Australian Journal of Basic and Applied Sciences, 5(7): 387-396, 2011
ISSN 1991-8178

Effect of *Cichorium endivia* Leaves on Some Biochemical Parameters in Streptozotocin-Induced Diabetic Rats

[1]Zeinab H. Kamel, [2]Iman Daw and [1]Mohamed Marzouk

[1]Department of Zoology, Faculty of Science, Cairo University, Egypt.
[2]Department of Zoology, Faculty of Science, Aljabal Algharbi University, Alzintan, Lybia.

Abstract: The present study was carried out to examine the effect of aqueous suspension of *Cichorium endivia* leaves powder or glibenclamide drug on some biochemical parameters in streptozotocin-induced diabetic rats. Adult male albino rats were randomly divided into 5 groups, (1) control group which was administered distilled water, (2) *Cichorium endivia* group in which rats received aqueous suspension of *C. endivia* leaves powder, 500 mg/kg b. wt., (3) diabetic group in which diabetic rats were administered distilled water, (4) diabetic–*Cichorium endivia* group in which diabetic rats were treated with aqueous suspension of *C. endivia* leaves powder, 500 mg/kg b. wt., and (5) diabetic–glibenclamide group in which diabetic rats were treated with glibenclamide drug, 600 mg/kg b. wt. All groups were induced by intragastric administration for six weeks. The *C. endivia* leaves powder or the glibenclamide drug produced significant hepatoprotective effects by decreasing the activities of serum aminotransferases (AST and ALT), alkaline phosphatase (ALP), lactate dehydrogenase (LDH) and liver malondialdehyde (MDA) level as well as liver superoxide dismutase (SOD) and catalase (CAT) activities, and increasing the liver glutathione peroxidase (GPx) and glutathione-S-transferase (GST) activities and reduced glutathione (GSH) level. In conclusion, the results obtained clearly indicate the role of oxidative stress in induction of diabetes and suggest protective and/or ameliorative effects of *C. endivia* leaves powder aqueous suspension, similar to the diabetic drug (glibenclamide) effects in this animal model.

Key words: *Cichorium endivia*- Liver- Streptozotocin- Biochemical parameters- Diabetes- Antioxidant enzymes

INTRODUCTION

Diabetes is a major degenerative disease in the world today (Ogbonnia *et al.,* 2008). It is a multifactorial disease which is characterized by hyperglycemia (Ugochukwu *et al.,* 2003), lipoprotein abnormalities (Scoppola *et al.,* 2001), raised basal metabolic rate (Owu *et al.,* 2006), defect in reactive oxygen species scavenging enzymes (Kesavulu *et al.,* 2000) and high oxidative stress induced damage to pancreatic β-cells (Nayeemunnisa, 2009). Streptozotocin (STZ) was recognized as a toxic agent for β-cells of the islets of Langerhans (Agarwal, 1980), and has since then been widely used for the induction of diabetes mellitus in experimental animals (Kim *et al.,* 2003). Damasceno *et al.* (2002) reported that STZ produced oxidative stress and depletion of antioxidant systems in both blood and tissue particularly, liver. The concentration of the reactive oxygen species are modulated by antioxidant enzymes-glutathione peroxidase (GPx), glutathione-S-transferase (GST), superoxide dismutase (SOD), catalase (CAT) and non-enzymatic scavengers like reduced glutathione (GSH) (Saxena *et al.,* 1993).

Management of diabetes mellitus without any side effects is still a challenge to the medical system. There is an increasing demand by patients to use natural products with antidiabetic activity, because insulin and oral hypoglycemic drugs have undesirable side effects (Kameswara and Appa, 2001). Medicinal plants are a good source of natural antioxidants believed to exert their effects by reducing the formation of the final active metabolite of the drug-induced systems or by scavenging the reactive molecular species to prevent their reaching a target site (Kaleem *et al.,* 2005). It has been documented that several medicinal plants show them fromhypoglycemic effects associated with a significant alteration in the activity of liver hexokinase (Bopanna *et al.,* 1997) and glucokinase (Kumari *et al.,* 1995). In addition, Mansour *et al.* (2002) and Eliza *et al.* (2009) demonstrated that the administration of several herb extracts could restore the changes in the activities of serum enzymes, like transaminases (AST & ALT) as well as alkaline phosphatase (ALP) and lactate dehydrogenase (LDH) of diabetic rats.

Corresponding Author: Zeinab H. Kamel, Zoology Dep., Faculty of Science, Cairo Univ.
E-mail zeinab_azza_4@yahoo.com

Cichorium endivia, a member of the sunflower family Asteraceae, is a typical Mediterranean plant indigenous to Europe, Western Asia and North America (Fernald, 1950). It is very valuable nutritionally, with a high content of dietary fibres, potassium and vitamin C (Kopeck, 1998). The antibacterial, antimalarial, cytotoxic, antidiabetic, no-mutagenic and other activities of chicory were evaluated previously **(**Petrovic *et al.,* 2004). Earlier investigations have reported that the ethanolic extract of *Cichorium intybus* has antidiabetic and hypolipidemic activities in STZ-induced diabetic rats (Pushparaj *et al.,* 2007). Similarly, Upur *et al.* (2009) mentioned that *Cichorium glandulosum* extract reduced serum AST, ALT and ALP activities in CCl_4 and galactosamine administration, which induced acute hepatotoxicity in mice, these results suggested that *C. glandulosum* is a potent hepatoprotective agent that could protect liver against the acute injury and this ability might be attributed to its antioxidant potential. Therefore, the present study aimed to examine the effect of *C. endivia* leaves powder (aqueous suspension) or the glibenclamide drug on some biochemical parameters of STZ-induced diabetic rats.

MATERIALS AND METHODS

Animals:

Adult male albino rats (*Rattus norvegicus*) weighing 105-150 g were used as experimental animals in this study. These animals were grouped and housed in polyacrylic cages (five animals per cage). The rats were provided with standard diet and water *ad libitum*. They were acclimatized under laboratory conditions for one week before experimentation.

