabolic control is beneficial in slowing the progression of renal dysfunction in diabetes[17].

Glimepiride could ameliorate the glomerular and tubular lesions that characterize diabetic renal dysfunction and subsequently recover renal morphology and function. Reno- protective effect glimepiride of was further evidenced by histological observations made on the renal tissue of glimepiride treated rats that revealed normal structure of renal parenchyma.

Intraperitoneal administration of AGE causes significant decrease in level of seum urea and creatinine in accordance with[10].

Fu and Ji[7] indicates that ginseng components could prevent diabetic renal damage by attenuating the oxi-

dative stress, inhibiting advanced glycation endproduct formation and of MAPK-Akt signaling cascade. Renoprotective effect of was further evidenced by histological observations made on the renal tissue of AGE treated rats that revealed no histological changes.

CONCLUSION

Although additional studies are needed, it could be suggested that AGE have Antidiabetic, Antihyperlipidemic mechanisms against Pancreas, liver and kidney injury induced by alloxan. The signaling mechanisms associated with protection against the liver damage and oxidative stress status induced by alloxan via intake AGE still need merit further investigations.

CONFLICT OF INTEREST STATEMENT

We declare that we have no conflict of interest.

ACKNOWLEDGEMENTS

REFERENCES

[1] K.A.Amin, E.M.Awad, M.A.Nagy; Effect of *Panax quinquefolium* on streptozotocin-induced diabetic rats: role of C-peptide, nitric oxide and oxidative stress, Int.J.Clin.Exp.Med., **4(2)**,136-47 **(2011)**.

[2] W.J.Banz, M.J.Iqbal, M.Bollaert, N.Chickris, B.James, D.A.Higginbotham, R.Peterson, L.Murphy; Ginseng modifies the diabetic phenotype and genes associated with diabetes in the male ZDF rat, Phytomedicine, **14(10)**, 681-9 **(2007)**.

[3] S.A.S.Batran, S.E.El-Gengaihi, O.A.Shabrawy; Some toxicological studies of Momordica charantia L. on albino rats in normal and alloxan diabetic rats, J.Ethnopharmacol., **108**, 236–242 **(2006)**.

[4] V.B.Brito, J.B.Da Rocha, G.O.Puntel, S.C.da Luz, N.B.Barbosa, N.R.de Carvalho, V.Folmer; Inhibition of δ-aminolevulinate dehydratase is not closely

related to the development of hyperglycemia in alloxan-induced diabetic mice, Exp Toxicol Pathol., **63(5)**, 443-51 **(2011)**.

[5] Y.P.Cao, Y.M.Hao, Q.J.Liu, J.Wang, H.Li, H.J.Duan; Therelationship between endoplasmic reticulum stress and its particular apoptosisway caspase-12 and apoptosis in renal cortex of diabetic rats, Zhongguo Ying Yong Sheng Li Xue Za Zhi, **27(2)**, 236-40 **(2011)**.

[6] M.Y.Donath, P.A.Halban; Decreased beta-cell mass in diabetes: significance, mechanisms and therapeutic implications, Diabetologia, **47**, 581 –589 **(2004)**.

[7] Y.Fu, L.L.Ji; Chronic ginseng consumption attenuates age-associated oxidative stress in rats, J Nutr, **133(11)**, 3603-9 **(2003)**.

[8] N.R.Hadi, F.Al-Amran, M.A.Hussein, F.A.Rezeg; Evaluation of the effects of glimepiride (Amaryl) and repaglinide (novoNorm) on atherosclerosis progression in high cholesterol-fed male rabbits, J Cardiovasc Dis Res, **3(1)**, 5-11 **(2012)**.

[9] J.Y.Joo, D.L.Huber; An integrative review of case management for diabetes, Prof Case Manag, **17(2)**, 72-85 **(2012)**.

[10] H.Y.Kim, K.S.Kang, N.Yamabe, R.Nagai, T.Yokozawa; Protective effect of heat-processed American ginseng against diabetic renal damage in rats, J Agric Food Chem. 17, **55(21)**, 8491-7 **(2007)**.

[11] V.P.Lebedev, S.V.Bilichenko, N.E.Ordyan, S.G.Pivina, S.P.Nechiporenko, A.A.Puzyrev, E.A.Mikheeva, K.K.Kubacheva; Transcranial electrostimulation activates reparative regeneration and the insulin-producing function of pancreatic B-cells in alloxan diabetes in rats, Neurosci Behav Physiol., **37(4)**, 341-7 **(2007)**.

[12] L.S.Lee, S.D.Wise, C.Chan, T.L.Parsons, C.Flexner, P.S.Lietman; Possible differential induction of phase 2 enzyme and antioxidant pathways by american ginseng, *Panax quinquefolius*, J Clin Pharmacol, **48(5)**, 599-609 **(2008)**.

