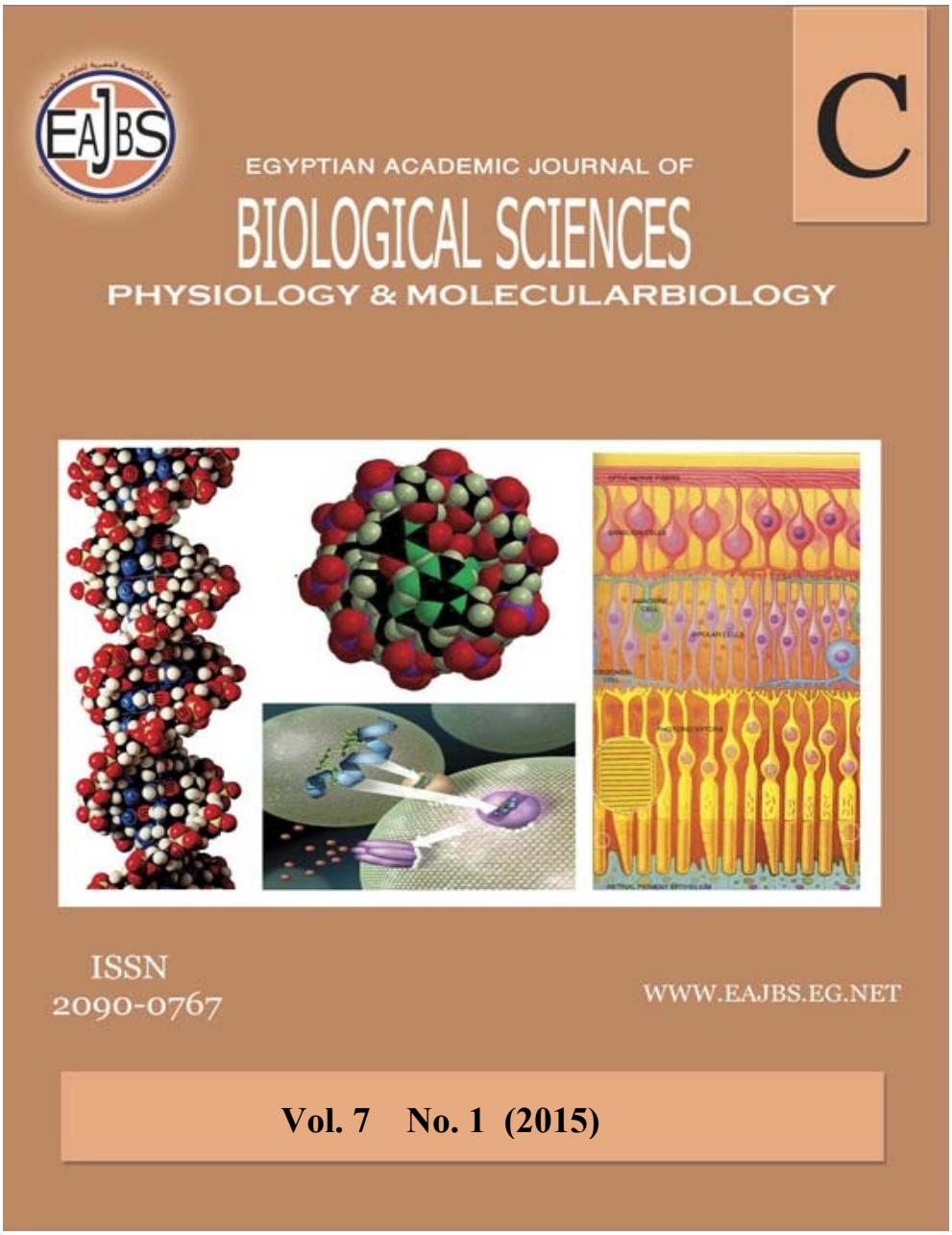

Egyptian Academic Journal of Biological Sciences is the official English language journal of the Egyptian Society for Biological Sciences, Department of Entomology, Faculty of Sciences Ain Shams University .

Physiology & molecular biology journal is one of the series issued twice by the Egyptian Academic Journal of Biological Sciences, and is devoted to publication of original papers that elucidateimportant biological, chemical, or physical mechanisms of broad physiological significance.

www.eajbs.eg.net

Egypt. Acad. J. Biolog. Sci., 7(1): 1 -10 (2015)

Egyptian Academic Journal of Biological Sciences
C. Physiology & Molecular Biology
ISSN 2090-0767
www.eajbs.eg.net

Evaluation of ameliorative effect of allicin (diallyl thiosulfinate) on experimentally induced diabetes mellitus in albino rats

Sasikumar Dhanarasu
Department of Biochemistry, College of Medicine, University of Hail, Kingdom of Saudi Arabia.
Email: drdskumar31@yahoo.com, s.dhanarasu@uoh.edu.sa

ARTICLE INFO
Article History
Received:4/1/2015
Accepted: 15/3/2015

***Keywords*:**
Allicin
Oxidative stress
Inflammatory markers
TNF-α
IL-6
Lipidperoxidation

ABSTRACT

Background: Diabetes causes increased oxidative stress, to play an important role in the pathogenesis of various diabetic complications. The present study investigated the protective potential of allicin against diabetes in rat model.