Chemicals and Drug:

Streptozotocin (STZ) was purchased from Sigma –Aldrich (St Louis, MO, USA). The glibenclamide drug (commercial name Daonil) was purchased from Aventis Pharma S.A.E., Germany. This drug was widely used in Egypt as diabetic treatment and control.

Plant Material:

Cichorium endivia is aerb belonging to the Asteraceae family. The plant leaves were collected from fields in Giza Province, Egypt. The plant leaves were identified and authenticated by the Herbarium of Botany Department, Faculty of Science, Cairo University. The leaves were dried in an incubator and grounded to a fine powder with a grinder. The *C. endivia* leaves powder was stored in dry place until used.

Experimental Design:

Animals were randomly divided into five groups (7rats/group), the 1st group served as normal control group (N.C.); rats of this group were administered daily 1 ml distilled water orally by gastric gavage. The 2nd group: normal *Cichorium endivia* group (N.Ch.); rats were orally administered with aqueous suspension of *C. endivia* leaves powder (500 mg/kg b. wt.) according to Sadeghi *et al.* (2008). The 3rd group: diabetic control group (D.C.); rats were rendered diabetic by a single intraperitoneal (I.P.) injection of STZ (50 mg/kg b. wt.) according to Kanter *et al.* (2003), in freshly prepared sodium citrate buffer (0.1 M, pH 4.5) after an overnight fast. After 48 hours of STZ injection, blood glucose level of each rat was determined. Rats with blood glucose range from 200-300 mg/dl were considered diabetic and included in the study. Rats of this group were administered daily 1 ml distilled water orally after 48 hours of STZ injection. The 4th group: diabetic–*Cichorium endivia* group (D.Ch.); diabetic rats were administered daily aqueous suspension of *C. endivia* leaves powder (500 mg/kg b. wt.) orally. The 5th group: diabetic–glibenclamide group (D.G.); diabetic animals were administered orally glibenclamide drug (600 mg/kg b. wt.) according to Chandramohan *et al.* (2009). At the end of the experimental period (six weeks), the rats were sacrificed; blood and liver samples were collected for different biochemical analyses.

Serum Preparation:

Blood samples were collected in centrifuge tubes and centrifuged at 3000 rpm for 20 min. Serum was stored at -20°C until used for biochemical assays. The appropriate kits (Biodiagnostic kits) were used for determination of the activities of serum aminotransferases AST & ALT according to Reitman and Frankel (1957), alkaline phosphatase (ALP) (Belfield and Goldberg, 1971) and lactate dehydrogenase (LDH) (Vassault *et al*., 1986).

Liver Tissue Preparation:

Liver was homogenized (10% w/v) in ice-cold 0.1 M phosphate buffer (pH 7.4) and centrifuged at 3000 rpm for 20 min. at 4°C. The resultant supernatant was used for determination of the activities of liver catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase (GPx) and glutathione-S-transferase (GST) according to the methods described by Aebi (1984), Nishikimi *et al.* (1972), Paglia and Valentine (1967) and Habig *et al.* (1974), respectively, as well as the levels of reduced glutathione (GSH) (Beutler *et al.*, 1963) and malondialdehyde (MDA) (Ohkawa *et al.*, 1979) using Biodiagnostic kits.

Statistical Analysis:

Values were expressed as mean ± SE. To evaluate differences between the groups studied, one way analysis of variance (ANOVA) with LSD post hoc test was used to compare the group means and $P<0.05$ was considered statistically significant. SPSS, for Windows (version 17.0) was used for the statistical analysis.

RESULTS AND DISCUSSION

Amelioration of Liver Functions in C. endivia treated Diabetic Rats:

The activities of serum AST, ALT, ALP, and LDH in the N.C., N.Ch., D.C., D.Ch. and D.G. are presented in Table 1 and Fig. 1. The results showed that the administration of aqueous suspension of *C. endivia* leaves powder to normal rats caused significant decrease ($P<0.05$) in serum AST, ALT, ALP, and LDH activities, as compared to the control group. On the other hand, the activities of these enzymes increased significantly ($P<0.05$) in serum of STZ-induced diabetic rats, as compared to the control group. Treatment of diabetic rats orally with aqueous suspension of *C. endivia* leaves powder caused significant decrease ($P<0.05$), of -8.10%, -22.88%, -9.45% and -12.77% in serum AST, ALT, ALP, and LDH activities, respectively, as well as treatment of diabetic rats with glibenclamide drug also caused significant decrease ($P<0.05$), of -8.12%, -19.89%, -15.59% and -13.28% in these serum enzymes, activities respectively, as compared to the diabetic group. It seems that *C. endivia* leaves powder and glibenclamide drug are useful in improvement the alteration recorded in such serum enzymes activities in the diabetic rats (Table 1 & Fig. 1).

Table 1: Effect of treatment with aqueous suspension of *C. endivia* leaves powder or glibenclamide drug for 6 weeks on serum AST, ALT, ALP and LDH activities of diabetic rats.

Parameters	Experimental groups				
	N.C.	N.Ch.	D.C.	D.Ch.	D.G.
AST (U/ml)	64.09 ± 0.79	60.70 ± 1.06 [a]	74.30 ± 1.02 [a]	68.28 ± 0.81 [b]	68.27 ± 1.77 [b]
% of change from N.C.			15.93		
% of change from D.C.				-8.10	-8.12
ALT (U/ml)	52.37 ± 1.40	44.26 ± 1.01 [a]	68.53 ± 2.60 [a]	52.85 ± 0.92 [b]	54.90 ± 1.67 [b]
% of change from N.C.			30.86		
% of change from D.C.				-22.88	-19.89
ALP (IU/L)	86.58 ± 2.56	76.50 ± 3.91 [a]	106.57 ± 1.84 [a]	96.50 ± 1.24 [b]	89.96 ± 1.34 [b]
% of change from N.C.			23.09		
% of change from D.C.				-9.45	-15.59
LDH (U/L)	46.87 ± 2.16	37.73 ± 1.11 [a]	64.23 ± 1.62 [a]	56.03 ± 1.47 [b]	55.70 ± 1.51 [b]
% of change from N.C.			37.04		
% of change from D.C.				-12.77	-13.28

All data are mean of seven rats ± SE

[a] Significant ($P < 0.05$) as compared to the control group (N.C.).

