

AQUEOUS EXTRACT OF *FICUS BENGALENSIS* BARK DECREASES OXIDATIVE ACTIVITY IN STREPTOZOTOCIN-INDUCED DIABETIC RATS

*JALARI RAMU

*Dr. JALARI RAMU, Associate Professor of Biochemistry, School of Medicine, Wolaita Sodo University Teaching and Referral Hospital, Ethiopia.

CORRESPONDING AUTHOR : *DR. JALARI RAMU

*Email ID: ramujalari@gmail.com

Mobile No: +91-9492633090 (India)

+251-947331518 (Ethiopia)

Article History

Received: 22.09.2021

Revised: 14.10.2021

Accepted: 28.11.2021

ABSTRACT

One of the major etiologies in pathogenesis of diabetes especially complications is oxidative stress. In this present investigation the aqueous extracts of *Ficus benghalensis* bark antioxidant properties had been studied in albino rats with streptozotocin-induced diabetes. Oral administration of *Ficus benghalensis* bark extracts for 6 weeks result as reduction of Thiobarbituric acid reactive substances (TBARS) and hydroperoxides. Aqueous extract of *Ficus benghalensis* bark at a dose of 150 mg/kg BW significantly increased in the reduced glutathione (GHS), superoxide dismutase (SOD), catalase (CAT) in liver of streptozotocin (STZ) -induced diabetic rats. These results clearly indicated that the antioxidant effect of aqueous extract and a dose at 500 µg/kg BW of glibenclamide reduced the oxidative stress in diabetic rats. Medicinal plants are not only a mean of health care, but make an important contribution to poverty eradication to livelihoods of poor communities all over the world. World trade in medicinal plants accounts for about 30 percent of the total drug market. Interest in medicinal plants business has been rapidly growing field and has commercial and socio-economic significance.

Keywords: Aqueous extract, *Ficus benghalensis* bark, Oxidative stress, Poverty eradication, Streptozotocin-induced diabetes.

INTRODUCTION:

Diabetes mellitus (DM) refers to a common metabolic disorder that leads hyperglycemia. Diabetes mellitus caused by a complex interaction of genetics and environmental factors. Diabetes mellitus is associated with significantly increased oxidative stress (Mc Coll *et al.*, 1997). Under this oxidative stress likely to affect the mechanical behavior of the membrane are those which involve the changes in lipid composition and peroxidation of endogenous membrane phospholipids reticulocyte and erythrocyte membranes are labile to lipidperoxidation owing to their content of polyunsaturated lipids and to the fact that they are directly exposed to molecular oxygen and involve in the generation of free radical intermediates and semi-stable peroxides (Tappel, 1973). Diabetics have

been shown to have increased levels of free radical activity and are more exposed to oxidative stress. These free radicals can react with polyunsaturated fatty acids and leads to the formation of peroxidation of membrane (Baynes, 1991). The Lipid peroxidation process is controlled by various cellular defense mechanisms of enzymatic and non-enzymatic scavenger systems (Halliwell and Gutteridge, 1994; Simmons, 1984). Thus, these defense mechanisms are controlled or altered in diabetes (Wohaieb and Godin, 1987). The locally available plants like *Ficus benghalensis* have the capability to cure diseases traditionally since long years, due to these reasons this plant was selected for the study. Some of studies clearly indicated that bark of *Ficus benghalensis* have a significant role to use as a natural antioxidant. Therefore, there is a need to the study the antioxidant properties of plant extract on diabetes mellitus. *Ficus benghalensis* bark was chosen to prepare the aqueous extract with water. An earlier study on this plant has been made with bark only. Hence in this investigation an attempt has been made to study the effect of *Ficus benghalensis* bark extract on tissue lipid peroxides and enzymatic antioxidants in rats with STZ induced diabetes. Medicinal plants are widely used in the health-care system all over the world (Jeyaprakash *et al.*, 2011). Interest in medicinal plants has been rapidly on the increase in the South Asian region due to growing awareness of their commercial and socio-economic significance.

MATERIALS & METHODS:

Male albino rats (Wistar strain) weighing 150-200gm were used in this present investigation. They were obtained from Ethiopian health and nutrition research institute (EHNRI), Addis Ababa and housed under standard husbandry conditions ($27^{\circ}\text{C} \pm 2^{\circ}\text{C}$, 50–65 % relative humidity and 12h:12h day-night cycle) and allowed standard fed with normal laboratory pellet diet and water. The experiment was conducted according to the ethical norms approved by Animal Ethics Committee Guidelines of the EHNRI. The bark of *Ficus benghalensis* were initially separated from the main plants body and rinsed with distilled water, and dried under shade paper towel in laboratory and then homogenized into fine particles and stored in air tight bottles and were used for all the extraction process. The *Ficus benghalensis* bark powdered juice was obtained with a Turmix electric extractor with 500ml of sterile distilled water. The juice was filtered and the residue was removed. The extract was concentrated under vacuum to get solid yield and freeze dried and the yield was calculated. The solid extract was stored $0-4^{\circ}\text{C}$ and used for further study.