Methodology: Animals were injected a single dose of streptozotocin (55mg/kg.b.wt) to induce diabetes. Control and diabetic rats were treated with allicin (20 mg/kg.b.wt) for 30 days. Glibenclamide (10mg/kg.b.wt) was used as a positive control. After the experimental regimen, the rats were sacrificed and the blood was collected and serum was separated. Blood sugar, body weight, total protein, oxidative stress markers (TBARS, CAT, SOD and GSH) and inflammatory markers (TNF-α and IL-6) were used to bio-monitor the protective potential of allicin.

Results: The experimental animals lost weight significantly, exhibited elevated blood sugar levels, decreased protein levels (p<0.05) in comparison to control animals; these alteration were corrected after allicin therapy. The significant elevation of TBARS content, reduction in total glutathione content and declined CAT and SOD activities in experimental animals; which were normalized following allicin therapy. The altered serum inflammatory markers were significantly reduced to near normal levels. No significant alterations in allicin alone administered rats.

Conclusion: We thus conclude that allicin has potent antihyperglycemic, anti-lipidperoxidative and anti- inflammatory potential in diabetic animals.

INTRODUCTION

Diabetes is a common life-long health condition caused by an absolute or relative lack of insulin and is characterized by hyperglycemia in the postprandial and fasting state. The number of people with type I and type II diabetes are dramatically increasing on a global scale (Leila, *et al*. 2007). The World Health Organization (WHO) estimates that more than 180 million people in the world have diabetes and that this number will double by 2030 (WHO, 2009).

Diabetes is a chronic metabolic disorder with numerous complications such as retinopathy, neuropathy, and peripheral vascular insufficiencies (Chehade and Mooradian, 2000).

Hyperglycemia induced oxidative stress results in overproduction of reactive oxygen species (ROS) leading to an imbalance between free radical production and the ability of the antioxidant defense system to remove free radicals (Ugochukwu and Cobourne, 2003). Streptozotocin (STZ) acts as a diabetogenic agent due its capacity to destroy pancreatic β-cells, possibly through a free radical mechanism (Halliwell and Gutteridge, 1994). Various hypotheses to explain the genesis of free radicals in diabetes include autoxidation processes of glucose, the non-enzymatic and progressive glycation of proteins with the consequently increased formation of glucose-derived advanced glycosylation end products (AGEs), and enhanced glucose flux through the polyol pathway (Tiwari and Rao, 2002).

The generated ROS produce oxidative stress and exert major effects on signaling pathways, which further influence cellular metabolism and trigger a low-grade inflammatory reaction (Dominiczak, 2003). Moreover, glucose in itself is pro-inflammatory and increases the levels of acute-phase inflammatory markers, including tumor necrosis factor-α (TNF-α), interleukin-6 (IL-6) and C-reactive protein (CRP). These acute-phase inflammatory markers are associated with insulin resistance and metabolic syndrome, suggesting a role for chronic low-grade inflammation in diabetes mellitus (Niehoff, *et al*. 2007).

Diabetes is still not completely curable by the present antidiabetic therapy. Insulin therapy is the only satisfactory approach in diabetic mellitus, even though it has several drawbacks like insulin resistance (Piédrola, *et al*. 2001), anorexia, brain atrophy, and fatty liver in chronic treatment (Weidmann, *et al*. 1993). There are several oral hypoglycemic agents used therapeutically but certain adverse effects and weak effectiveness of them have led to the search for more effective agents. WHO recommended that medicinal plant research warrant attention to search for anti-diabetic properties (WHO, 1980). Our previous studies indicated that medicinal plant modulates the effect of circulatory antioxidants against oral carcinogenesis (Sasikumar, *et al.* 2010). The protective effects of medicinal plants have been attributed to various phytoconstituents, such as carotenoids, vitamin C and phenolic acids (Tan, *et al.* 2011). In particular, several studies have also focused on the biological activities of flavonoids, a group of plant polyphenolic compounds that possess potent antioxidant effects (Xiao, *et al.* 2011).

Allicin, diallyl thiosulfinate (or S-(2-propenyl) 2-propene-1-sulfinothioate), is the main biologically active compound derived from garlic (*Allium sativum* L.). Chemical structure of allicin is show in Fig. 1.

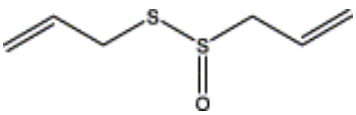

Fig. 1: Chemical structure of allicin [S-(2-propenyl) 2-propene-1-sulfinothioate or diallyl thiosulfinate].