[b] Significant ($P < 0.05$) as compared to the diabetic group (D.C.).

Changes in the Level of Lipid Peroxidation and Antioxidant Enzymes in the Liver of C. endivia treated Diabetic Rats:

Oxidative stress markers, liver CAT, SOD, GPx and GST activities as well as GSH and MDA levels of the different experimental groups are shown in Table 2. Administration of aqueous suspension of *C. endivia* leaves powder to normal rats had no effect on the studied oxidative stress markers in the liver, except SOD activity which decreased significantly ($P<0.05$), as compared to the control group. Intraperitoneal injection of STZ caused significant increase ($P<0.05$) in liver CAT, SOD activities and MDA level of rats, as compared to the normal control group (Fig. 2). On the other hand, STZ administration showed significant decrease ($P<0.05$) in the activities of liver GPx, GST and the level of GSH, as compared to the control group (Table 2 & Fig. 3). It is interesting to notice that the diabetic rats treated with aqueous suspension of *C. endivia* leaves powder showed significant decrease ($P<0.05$), of -9.51%, in liver SOD activity, and significant increase ($P<0.05$), of 38.46% and 23.06%, in GST activity and GSH level, respectively, as compared to the diabetic

group. Concerning the effect of glibenclamide drug on the diabetic rats, it seems that this drug ameliorates the alteration recorded in the liver antioxidant enzymes since, a significant decrease (P<0.05), of -29.17%, -12.33% and -11.96%, in the activities of liver CAT, SOD and the level of MDA, respectively, was as compared to the diabetic group. On the other hand, the activities of liver GPx, GST and the level of GSH of diabetic rats treated with glibenclamide drug showed significant increase (P<0.05), of 12.14%, 53.85% and 28.58%, respectively, as compared to the diabetic group (Table 2 & Fig. 3).

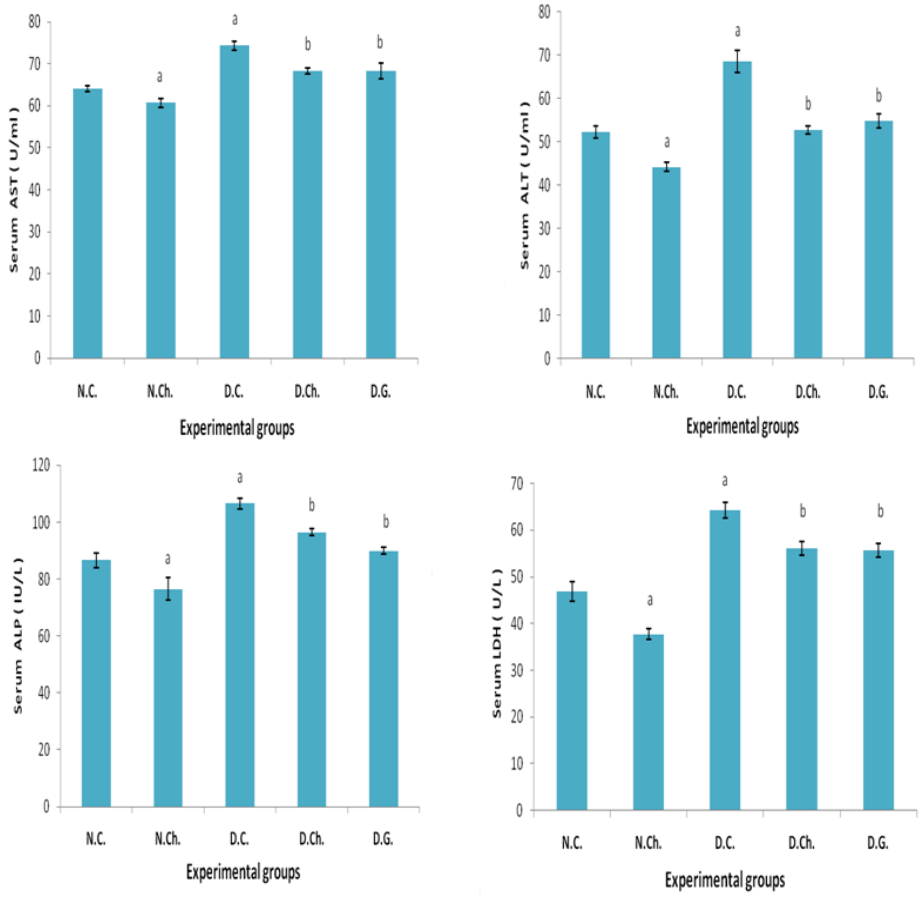
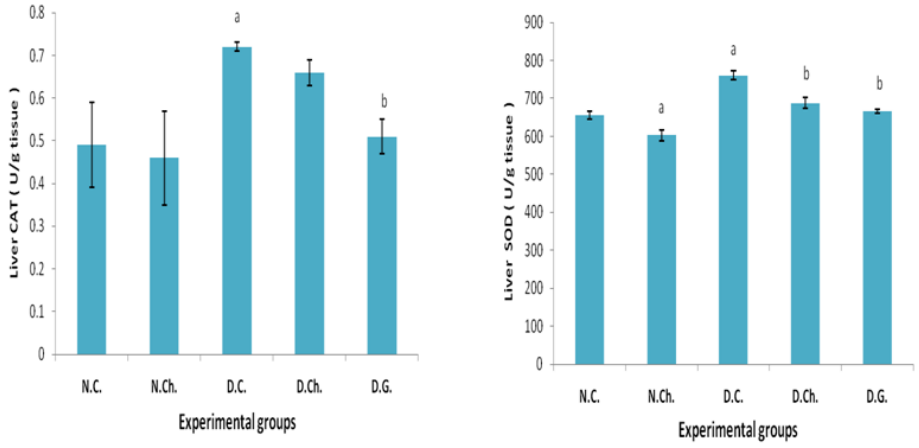


Fig. 1: Serum AST, ALT, ALP and LDH activities (Mean ± SE) of experimental groups.
a Significant (P<0.05) as compared with the normal control (N.C.).
b Significant (P<0.05) as compared with the diabetic control (D.C.).