Diabetes was induced experimentally in albino rats by freshly prepared solution of streptozotocin (Sigma Chemical Co. St. Louis, USA) (40 mg/kg) in 0.1mol/L cold citrate buffer, pH 4.5, was injected intraperitoneally (Siddique *et al.*, 1987). After 48 h the blood glucose levels were determined by using Autoanalyzer (Microlab 2000), rats with moderate diabetes having glycosuria and hyperglycemia (i.e., blood glucose of 250-300 mg/dL) had taken for the experiment. The experimental animals were divided into four groups. Each group has 5 rats which are group 1- normal rats; group 2- diabetic control (STZ treated); group 3 - diabetic rats were treated with aqueous extract of *Ficus benghalensis* bark 150 mg/kg body weight by oral administered with an intra gastric tube (Roman-Ramos *et al.*, 1995); and group 4 - diabetic rats were treated with glibenclamide daily for 42 days (Pari and Uma Maheswari, 2000). After 6 weeks, the albino rats were sacrificed at the end of the experimental period of 42 days and blood was collected for the estimation of glucose. The liver and kidneys were dissected out, washed in ice-cold saline, patted dry and weighed.

Fasting blood glucose was measured by the O-toluidine method (Sasaki *et al.*, 1972). Hydroperoxide was determined by Jiang *et al.*, (1992). Protein content in tissue homogenate was measured by the method of Lowry *et al.*, (1951). Lipid peroxidation was measured by measuring the levels of MDA by the method of Nichans *et al.*, 1968. Reduced glutathione (GSH) (Ellman, 1959), Superoxide dismutase (SOD) (Madesh and Balasubramanian, 1998), Catalase (CAT) (Bergmayer, 1983), Thiobarbituric acid reactive substances (TBARS), (Fraga *et al.*, 1988), Glutathione peroxidases (GP_x) (Rotruck *et al.*, 1973) and glutathione-S-transferase (GST) (Habig *et al.*, 1974) were analyzed in the normal, diabetic induced and drug treated rats. The study was carried out following approved from the Faculty of Medicine Institutional Review Board on the use and care of experimental animals.

RESULTS:

Effect of oral administration of aqueous extract of *Ficus benghalensis* bark and glibenclamide on blood glucose in normal and STZ-induced diabetic rats showed in Table 1. There were significantly decreased blood glucose levels in treated diabetic rats compared to untreated diabetic rats. The results reveal that *Ficus benghalensis* bark extract was more effective than glibenclamide. Table 2 and 3 presents the concentration of TBARS and hydroperoxides including lipid peroxidation in liver and kidney tissues of normal and STZ-induced diabetic rats. There was a significant elevation in liver and kidney tissues TBARS, hydroperoxides and lipid peroxidation during diabetes compared to the corresponding control group. The oral administration of aqueous extract of *Ficus benghalensis* bark and glibenclamide significantly decreased the levels of TBARS, hydroperoxides and lipid peroxidation in liver and kidney of rats with streptozotocin-induced diabetes.

Table-1: Effect of aqueous extract of *Ficus benghalensis* bark on blood glucose in normal and STZ-induced diabetic rats.

Group	Blood glucose (mg/dL)
Normal	80.10 ± 2.20
Diabetic control	274.20 ± 13.50
Diabetic + <i>Ficus benghalensis</i> (250 mg/kg)	93.47 ± 4.12
Diabetic + Glibenclamide (500 µg/kg)	99.85 ± 7.25

Table-2: Effect of aqueous extract of *Ficus benghalensis* bark on concentration of TBARS and hydroperoxides in liver and kidney of normal and STZ-induced diabetic rats.

Group	TBARS (mmol/L per 100 g tissue)		Hydroperoxides (mmol/L per 100 g tissue)	
	Liver	Kidney	Liver	Kidney
Normal	0.792 ± 0.06	1.350 ± 0.11	65.20 ± 2.15	51.63 ± 2.43
Diabetic control	1.620 ± 0.16	2.062 ± 0.13	92.85 ± 3.75	70.45 ± 4.20
Diabetic + <i>Ficus benghalensis</i> (250 mg/kg)	1.150 ± 0.07	1.420 ± 0.04	78.59 ± 3.64	62.38 ± 3.91
Diabetic + Glibenclamide (500 µg/kg)	1.261 ± 0.08	1.630 ± 0.06	83.41 ± 4.25	67.50 ± 2.40

Table-3: Effect of aqueous extract of *Ficus benghalensis* bark on concentration of lipidperoxides in liver and kidney of normal and STZ-induced diabetic rats.