It is produced by the interaction of the enzyme alliinase. Allicin became an object of interest due to its potential to confer a vast spectrum of health beneficial effects including: anti-microbial, antifungal and antiparasitic (Koch and Lawson, 1996), antihypertensive (Elkayam, *et al.* 2001), cardioprotective (Gonen, *et al.* 2005), anti-inflammatory (Lang, *et al*. 2004) and anticancer activities (Koch and Lawson, 1996, Hirsch, *et al*. 2000).

However, no evidence is available on the ability of allicin on ameliorative effects associated with free radical scavenging and inflammatory response on diabetes. Hence, the present study was designed to evaluate the therapeutic efficacy of allicin on diabetic animals.

MATERIALS AND METHODS

Animals

Male albino rats (Wistar Stain), weighing 250–290 g were used for the study. The animals were obtained from the Central Animal House, King Saud University, Riyadh, KSA. The rats were housed in polypropylene cages and were maintained in controlled atmosphere (temperature of 22±2°C and 55±5 % humidity) with a 12 hr light:dark (LD) cycles in an experimental room. The normal laboratory diet (Manufactured by Grain Silos and Flour Mills Organization, Riyadh, KSA) and water given *ad libitum*.

Chemicals

Streptozotocin was obtained from Sigma Aldrich Chemical Pvt Limited, Bangalore, India. Allicin was purchased from Shanghai Harvest Pharmaceutical Co., Ltd. (Shanghai, China). All other chemicals used were of analytical grade.

Induction of diabetes

The STZ was dissolved in citrate buffer (pH 4.5), and rats were made diabetic by injection of a single dose of STZ (55 mg/kg, i.p). They were given 5% of glucose in drinking water for the first 24 h to encounter any initial hypoglycemia. On the 3rd day animals were checked for serum blood glucose levels and those with more than 300 mg/dl were used in the study (Rathod, *et al*. 2008).

Experimental design

The rats were divided into 5 groups of 6 rats each as follows. Group I – Normal animals injected with vehicle (citrate buffer), Group II, III and IV rats were induced diabetes. After diabetic induction group II received vehicle only. Group III diabetic rats were treatment with allicin (20 mg/kg body weight, orally) for 30 days by gastric intubation using force-feed needle. Group IV diabetic received glibenclamide (10 mg/kg, p.o.). Group V rats received oral administration of allicin alone throughout the experimental period. After the experimental regimen, the rats were fasted overnight and were sacrificed by cervical dislocation under light ether anesthesia, and the blood was collected on decapitation and serum was separated by centrifugation (20 min at 2000 rpm) and stored at –20ºC for biochemical assays.

Determination of oxidative stress markers

Thiobarbituric acid reactive substances (TBARS) were assayed in plasma according to the methods of Yagi, (1987). Plasma was deproteinised with 10% phosphotungstic acid and the precipitate was treated with thiobarbituric acid at 90^{o}C for 1 hour. After cooling, 5.0ml of n-butanol was added and the mixture was shaken vigorously and centrifuged at 1000g for 15 minutes. The pink color formed gives a measure of TBARS. Reduced glutathione (GSH) was determined by the method of Beutler and Kelly, (1963). The technique involved in protein precipitation by meta phosphoric acid and spectrophotometric assay at 412 nm of the yellow derivative obtained by the reaction of supernatant with 5,5'-

dithio-bis-2-nitrobenzoic acid (DTNB). Superoxide dismutase (SOD) activity was assayed by the method of Kakkar, *et al.* (1984). The assay of superoxide dismutase was based on the inhibition of formation of NADH-phenazine methosulphate nitroblue tetrazolium formazan. The colour formed at the end of the reaction was extracted into butanol layer and measured at 520nm. Catalase (CAT) activity was assayed using the method of Sinha, (1972). The method is based on the utilization of H_2O_2 by the enzyme. The color developed was read at 620 nm.

Estimation of sugar and protein

Blood glucose was determined by the method of Sasaki and Matsy, (1972) using O-toluidine reagent. The protein content was estimated using a previously reported method (Lowry, 1951).

Determination of inflammatory markers

Serum TNF-α concentration was estimated by using the CYTELISA rat TNF-α obtained from CYTIMMUNE Sciences Inc., Maryland, USA. The intra assay variation was 6.7%. To avoid inter assay variation all samples were run at one time. Optical density of each well was determined by using a microplate reader (Thermo Labsystems, Finland). Serum IL-6 was estimated by using the ELISA Quantikine rat IL-6 immunoassay kit obtained from R and D Systems Inc., Minneapolis, MN, USA. The intra assay variation was 5.5%. To avoid inter assay variation all samples were run at one time. Optical density of each well was determined by using a microplate reader (Thermo Labsystems, Finland).