[13] C.Li, Y.Gao, Y.Xing, H.Zhu, J.Shen, J.Tian; Fucoidan, a sulfated polysaccharide from brown algae, against myocardial ischemia–reperfusion injury in rats via regulating the inflammation response, Food and Chemical Toxicology, **(2011)**.

[14] J.Z.Luo, L.Luo; Ginseng on hyperglycemia: effects and mechanisms. Evid Based Complement Alternat Med, **6(4)**, 423-7 **(2009)**.

[15] L.Ladrière, F.Malaisse-Lagae, J.Fuhlendorff, W.J.Malaisse; Repaglinide, glibenclamide and glimepiride administration to normal and hereditarily diabetic rats, Eur J Pharmacol, **335(2-**3), 227-34 **(1997)**.

[16] E.Mandache, M.Penescu; Nanostructural features of diabetic podocytopathy, Rom.J.Morphol. Embryol, **53(1)**, 23-7 **(2012)**.

[17] A.L.McCall; Clinical review of glimepiride, Expert Opin Pharmacother., **2(4)**, 699-713 **(2001)**.

[18] M.Emoto, T.Konishi, Y.Ikuno, E.Lee, M.Teramura, K.Motoyama; Glimepiride increases high-density lipoprotein cholesterol via increasing adiponectin levels in type 2 diabetes mellitus, Metabolism, **58(2)**, 143-8 **(2009)**.

[19] M.A.Nagy, M.A.Bastawy; Renoprotective effects of Egyptian herbal formula during experimental diabetes, Access Scientific Reports, **1(9)**, (1-9) **(2012)**.

[20] A.Pfützner, T.Schöndorf, D.Seidel, K.Winkler, S.Matthaei, A.Hamann, T.Forst; Impact of rosiglitazone on beta-cell function, insulin resistance, and adiponectin concentrations: results from a double-blind oral combination study with glimepiride, Metabolism, **55(1)**, 20-5 **(2006)**.

[21] S.I.Rabbani, K.Devi, S.Khanam; Role of Pioglitazone with Metformin or Glimepiride on Oxidative Stress-induced Nuclear Damage and Reproductive Toxicity in Diabetic Rats, Malays.J.Med.Sci., **17(1)**, 3-11 **(2010)**.

[22] B.Rosenkranz, V.Profozic, Z.Metelko, V.Mrzljak, C.Lange, V.Malerczyk; Pharmacokinetics and safety of glimepiride at clinically effective doses in diabetic patients with renal impairment, Diabetologia, **39(12)**, 1617-24 **(1996)**.

[23] B.Santamaría, A.B.Sanz, A.C.Ucero, M.D.Sánchez-Niño, A.Benito-Martín, A.Reyero, M.Ruiz Ortega, R.Selgas, A.Ortiz; Apoptosis: from advances in PD to therapeutic targets in DM. Nefrologia 28, **6**, 23-6 **(2008)**.

[24] I.A.Van den Oever, H.G.Raterman, M.T.Nurmohamed, S.Simsek; Endothelial dysfunction, inflammation, and apoptosis in diabetes mellitus, Mediators Inflamm, 792393 **(2010)**.

[25] R.Sugimoto, M.Enjoji, M.Kohjima, S.Turuta, M.Fukushima, M.Iwao, T.Sonta, K.Kotoh, T.Inoguchi, M.Nakamura; High glucose stimulates hepatic stellate cells to proliferate and to produce collagen through free radical production and activation of mitogen-activated protein kinase, Liver Int, **25**, 1018–1025 **(2005)**.

[26] V.Vuksan, J.L.Sievenpiper, J.Wong, Z.Xu, U.Beljan-Zdravkovic, J.T.Arnason, V.Assinewe, M.P.Stavro, A.L.Jenkins, L.A.Leiter, T.Francis; American ginseng (*Panax quinquefolius* L.) attenuates post-

Regular Paper

prandial glycemia in a time-dependent but not dose-dependent manner in healthy individuals, Am J Clin Nutr, **73(4)**, 753-8 **(2001)**.

[27] WHO; WHO expert committee on Diabetes mellitus, Technical reports series World Health Organization, Geneva **(1980)**.

[28] P.Wlodarczyk, M.Paluch, Z.Wojnarowska, L.Hawelek, K.Kaminski, J.Pilch; Theoretical and experimental studies on the internal mobility of two sulfonylurea agents: glibenclamide and glimepiride, J.Phys.Condens.Matter., **23(42)**, 425901 **(2011)**.

[29] U.C.Yadav, K.Moorthy, N.Z.Baquer; Combined treatment of sodium orthovanadate and Momordica charantia fruit extract prevents alterations in lipid profile and lipogenic enzymes in alloxan diabetic rats, Mol Cell Biochem., **268(1-2)**, 111-20 **(2005)**.

[30] J.Yeo, Y.M.Kang, S.I.Cho, M.H.Jung; Effects of a multi-herbal extract on type 2 diabetes, Chin Med., **4**, 6:10 **(2011)**.