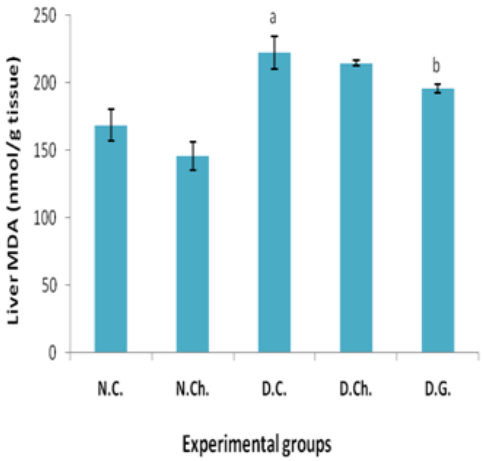


Fig. 2: Liver CAT, SOD activity and MDA concentration (Mean ± SE) of experimental groups.
a Significant (P<0.05) as compared with the normal control (N.C.).
b Significant (P<0.05) as compared with the diabetic control (D.C.).

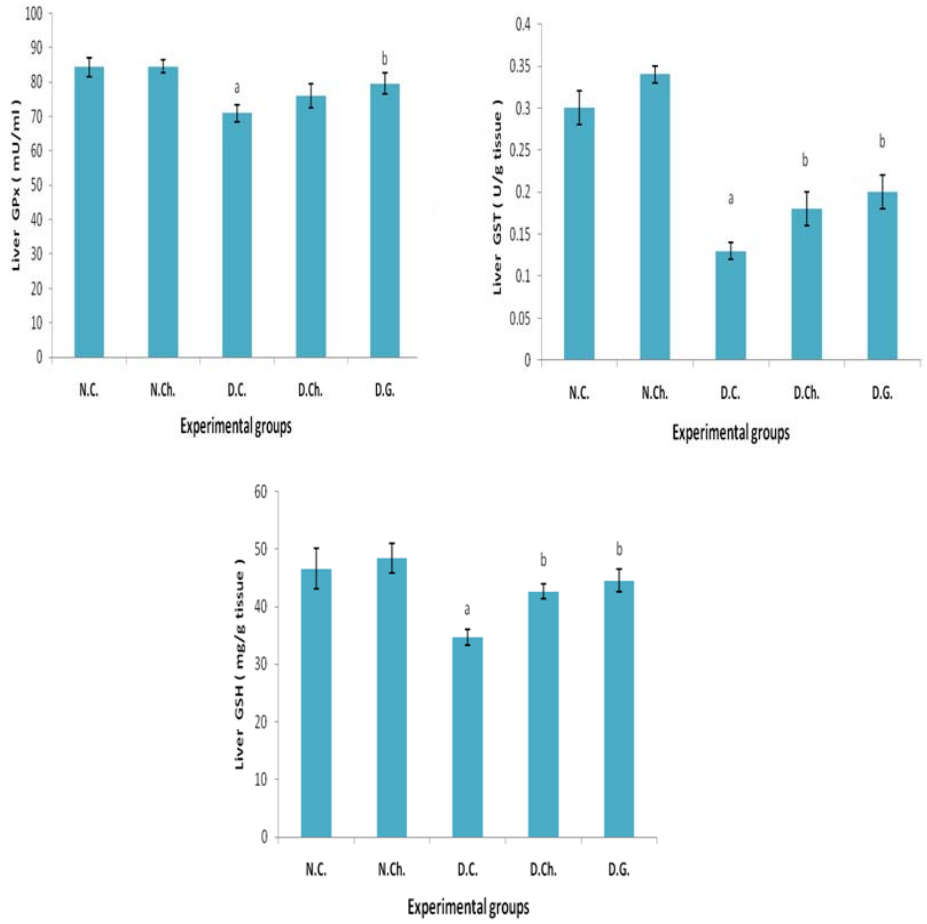


Fig. 3: Liver GPx, GST activity and GSH level (Mean ± SE) of experimental groups.
a Significant (P<0.05) as compared with the normal control (N.C.).
b Significant (P<0.05) as compared with the diabetic control (D.C.).

Table 2: Effect of treatment with aqueous suspension of *C. endivia* leaves powder or glibenclamide drug for 6 weeks on liver CAT, SOD, GPx and GST activities as well as GSH and MDA levels of diabetic rats.

Parameters	Experimental groups				
	N.C.	N.Ch.	D.C.	D.Ch.	D.G.
CAT (U/g tissue)	0.49 ± 0.10	0.46 ± 0.11	0.72 ± 0.01 [a]	0.66 ± 0.03	0.51 ± 0.04 [b]
% of change from N.C.			46.94		
% of change from D.C.				-8.33	-29.17
SOD (U/g tissue)	655.89 ± 9.96	602.55 ± 14.01 [a]	760.30 ± 11.35 [a]	688.01 ± 13.75 [b]	666.53 ± 5.95 [b]
% of change from N.C.			15.92		
% of change from D.C.				-9.51	-12.33
GPx (mU/ml)	84.26 ± 2.70	84.41 ± 1.90	70.93 ± 2.55 [a]	76.05 ± 3.47	79.54 ± 2.99 [b]
% of change from N.C.			-15.82		
% of change from D.C.				7.22	12.14
GST (U/g tissue)	0.30 ± 0.02	0.34 ± 0.01	0.13 ± 0.01 [a]	0.18 ± 0.02 [b]	0.20 ± 0.02 [b]
% of change from N.C.			-56.67		
% of change from D.C.				38.46	53.85
GSH (mg/g tissue)	46.59 ± 3.47	48.43 ± 2.56	34.64 ± 1.37 [a]	42.63 ± 1.32 [b]	44.54 ± 1.94 [b]
% of change from N.C.			-25.65		
% of change from D.C.				23.06	28.58
MDA (nmol/g tissue)	168.77 ± 11.54	145.83 ± 10.30	222.56 ± 12.08 [a]	214.48 ± 1.79	195.93 ± 3.14 [b]
% of change from N.C.			31.87		
% of change from D.C.				-3.63	-11.96

All data are mean of seven rats ± SE.
[a] Significant ($P < 0.05$) as compared to the control group (N.C.).
[b] Significant ($P < 0.05$) as compared to the diabetic group (D.C.).