Statistical Analysis

The analysis was done on SPSS 20.0 statistical package. The data are expressed as mean±SD. Statistical comparisons were performed by one way analysis of variance (ANOVA) followed by Duncan's Multiple Range Test (DMRT). The results were considered statistically significant at p-values less than 0.05.

RESULTS

Fig. 2 shows the effects of allicin on body weights of normal and experiment animals. Body weights were similar in all the groups at baseline. The body weights of 30 days diabetic rats were significantly lower than non-diabetic controls. The rats started regaining weight after instituting allicin therapy and reached their baseline levels within 1 month. Administration of allicin alone did not show any effect on body weights.

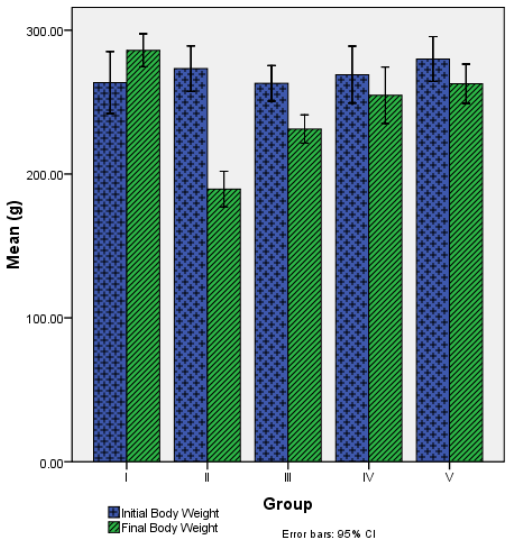

Fig. 2: Effects of allicin on body weight in diabetes induced albino rats.

The blood sugar and total protein levels of control and experimental animals in each group are depicted in Table 1. Blood glucose levels were elevated in the diabetic animals compared to control animals. However, this increase in blood sugar levels in allicin and reference drug (Glibenclamide) treated rats were significantly ($p<0.05$) decreased. The levels of serum protein were significantly decreased ($p<0.05$) in diabetic rats compared to control rats, whereas in allicin treated rats, these levels were significantly increased ($p<0.05$) compared to group II rats. The altered levels of blood sugar and serum protein were normalized by treated with glibenclamide. No significant alterations in these parameters were noted in allicin alone administered group of rats.

Table 1: Effects of allicin on blood sugar and serum protein in diabetic induced albino rats (Mean±SD; n = 6).

Parameters	Group I (Normal)	Group II (Diabetic)	Group III (Diabetic + allicin)	Group IV (Diabetic + Glibenclamide)	Group V (allicin alone)
Blood sugar (mg/dl)	99.33 ± 7.56^{a}	335.89 ± 22.92^{b}	126.65 ± 11.78^{c}	116.91 ± 11.21^{cd}	109.23 ± 8.62^{ad}
Serum protein (g/dl)	7.09 ± 0.44^{a}	4.03 ± 0.37^{b}	6.83 ± 0.59^{a}	6.98 ± 0.56^{a}	6.45 ± 0.55^{a}

Values not sharing a common superscript letter differ significantly at $p<0.05$.

Table 2 represents the activity of oxidative stress markers in the different experimental groups. There was significant elevation in the TBARS content, with a concomitant reduction in the total glutathione content in the diabetic rats. Treatment with allicin and glibenclamide reverses the TBARS content back to normal level and increases the GSH content. The activities of CAT and SOD enzymes were also found to be declined in the plasma of STZ-induced rats which was restored to normal levels on allicin and glibenclamide treatment. Allicin only administered rats registered no significant changes (group V).

Table 2: Status of TBARS and antioxidants in plasma of control and experimental animals (Mean±SD; n=6).

Parameters	Group I (Normal)	Group II (Diabetic)	Group III (Diabetic + allicin)	Group IV (Diabetic + Glibenclamide)	Group V (allicin alone)
TBARS (nmoles/mL)	3.22 ± 0.20^{a}	4.93 ± 0.38^{b}	3.57 ± 0.27^{c}	3.32 ± 0.25^{ac}	3.21 ± 0.24^{a}
CAT (nmol/min/ml)	84.39 ± 6.43^{a}	53.61 ± 3.47^{b}	80.83 ± 5.38^{a}	83.48 ± 4.88^{a}	83.01 ± 6.32^{a}
GSH (mg/dl)	21.90 ± 1.67^{a}	13.83 ± 1.28^{b}	19.73 ± 1.3^{c}	20.71 ± 1.41^{ac}	21.78 ± 1.66^{a}
SOD (U*/ml)	104.48 ± 6.52^{a}	70.00 ± 5.33^{b}	98.57 ± 6.32^{a}	102.02 ± 7.77^{a}	103.87 ± 6.33^{a}

*The amount of enzyme required to inhibit 50% nitroblue tetrazolium (NBT) reduction.
Values not sharing a common superscript letter differ significantly at $p<0.05$.

Table 3 shows the effects of allicin on inflammatory markers. STZ-treated rats compared with normal group I animals showed a significant rise in serum acute-phase inflammatory markers (TNF-α and IL-6). Treatment with allicin and Glibenclamide significantly reduced the serum TNF-α and IL-6 to near normal levels compared with group II diabetic animals. Administration of allicin alone

did not show any effect on inflammatory markers.

Table 3: Effects of allicin on inflammatory markers in diabetic induced albino rats (Mean±SD; n=6).

Parameters	Group I (Normal)	Group II (Diabetic)	Group III (Diabetic + allicin)	Group IV (Diabetic + Glibenclamide)	Group V (allicin alone)
TNF-α (pg/ml)	100.01±7.62[a]	199.90±15.22[b]	115.99±8.83[c]	103.92±6.93[ac]	111.89±8.52[ac]
IL-6 (pg/ml)	381.04±29.01[a]	596.46±36.54[b]	402.05±30.61[a]	399.61±30.43[a]	401.07±30.54[a]

Values not sharing a common superscript letter differ significantly at $p<0.05$.

DISCUSSION

Rats treated with STZ display many features seen in human beings with uncontrolled diabetes, including hyperglycemia, hypoinsulinemia, increased urinary glucose levels, and consequently polyurea as well as weight loss (De Angelis, *et al*. 2002). The destruction of β-cells during diabetes ultimately causes physic-metabolic abnormalities such as decrease in body weight gain and increase in food and water intake (Rodriguez, *et al*. 1997). In addition, diabetic rats showed a clear muscle atrophy involving a decrease in both skeletal muscle mass and protein content. These changes were related to important alterations in protein turnover in skeletal muscle (Pepato, *et al*. 1996). Hence, a notable decrease in the body weight change observed in the diabetic group of rats (group II) might be the result of protein wasting due to the unavailability of carbohydrates for energy metabolism (Brodsky, 1998). The improvement in body weight gain in diabetic rats supplemented with allicin highlight the body glucose homeostasis which in turn promotes the body weight gain.

Hyperglycemia causes oxidative stress by inducing the overproduction of ROS, which results in an imbalance between free radical production and the ability of the antioxidant defense system to remove free radicals (Ugochukwu and Cobourne, 2003). Increased oxidative stress is a widely accepted participant in the development and progression of diabetes and its complications (Ceriello, 2000). Diabetes is usually accompanied by increased production of free radicals or impaired antioxidant defenses (Saxena, *et al*. 1993). ROS results in lipid peroxidation and subsequently increased in TBARS levels leading to degradation of cellular macromolecules. A marked increase in the concentration of TBARS in STZ-induced diabetic rats indicates enhanced lipidperoxidation (LPO) leading to tissue injury and failure of the antioxidant defense mechanisms to prevent overproduction of ROS. It has been proposed that streptozotocin (STZ) acts as a diabetogenic agent owing to its ability to destroy pancreatic β-cells, possibly by a free radical mechanism (Halliwell and Gutteridge, 1994). The level of LPO in cell is controlled by various cellular defense mechanisms consisting of enzymatic and non-enzymatic scavenging systems which are altered in diabetes. Moreover, disturbances of antioxidant defense systems in diabetes showed alteration in antioxidant enzyme levels, such as superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and glutathione reductase (GR) along with impaired glutathione (GSH) metabolism (McLennan, Heffermen and Wright, 1991).

SOD and CAT are the two major scavenging enzymes that remove free

radicals *in vivo*. A decreased activity of these antioxidants of group II animals can lead to an excess availability of superoxide anion ($O2^{\cdot-}$) and hydrogen peroxide (H_2O_2), which in turn generate hydroxyl radicals ($\cdot OH$), resulting in initiation and propagation of LPO (Jin, *et al* 2008). The SOD and CAT play a prominent role in scavenging free radical and restoring antioxidant activities in the plasma and it protects cell protein and cell membranes against oxidative stress (Rahimi, *et al*. 2005) of diabetic animals. These observations may clearly indicate that the increased levels of SOD and CAT in allicin treated animals has free radical scavenging activity, which may exert a beneficial effect against pathological alterations caused by reactive oxygen species.

Glutathione plays an important role in the endogenous non-enzymatic antioxidant system. Primarily, it acts as reducing agent and detoxifies hydrogen peroxide in presence of an enzyme, glutathione peroxidase (Ketterer, *et al*. 1976). The depleted GSH may be due to reduction in GSH synthesis or degradation of GSH by oxidative stress (Loven, *et al*. 1986) in diabetic induced animals. Treatment with allicin increased the activity of antioxidants, which scavenge the free radicals generated during diabetes.