Discussion:

Streptozotocin-induced hyperglycemia has been described as a useful experimental model to study the activity of hypoglycemic agents (Junod *et al.,* 1969). It damages the β-cells of the islets of Langerhans also hepatocytes, nephrons and cardiomyocytes (Selvan *et al.,* 2008). STZ induces oxidative stress, which results from enhanced free radical formation and/or defects in antioxidants defense causes severe tissue damage and may lead to number of diseases like coronary artery disease, atherosclerosis, cancer and diabetes (Chakraborty and Das, 2010). The present study was designed to investigate the effect of aqueous suspension of *C. endivia* leaves powder or glibenclamide drug on some biochemical parameters in STZ-induced diabetic rats.

Aminotransferases (AST and ALT) mediate the catalysis of aminotransfer reactions and they are markers for clinical diagnosis of liver injury (Li *et al.,* 2007). Alkaline phosphatase (ALP) is a hydrolase enzyme responsible for removing phosphate group from nucleotides and proteins, it is produced primarily in liver and brain (Han *et al.,* 2006), and it is a marker of hepatic functions (Yoo *et al.,* 2008). Lactate dehydrogenase (LDH) is a general indicator of acute or chronic hepatic damage, as well as determining organ, cell and tissue condition (Yoo *et al.,* 2008). The present study demonstrated significant increase in the activities of serum AST, ALT, ALP, and LDH of diabetic rats as compared to the control group. This increase was mainly due to the leakage of these enzymes from the liver cytosol into the blood stream (Navarro *et al.,* 1993), which gives an indication on the hepatotoxic effect of STZ. Ohaeri (2001) also found that liver was necrotized in STZ-induced diabetic rats. Experimental diabetes induced by STZ in rats causes liver tissue damage, which is reflected in the increase of serum AST and various hepatic enzymes, such as ALT (Maiti *et al.,* 2004). Khan *et al.* (2010) suggested that the elevation in serum AST and ALT activities in diabetic rats, could be related to excessive accumulation of amino acids (glutamate and alanine) in the serum of diabetic animals, as a result of amino acids mobilization from protein stores. Treatment with either aqueous suspension of *C. endivia* leaves powder or glibenclamide drug produced significant decrease in serum AST, ALT, ALP, and LDH activities, as compared to the diabetic group. It seems that *C. endivia* leaves powder or glibenclamide drug ameliorate the changes observed in the various hepatic enzymes of STZ-induced diabetic rats. The *C. endivia* leaves powder is better than glibenclamide drug in lowering serum ALT activity. Meanwhile, the effect of glibenclamide drug is more pronounced than that of *C. endivia* leaves powder in reducting the activity of serum ALP of diabetic rats. Aqueous suspension of *C. endivia* leaves powder and glibenclamide drug nearly appeared to have the same effect in decreasing the activity of serum AST and LDH of diabetic rats.

Superoxide dismutase (SOD) is one of the important antioxidant enzymes and scavenges the superoxide radicals by converting them to H_2O_2 and molecular oxygen (Selvan *et al.,* 2008). Catalase (CAT) is a hemoprotein which catalyzes the reduction of hydrogen peroxide and protects the tissue from highly reactive hydroxyl radicals (Searle and Wilson, 1980). In the present study, hepatic SOD and CAT of the STZ-induced diabetic rats were increased significantly as compared to the control group. The elevation of liver SOD activity

may be due to the increase dismutation of superoxide anions, and the increase in the SOD activity may inturnprotect CAT against enzyme inactivation by $\bar{O}_2$ anions, as these anions have been shown to inactive CAT (Selvam and Anuradha, 1990). It is possible that the increase in oxygen radical during diabetes could increase CAT activity, which in turn would protect SOD inactivation by H_2O_2 and hence caused an increase in SOD activity, H_2O_2 is known to inactivate SOD (Bray *et al.,* 1974). In accordance with our study, Cho *et al.* (2002) recorded an increase in CAT and SOD activities in the liver of STZ-induced diabetic rats. Regarding the effect of glibenclamide drug administration to diabetic rats there was significant reduction in hepatic SOD and CAT activities as compared to the diabetic group. However, treatment of the diabetic rats with aqueous suspension of *C. endivia* leaves powder produced only a significant decrease in their liver SOD activity.

Glutathione peroxidase (GPx), a selenium containing enzyme present in significant concentrations, detoxifies H_2O_2 to H_2O through the oxidation of reduced glutathione (Bruce *et al.,* 1982). It is sensitive to lower concentration of H_2O_2 (Bagri *et al.,* 2009). Glutathione-S-transferase (GST) catalyzes the reduction of hydrogen peroxide and hydroperoxides to non-toxic products (Bruce *et al.,* 1982). It catalyzes the conjugation of glutathione to a wide range of electrophiles and represents a protective mechanism against oxidative stress (Nowier *et al.,* 2009). The present investigation revealed significant decrease in liver GPx and GST activities in the STZ-induced diabetic rats, as compared to the control group. In accordance with our results, Schettler *et al.* (1994) demonstrated that the reduced antioxidant production may be due to the increase in oxygen metabolites that causes a decrease in the activity of the antioxidant defense system. Moreover, Kennedy and Baynes (1984) reported that the decrease in antioxidant enzyme activity in diabetes mellitus may be due to non-enzymatic glycosylation of the enzymes. Again, Al-Wabel *et al.* (2008) suggested that the depletion of GSH content also may lower GST enzyme, because GSH is required as a substrate for GST activity. Treatment the diabetic rats with glibenclamide drug increased significantly the decline observed in the activities of liver GPx and GST, as compared to the diabetic group. Meanwhile, a significant increase in liver GST activity of diabetic rats was recorded after treatment with *C. endivia* leaves powder aqueous suspension, as compared to the diabetic group.