Increased oxidative stress is supported by raised cytokine levels (Esposito, *et al*. 2002). High amounts of circulating inflammatory cytokines, such as TNF-α and IL-6, contribute significantly to insulin resistance in muscle and adipose tissue (Hotamisligil, *et al*. 1995) of diabetic animals. Tumor necrosis factor-alpha (TNF-α) plays a key role in the pathogenesis diabetes (Argile´s, *et al*. 1994). TNF-α inhibits insulin action in animal and in vitro models (Bruun, *et al*. 2003). Human cross-sectional studies have shown that elevated circulating TNF-α level are associated with insulin resistance and diabetes mellitus (Mishima, *et al*. 2001; Fernandez-Real, *et al*. 2002).

Many mechanisms may account for the metabolic effect of TNF-α, including the down regulation of genes related to normal insulin action and direct effects on glucose transport, and pancreatic β-cells (Grimble, 2002). IL-6 was among the first cytokines implicated as a predictor or pathogenetic marker of insulin resistance and cardiovascular disease (Ferna´ndez-Real and Ricart, 2003). Therefore, these mediators have detrimental effects on glucose metabolism. In the present study, TNF-α and IL-6 were observed to be significantly increased in diabetic rats and reverted back to normal after allicin and glibenclamide therapy. Allicin treatment reduced the serum TNF-α and IL-6 levels of diabetic rats, suggesting anti-inflammatory activity of the bark extract. Furthermore, the allicin was found to be as ameliorative potential of the reference drug glibenclamide.

The results obtained in this study clearly indicate that oral administered allicin to diabetic rats showed significant antihyperglycemic, anti-lipidperoxidative effects and this effect may be attributed at least in part to its anti-inflammatory properties as evident by attenuating TNF-α and IL-6 levels in diabetic rats. We thus conclude that allicin have potent ameliorative potential in STZ-induced diabetic rats.

ACKNOWLEDGEMENT

I wish to express my sincere thanks and gratitude to Prof. (Dr). Hussain Gadelkarim Ahmed for his valuable suggestions and also thankful to the University of Hail, Kingdom of Saudi Arabia for providing necessary research facilities.

REFERENCES

Argile´s JM, Lo´pez-Soriano J, and Lo´pez-Soriano FJ. (1994). Cytokines and diabetes: the final step?

Involvement of TNF-a in both type I and II diabetes mellitus. Hormone and Metabolic Research, 26: 447–9.

Beutler E and Kelly BM. (1963). The effect of sodium nitrite on red cell GSH. Experientia, 19 (2): 96-97.

Brodsky IG. (1998). Nutritional effects of dietary protein restriction in insulin-dependent diabetes mellitus. The Journal of Nutrition, 128: 337S-339S.

Bruun JM, Verdich C, Toubro S, Astrup A and Richelsen B. (2003). Association between measures of insulin sensitivity and circulating levels of interleukin-8, interleukin-6 and tumor necrosis factor-α. Effect of weight loss in obese men. European Journal of Endocrinology, 148: 535–542.

Ceriello A. (2000). Oxidative stress and glycemic regulation. *Metabolism* 49: 27– 29.

Chehade JM and Mooradian AD. (2000). A rational approach to drug therapy of type 2 diabetes mellitus. *Drugs,* 60: 95–113.

De Angelis K, Schaan BD, Maeda CY, Dall'Ago P, Wichi RB and Irigoyen MC. (2002). Cardiovascular control in experimental diabetes. *Brazilian Journal of Medical and Biological Research* 35(9): 1091-1100.

Dominiczak MH. (2003). Obesity, glucose intolerance and diabetes and their links to cardiovascular disease. Implications for laboratory medicine. *Clinical Chemistry of Laboratory Medicine* 41: 1266–1278.

Elkayam A, Mirelman D, Peleg E, Wilchek M, Miron T, Rabinkov A, *et al*. (2001). The effects of allicin and enalapril in fructose-induced hyperinsulinemic hyperlipidemic hypertensive rats. *Am J Hypertens* 14:377–381.

Esposito K, Nappo F, Marfella R, Giugliano G, Giugliano F, Ciotola M, *et al*. (2002). Inflammatory cytokine concentrations are acutely increased by hyperglycemia in humans. Role of oxidative stress. *Circulation* 106: 2067–2072.

Fernandez-Real JM and Ricart W. (2003). Insulin resistance and chronic cardiovascular inflammatory syndrome. Endocrine Reviews, 24: 278–301.

Fernandez-Real JM, Lainez B, Vendrell J, Rigla M, Castro A, Penarroja G, *et al*. (2002). Shedding of TNF-α receptors, blood pressure, and insulin sensitivity in type 2 diabetes mellitus. American Journal of Physiology-Endocrinology and Metabolism, 282: E952–959.

Gonen A, Harats D, Rabinkov A, Miron T, Mirelman D, Wilchek M, *et al*. (2005). The antiatherogenic effect of allicin: possible mode of action. *Pathobiology* 72:325–34.