Reduced glutathione (GSH) normally plays the role of an intracellular radical scavengers and it is the substrate of many xenobiotic elimination reactions (Gregus *et al.,* 1996). It protects the cellular system against the toxic effect of lipid peroxidation (Bagri *et al.,* 2009). Viewed in conjunction with the report of Shabeer *et al.* (2009), the present reduction in hepatic GSH level following STZ administration may be probably due to its increased utilization by the hepatic cells, which could be result of decreased synthesis or increased degradation of GSH by oxidative stress in diabetes. Again, Veerapur *et al.* (2010) reported that the generation of oxygen radicals by the increase in the level of glucose caused tissue GSH depletion, which attempted to overcome the deleterious effects of lipid peroxidation. The present study revealed that liver GSH level of diabetic rats increased significantly after treatment with either *C. endivia* leaves powder aqueous suspension or glibenclamide drug, as compared to the diabetic group.

In diabetes, hypoinsulinaemia increased the activity of the enzyme fatty acyl coenzyme and coenzyme A oxidase, which initiated β-oxidation of fatty acids resulting in lipid peroxidation (Baynes, 1995). The increase in oxygen free radicals in diabetes could be primarily due to the increase in the blood glucose level, which upon autoxidation generated free radicals, and secondarily due to the effect of the diabetogenic agent (streptozotocin or alloxan) (Szkudelski, 2001). Malondialdehyde (MDA) is one of the lipid peroxidation products frequently used to determine the oxidant/antioxidant balance in diabetic patient (Cheeseman and Slate, 1993). The present study revealed that the hepatic MDA level increased significantly after intraperitoneal injection of STZ as compared to the control group. In agreement with our study Punitha *et al.* (2006), Selvan *et al.* (2008) and Chakraborty and Das (2010) reported that the administration of STZ in rats elevated hepatic MDA level. The elevation in hepatic MDA may be due to the high concentration of lipid, which is one of the characteristic remarks in liver of diabetic rats, which resulted in the activation of NADPH dependent microsomal lipid peroxidation in liver (Patel *et al.,* 2009). Treatment with glibenclamide drug showed significant decrease in liver MDA level of diabetic rats. But after treatment with *C. endivia* leaves powder aqueous suspension, the level of liver MDA of diabetic rats showed insignificant decrease, as compared to the diabetic group. The results suggest that aqueous suspension of *C.endivia* leaves powder or glibenclamide drug may effectively normalize the impaired antioxidants status in STZ-induced diabetes. Moreover, glibenclamide drug may be more powerful free radical scavenger than *C. endivia* leaves powder.

In conclusion, the results obtained clearly indicate the role of oxidative stress in induction of diabetes and suggest the ameliorative effect of *C.endivia* leaves powder aqueous suspension, similar to the diabetic drug (glibenclamide) at least in such animal model. However, further studies are necessary to isolate and identify the principle components of *C. endivia* leaves powder and elucidate the mechanisms of their action.

REFERENCES

Aebi, H., 1984. Catalase *in vitro*. Methods in Enzymology, 105: 121-126.

Agarwal, M., 1980. Streptozotocin: mechanism of action. FEBS Lett, 120: 1-3.

Al-Wabel, N.A., H.M. Mousa, O.H. Omer and A.M. Abdel-Salam, 2008. Biological evaluation of aqueous herbal extracts and stirred yoghurt filtrate mixture against alloxan-induced oxidative stress and diabetes in rats. Int. J. Pharmacol., 4: 135-139.

Bagri, P., M. Ali, V. Aeri, M. Bhowmik, and S. Sultana, 2009. Antidiabetic effect of *Punica granatum* flowers: Effect on hyperlipidemia, pancreatic cells lipid peroxidation and antioxidant enzymes in experimental diabetes. Food Chem. Toxicol., 47: 50-54.

Baynes, J.W., 1995. Reactive oxygen in the aetiology and complications of diabetes. In: Ioannides, C., Flatt, P.R. (Eds.), Drug, Diet and Disease, Mechanistic Approach to Diabetes Volume 2. Ellis Horwood Limited, Hertfordshive, pp: 230-231.

Belfield, A. and D.M. Goldberg, 1971. Revised assay for serum phenylphosphatase activity using 4-amino-antipyrine. Enzyme, 12: 561-573.

Beutler, E., O. Duron, and B.M. Kelly, 1963. Improved method for the determination of blood glutathione. J. Lab. Clin. Med., 61: 882-888.

Bopanna, K.N., J. Kannan, S. Godgil, R. Balaraman, S.P. Rathod, 1997. Antidiabetic and antihyperlipidemic effects of neem seed kernel powder on alloxan-diabetic rabbits. Indian J. Pharmacol., 29: 162-72.

Bray, R.C., S.A. Cockle, E. Martin-Fielder, P.B. Roberts, G. Rotillo, and L. Calabrase, 1974. Reduction and inactivation of superoxide dismutase by hydrogen peroxide. Biochem. J., 139: 43-48.

Bruce, A., D. Freeman, and C. James, 1982. Biology of disease-free radicals and tissue injury. Lab. Invest., 47: 412-426.

Chakraborty, U. and H. Das, 2010. Antidiabetic and Antioxidant activities of *Cinnamomum tamala* leaf extracts in STZ-treated diabetic rats. Global J. Biotechnol. Biochem., 5(1): 12-18.

Chandramohan, G., K.S. Al-Numair, and K.V. Pugalendi, 2009. Restoration of altered plasma, erythrocyte and liver antioxidant levels by 3-hydroxymethyl xylitol in streptozotocin-diabetic rats. Int. J. Integr. Biol., 5: 176-181.

Cheeseman, K.H. and T.F. Slate, 1993. An introduction to free radical biochemistry. Br. Med. Bull., 49: 481-493.

Cho, S.Y., J.Y. Park, E.M. Park, M.S. Choi, M.K. Lee, S.M. Jeon, M.K. Jang, M.J. Kim, and Y.B. Park, 2002. Alternation of hepatic antioxidant enzyme activities and lipid profile in streptozotocin-induced diabetic rats by supplementation of dandelion water extract. Clin. Chim. Acta, 317: 109-117.