Grimble RF. (2002). Inflammatory status and insulin resistance. *Current Opinion in Clinical Nutrition and Metabolic Care* 5(5): 551–9.

Halliwell B and Gutteridge JM. (1994). Lipid peroxidation, oxygen radicals, cell damage and antioxidant therapy. *The Lancet* 344: 1396-7.

Hirsch K, Danilenko M, Giat J, Miron T, Rabinkov A, Wilchek M, *et al*. (2000). Effect of purified allicin, the major ingredient of freshly crushed garlic, on cancer cell proliferation. *Nutr Cancer*. 38:245–54.

Hotamisligil GS, Arner P, Caro JF, Atkinson RL and Spiegelman BM. (1995). Increased adipose tissue expression of tumour necrosis factor-alpha in human obesity and insulin resistance. The Journal of Clinical Investigation, 95 (5): 2409-2415.

Jin L, Xue HY, Jin LJ, Li SY and Xu YP. (2008). Antioxidant and pancreas protective effect of aucubin on rats with streptozotocin-induced diabetes. European Journal of Pharmacology, 582: 162-7.

Kakkar P, (1984). Das B and Viswanathan PN. A modified spectrophotometric assay of superoxide dismutase. *Indian Journal of Biochemistry and Biophysics* 21: 130-132.

Ketterer B, Tipping E, Beale D, Meuwissen JATP. Ligandin, glutathione and carcinogen binding. In: Arias IM, (1976). Jakoby WB (eds.). *Glutathione: Metabolism and Function*, Raven Press, New York. 243–253

Koch HP and Lawson LD. (1996). Garlic: the Science and therapeutic application of *Allium sativum* L. and related species. 2nd ed. Baltimore: Williams & Wilkins;

Lang A, Lahav M, Sakhnini E, Barshack I, Fidder HH, Avidan B, *et al*. (2004). Allicin inhibits spontaneous and TNF-alpha induced secretion of proinflammatory cytokines and chemokines from intestinal epithelial cells. *Clin Nutr*. 23:1199–208.

Leila Z, Eliandra DS, Luisa HC, Anildo CJ, Moacir GP, Bruno S, *et al*. (2007). Effect of crude extract and fractions from *Vitex megapotamica* leaves on hyperglycemia in alloxan-diabetic rats. *Journal of Ethnopharmacology* 109: 151–155.

Loven D, Schedl H, Wilson H, Daabees TT, Stegink LD, Diekus M, *et al*. (1986). Effect of insulin and oral glutathione on glutathione levels and superoxide dismutase activities in organs of rats with streptozocin-induced diabetes. Diabetes, 35:503-507.

Lowry OH, Rosebrogh NJ, Farr AL and Randall RJ. Protein measurement with the Folin phenol reagent. The *Journal of Biological Chemistry* 1951; 193: 265-275.

McLennan S, Heffermen V, and Wright S. (1991). Change in hepatic glutathione metabolism in diabetes. *Diabetes* 40: 344-8.

Mishima Y, Kuyama A, Tada A, Takahashi K, Ishioka T and Kibata M. (2001). Relationship between serum tumor necrosis factor-α and insulin resistance in obese men with Type 2 diabetes mellitus. *Diabetes Research and Clinical Practice* 52: 119–23.

Niehoff AG, Van Haeften TW, Onland-Moret NCh, Elbers CC, Wijmenga C and Van Der Schouw YT. (2007). C-reactive protein is independently associated with glucose but not with insulin resistance in healthy men. *Diabetes Care* 30: 1627–1629.

Pepato MT, Migliorini RH, Goldberg AL and Kettelhut IC. (1996). Role of different proteolytic pathways in degradation of muscle protein from streptozotocin-induced rats. American Journal of Physiology-Endocrinology and Metabolism 271: E340-E347.

Piédrola G, Novo E, (2001). Escobar F and García-Robles R. White blood cell count and insulin resistance in patients with coronary artery disease. *Annales d Endocrinologie* 62: 7–10.

Rahimi R, Nikfar S, Larijan B, Abdollahi M. (2005). A review on the role of antioxidant in the management of diabetes and its complications. Biomedicine and Pharmacotherapy, 59: 365-73.

Rathod N, Raghuveer I, Chitme HR and Chandra R. (2008). Antidiabetic activity of Nyctanthes arbortristis. Pharmacognosy Magazine, 16: 335-340.

Rodriguez T, Alvarez B, Busquets S, Carbo N, Lopez-Soriano FJ and Argiles JM. (1997). The increased skeletal muscle protein turnover of the streptozotocin diabetic rat is associated with high concentrations of branched-chain amino acids. *Biochemical and Molecular Medicine* 61: 87-94.

Sasaki T. and Matsy, S. (1972). Effect of acetic acid concentration on the color reaction in the *O*-toluidine boric acid method for blood glucose estimation. *Japanese Journal of Clinical Chemistry* 1: 346-53.

Sasikumar D, Mathi S, Abdel-Tawab Salama SM, Manoharan S and Prema S. (2010). *Terminalia arjuna* (Roxb.) modulates circulatory antioxidants on 7,12-dimethylbenz(a)anthracene-induced hamster buccal pouch carcinogenesis. *Oman medical journal* 25(4): 276-281.

Saxena AK, Srivastava P, Kale RK and Baquer NZ. (1993). Impaired antioxidant status in diabetic rat liver. Effect of vanadate. *Biochemical Pharmacology* 45(3): 539–542.

Sinha AK. (1972). Colorimetric assay of catalase. *Analytical Biochemistry* 47(2): 389- 394.

Tan AC, Konczak I, Sze DM and Ramzan I. (2011). Molecular pathways for

cancer chemoprevention by dietary phytochemicals. *Nutr Cancer* 63: 495–505.

Tiwari AK and Rao JM. (2002). Diabetes mellitus and multiple therapeutic approaches of phytochemicals: Present status and future prospects. *Current Science* 83: 30–38.

Ugochukwu NH and Cobourne MK. (2003). Modification of renal oxidative stress and lipid peroxidation in streptozotocin-induced diabetic rats treated with extracts from *Gongronema latifolium* leaves. *International Journal of Clinical Chemistry and Diagnostic Laboratory Medicine* 336: 73–81.

Weidmann P, Boehlen LM and de Courten M. (1993). Pathogenesis and treatment of hypertension associated with diabetes mellitus. *American Heart Journal* 125: 1498–1513.

WHO. Diabetes: What is diabetes? World Health Organization 2009. Available from http://www.sho.int/mediacentre/factsheets/fs312/en/intex.html [accessed 3 Mar 2010].

World Health Organization. Expert (1980). committee diabetes mellitus. *Technical Report Second Series* 646: Geneva: 61.

Xiao ZP, Peng ZY, Peng MJ, Yan WB, Ouyang YZ, Zhu HL. (2011). Flavonoids health benefits and their molecular mechanism. *Mini Rev Med Chem* 11: 169–77.

Yagi K. (1987). Lipid peroxides and human diseases. *Chemistry and Physics of Lipids*, 45(2-4): 337-351.

ARABIC SUMMARY

تقييم الخصائص الإصلاحية لمادة الأليسين (ثنائي الأليل ثيو سلفونيت) ضد داء السكري المستحث تجريبيا في الفئران

ساسي مومار دونا راسو

قسم الكيمياء الحيوية، كلية الطب، جامعة حائل، المملكة العربية السعودية.

خلفية : من المعروف أن مرض السكري يزيد من الجهد التأكسدي مما ينجم عنه الكثير من المضاعفات المرضية. وتبحث هذه الدراسة الحالية في الإمكانية الوقائية لمادة الأليسين ضد مرض السكري في نموزج الفئران.

المنهجية: ستحقن الحيوانات بجرعة واحدة من مادة "بالستربتوزوتوسين" (٥٥ ملجم\كجم) لإحداث داء السكري. ثم تعالج فئران مجموعة التحكم والفئران المريضة بمادة الأليسين لمدة ٣٠ يوما. وقد تم استخدام مادة غليبي نكلاميد (١٠جم\كجم) كمجموعة تحكم موجبة بعد النظام التجريبي ثم جمعت عينات الدم من الفئران بعد ذبحها لفصل المصل عن الدم. واستخدمت كل من قياسات سكر الدم ووزن الجسم والبروتينات الكلية ودلائل الإجهاد التأكسدي ودلائل الإلتهابات للرصد الحيوي لمعرفة الإمكانية الوقائية لمادة الأليسين.

النتائج: لوحظ في حيوانات التجربة فقدا معنويا لأوزانها وارتفاعا في سكر الدم مع انخفاض في البروتينات الكلية (وذلك عند درجة معنوية ٠.٠٥%) مقارنة بمجموعة التحكم. بيد أن هذه الإعتلالات تم تصحيحها بعد العلاج بمادة الأليسين لتستعيد محتوي TBARS والإنخفاض في القلوبيولينات الكلية ونشاطي إنزيمي CAT و SOD بصورة طبيعية في حيوانات التجربة. كما انخفضت التغيرات في دلائل الإلتهابات لمستوياتها الطبيعية. ولم يكن هناك تغيرا في الفئران المعطاة مادة الأليسبن بمفرده.

الخاتمة: بذا نستنتج أن للأليسين إمكانية علاجية كمضاد للإرتفاع في سكر الدم ومضاد لأكسدة الدهون ومضاد للإلتهابات في الحيوانات المصابة بداء السكري.

كلمات مفتاحية: أليسين، الجهد التأكسدي، دلائل الإلتهابات، عامل نخر الأورام، بيروكسيد الدهون.