Damasceno, D.C., G.T. Volpato, M. Paranhos-Calderon, and M.V. Cunha-Rudge, 2002. Oxidative stress and diabetes in pregnant rats. Anim. Reprod. Sci., 15(72): 235-244.

Eliza, J., P. Daisy, S. Ignacimuthu, and V. Duraipandiyan, 2009. Normo-glycemic and hypolipidemic effect of costunolide isolated from *Costus speciosus* (Koen ex. Retz.)Sm. in streptozotocin-induced diabetic rats. Chem. Biol. Interact., 179: 329-334.

Fernald, M.L., 1950. Gray's Manual of Botany. 8th ed. American Book Co., New York.

Gregus, Z., T. Fekete, E. Halaszi, and C.D. Klaassen, 1996. Lipoic acid impairs glycine conjugation of benzoic acid and renal excretion of benzoylglycine. Drug Metab. Dispos., 24: 682-688.

Habig, W.H., M.J. Pabst, and W.B. Jakoby, 1974. Glutathione-S-transferase: the first enzymatic step in mercapturic acid formation. J. Biol. Chem., 249: 7130-7139.

Han, K.H., N. Hashimoto, K. Shimada, M. Sekikawa, T. Noda, H. Yamauchi, M. Hashimoto, H. Chiji, D.L. Topping, and M. Fukushima, 2006. Hepatoprotective effects of purple potato extract against D-galactosamine-induced liver injury in rats. Biosci. Biotechnol. Biochem., 70: 1432-1437.

Junod, A., A.E. Lambert, W. Stauffacher, and A.E. Renold, 1969. Diabetogenic action of streptozotocin. Relationship of dose to metabolic response. J. Clin. Invest., 48: 2129-2139.

Kaleem, M., Sheema, H. Sarmed, and B. Bano, 2005. Protective effects of *Piper nigrum* and *Vinca rosea* in alloxan-induced diabetic rats. Indian J. Physiol. Pharmacol., 49: 65-71.

Kameswara, R.B. and R.C.H. Appa, 2001. Hypoglycemic and antihyperglycemic activity of alternifolium Walp. Seed extracts in normal and diabetic rats. Phytomed., 8: 88-93.

Kanter, M., I. Meral, Z. Yener, H. Ozbek, and H. Demir, 2003. Partial regeneration/proliferation of the β-cells in the islets of Langerhans by Nigella Sativa L. in streptozotocin-induced diabetic rats. Tohoku J. Exp. Med., 201(4): 213-219.

Kennedy, L. and J.W. Baynes, 1984. Non-enzymatic glycosylation and the chronic complications of diabetes: An overview. Diabetologia, 26(2): 93-98.

Kesavulu, M.M., R. Giri, R.B. Kameswara, and C. Apparao, 2000. Lipid peroxidation and antioxidant enzyme levels in type 2 diabetic with microvascular complications. Diabetic Metabol., 26: 387-392.

Khan, M.R.I., M.A. Islam, M.S. Hossain, M. Asadujjaman, M.I.I. Wahed, B.M. Rahman, A.S.M. Anisuzzaman, S.M. Shaheen, and M. Ahmed, 2010. Antidiabetic effects of the different fractions of ethanolic extracts of *Ocimum sanctum* in normal and alloxan induced diabetic rats. J. Sci. Res., 2(1): 158-168.

Kim, M.J., G.R. Ryu, J.S. Chung, S.S. Sim, D.J. Min Dos Rhie, S.H. Yoon, S.J. Hahn, M.S. Kim, and Y.M. Jo, 2003. Protective effect of epicatechin against the toxic effects of STZ on rat pancreatic islets: in vivo and in vitro. Pancreas, 26: 292-299.

Kopeck, K., 1998. Tabulky nutričních hodnot ovoce a zeleniny. Praha, ÚZPI: 72.

Kumari, K., B.C. Mathew, and K.T. Augusti, 1995. Antidiabetic and hypolipidemic effect of S-methyl cysteine sulfoxide isolated from Allium cepa Linn. Indian J. Biochem. Biophy., 32: 49-54.

Li, B., Z. Wang, J.J. Fang, C.X. Xu, and W.X. Chen, 2007. Evaluation of prognostic markers in severe drug-induced liver disease. World J. Gastroenterol., 13(4): 628-632.

Maiti, R., D. Jana, U.K. Das, and D. Ghosh, 2004. Antidiabetic effect of aqueous extract of seed of *Tamarindus indica* in streptozotocin-induced diabetic rats. J. Ethnopharmacol., 92: 85-91.

Mansour, H.A., A.S.A. Newairy, M.I. Yousef, and S.A. Sheweita, 2002. Biochemical study on the effects of some Egyptian herbs in alloxan-induced diabetic rats. Toxicology, 170: 221-228.

Navarro, C.M., P.M. Montilla, A. Martin, J. Jimenez, and P.M. Utrilla, 1993. Free radicals scavenger and antihepatotoxic activity of Rosmarinus. Plant Med., 59: 312-314.

Nayeemunnisa, A., 2009. Alloxan diabetes-induced oxidative stress and impairment of oxidative defense system in rat brain: neuroprotective effects of *Cichorium intybus*. Int. J. Diabetes Metab., 17: 105-109.

Nishikimi, M., N.A. Roa, and K. Yogi, 1972. The occurrence of superoxide anion in the reaction of reduced phenazine methosulfate and molecular oxygen. Biochem. Bioph. Res. Common., 46(2): 849-854.

Nowier, S.R., N.K. Kashmiry, H.A. Abdel Rasool, H. Morad, and S. Ismail, 2009. Association of type 2 diabetes mellitus and glutathione-S-transferase (GSTM1 and GSTT1) genetic polymorphism. Res. J. Medicine Med. Sci., 4(2): 181-188.

Ogbonnia, S.O., J.I. Odimegwu, and V.N. Enwuru, 2008. Evaluation of hypoglycemic and hypolipidemic effects of ethanolic extracts of Treculia africana Decne and Bryophyllum pinnatum Lam. and their mixture on streptozotocin (STZ)-induced diabetic rats. Afr. J. Biotechnol., 7(15): 2535-2539.

Ohaeri, O.C., 2001. Effect of garlic oil on the levels of various enzymes in the serum and tissue of streptozotocin diabetic rats. Biosci. Rep., 21: 19-24.

Ohkawa, H., W. Ohishi, and K. Yagi, 1979. Assay for lipid peroxides in animal tissue by thiobarbituric acid reaction. Anal. Biochem., 95(2): 351-358.

Owu, D.U., A.B. Antai, K.H. Udofia, A.O. Obembe, K.O. Obasi, and M.U. Eteng, 2006. Vitamin C improves basal metabolic rate and lipid profile in alloxan-induced diabetes mellitus in rats. J. Biosciences, 31(5): 575-579.

Paglia, D.E. and W.N. Valentine, 1967. Studies on the qualitative characterization of erythrocyte glutathione peroxidase. J. Lab. Clin. Med., 70: 158-169.

Patel, S.S., R.S. Shah, and R.K. Goyal, 2009. Antihyperglycemic, antihyperlipidemic and antioxidant effects of Dihar, a polyherbal ayurvedic formulation in streptozotocin-induced diabetic rats. Ind. J. Exp. Biol., 47: 564-570.

Petrovic, J., A. Stanojkovic, L.J. Comic, and S. Curcic, 2004. Antibacterial activity of *Cichorium intybus*. Fitoterapia, 75(7-8): 737-739.

Punitha, I.S.R., A. Shirwaikar, and A. Shirwaikar, 2006. Antidiabetic activity of benzyl tetra isoquinoline alkaloid berberine in streptozotocin-nicotinamide induced type 2 diabetic rats. Diabetologia Croatica, 34: 117-128.

Pushparaj, P.N., H.K. Low, J. Manikandan, B.K.H. Tan, and C.H. Tan, 2007. Anti-diabetic effects of *Cichorium intybus* in streptozotocin-induced diabetic rats. J. Ethnopharmacol., 111(2): 430-434.

Reitman, S. and S. Frankel, 1957. A colorimetric method for the determination of serum glutamic oxaloacetic and glutamic pyruvic transaminases. Amer. J. Clin. Pathol., 28: 56-58.

Sadeghi, H., N.M. Reza, G. Izadpanah, and S. Sohailla, 2008. Hepatoprotective effect of *Cichorium intybus* on CCl_4-induced liver damage in rats. Afr. J. Biochem. Res., 2(6): 141-144.

Saxena, A.K., P. Srivastava, R.K. Kale, and N.Z. Baquer, 1993. Impaired antioxidant status in diabetic rat liver. Effect of vanadate. Biochem. Pharmacol., 45(3): 539-542.

Schettler, V., E. Wieland, R. Verwiebe, P. Schuff-Werner, F. Scheler, and M. Oellerich, 1994. Plasma lipids are not oxidized during hemodialysis. Nephron, 67: 42-47.

Scoppola, A., F.R. Montecchi, G. Mezinger, and A. Lala, 2001. Urinary mevalonate excretion rate in type 2 diabetes: role of metabolic control. Atherosclerosis, 156: 357-361.

Searle, A.J. and R.L. Wilson, 1980. Glutathione peroxidase: effect of superoxide, hydroxyl and bromine free radicals on enzymic activity. Int. J. Radi. Biol., 37: 213-217.

Selvam, R. and C.V. Anuradha, 1990. Effect of oral methionine on tissue lipid peroxidation and antioxidants in alloxan-induced diabetic rats. J. Nutr. Biochem., 1: 653-658.

Selvan, V.T., L. Manikandan, G.P. Senthil Kumar, R. Suresh, B.B. Kakoti, P. Gomathi, D.A. Kumar, P. Saha, M. Gupta, and U.K. Mazumder, 2008. Antidiabetic and antioxidant effect of methanol extract of *Artanema sesamoides* in streptozotocin-induced diabetic rats. International Journal Applied Research in Natural Products, 1(1): 25-33.

Shabeer, J., R.S. Srivastava, and S.K. Singh, 2009. Antidiabetic and antioxidant effect of various fractions of *Phyllanthus simplex* in alloxan diabetic rats. J. Ethnopharmacol., 124: 34-38.

Szkudelski, T., 2001. The mechanism of alloxan and streptozotocin action in β-cells of the rat pancreas. Physiol. Res., 50: 536-546.

Ugochukwu, N.H., N.E. Babady, M. Cobourne, and S.R. Gasset, 2003. The effect of *Gangronema latifolium* extracts on serum lipid profile and oxidative stress in hepatocytes of diabetic rats. J. Biosciences, 28(1): 1-5.

Upur, H., N. Amat, B. Blažeković, and A. Talip, 2009. Protective effect of *Cichorium glandulosum* root extract on carbon tetrachloride-induced and galactosamine-induced hepatotoxicity in mice. Food Chem. Toxicol., 47: 2022-2030.

Vassault, A., D. Grafmeyer, C.I. Naudin, G. Dumont, M. Bailly, J. Henny, M.F. Gerhardt, and P. Georges, 1986. Protocole for the validation of methods . Ann. Biol. Clin., 44: 686-745.

Veerapur, V.P., K.R. Prabhakar, V.K. Parihar, P. Bansal, K.K. Srinivasan, K.I. Priyadarsini, and M.K. Unnikrishnan, 2010. Antidiabetic, hypolipidaemic and antioxidant activity of *Dodonaea viscosa* aerial parts in streptozotocin-induced diabetic rats. International Journal of Phytomedicine, 2: 59-70.

Yoo, Y.M., J.H. Nam, M.Y. Kim, J. Choi, and H.J. Park, 2008. Pectolinarin and pectolinarigenin of *Cirsium setidens* prevent the hepatic injury in rats caused by D-galactosamine *via* an antioxidant mechanism. Biol. Pharm. Bull., 31(4): 760-764.