Group	Tissue MDA (mmol/L per 100 g tissue)	
	Liver	Kidney
Normal	292.58 ± 1.85	288.31 ± 1.15
Diabetic control	358.29 ± 1.28	356.45 ± 1.73
Diabetic + <i>Ficus benghalensis</i> (250 mg/kg)	305.92 ± 1.56	273.48 ± 1.55
Diabetic + Glibenclamide (500 µg/kg)	335.75 ± 1.25	297.86 ± 1.35

Table-4: Effect of aqueous extract of *Ficus benghalensis* bark on the level of reduced glutathione in liver and kidney of normal and STZ-induced diabetic rats.

Group	Reduced Glutathione (mg/100 g tissue)	
	Liver	Kidney
Normal	46.80 ± 3.78	32.18 ± 1.61
Diabetic control	21.40 ± 2.91	20.68 ± 1.26
Diabetic + <i>Ficus benghalensis</i> (250 mg/kg)	41.20 ± 1.87	27.90 ± 1.45
Diabetic + Glibenclamide (500 µg/kg)	36.12 ± 2.65	25.20 ± 1.25

Table-5: Effect of aqueous extract of *Ficus benghalensis* bark on activities of SOD, Catalase of liver and kidney of normal and STZ-induced diabetic rats.

Group	SOD (U/mg protein)		CAT (U/mg protein)	
	Liver	Kidney	Liver	Kidney
Normal	9.00 ± 0.25	13.52 ± 0.78	81.25 ± 5.65	40.00 ± 2.25
Diabetic control	3.50 ± 0.23	8.40 ± 0.35	40.20 ± 3.45	22.28 ± 1.15
Diabetic + <i>Ficus benghalensis</i> (250 mg/kg)	5.89 ± 0.35	10.75 ± 0.65	65.47 ± 3.39	34.62 ± 1.58
Diabetic + Glibenclamide (500 µg/kg)	4.95 ± 0.11	9.80 ± 0.72	60.50 ± 2.79	29.45 ± 1.16

Table-6: Effect of aqueous extract of *Ficus benghalensis* bark on activities of GP_x and GST in liver and kidney of normal and STZ-induced diabetic rats.

Group	GP _x (U/mg protein)		GST (U/mg protein)	
	Liver	Kidney	Liver	Kidney
Normal	8.35 ± 0.65	6.02 ± 0.56	6.25 ± 0.41	5.90 ± 0.56
Diabetic control	5.28 ± 0.40	4.15 ± 0.35	2.95 ± 0.16	2.25 ± 0.38
Diabetic + <i>Ficus benghalensis</i> (250 mg/kg)	6.90 ± 0.50	6.05 ± 0.15	4.85 ± 0.43	3.89 ± 0.49
Diabetic + Glibenclamide (500 µg/kg)	6.25 ± 0.22	5.15 ± 0.20	3.90 ± 0.35	3.46 ± 0.24

The levels of liver and kidney reduced glutathione (GSH) were significantly decreased to near normal levels during diabetes compared to the corresponding control groups showed in Table 4. The oral administration of aqueous extract of *Ficus benghalensis* bark and glibenclamide increased the content of GSH in the liver and kidney of diabetic rats. The results showed that the *Ficus benghalensis* bark extract was more effective than glibenclamide.

Effect of oral administration of aqueous extract of *Ficus benghalensis* bark and glibenclamide on the activities of SOD, CAT, GP_x, and GST were presented in Table 5 and 6. During diabetes there was a significant reduction in the activities of SOD, catalase, GP_x and GST in tissues such as liver and kidney. Administration of aqueous extract of *Ficus benghalensis* bark and glibenclamide increased the activity of SOD, catalase, GP_x and GST in STZ-induced diabetic rats. The results reveal that the *Ficus benghalensis* bark extract was more prominent compared with glibenclamide.

DISCUSSION:

The present investigation which shows beneficial antioxidant effect with aqueous extract of *Ficus benghalensis* bark in STZ-induced diabetic rats. The several workers reported that STZ-induced diabetes and insulin deficiency leads to increased blood glucose levels (Chaude *et al*, 2001). STZ at lower doses 50 mg - 60 mg/kg BW produces partial destruction of pancreatic β-cells with permanent of diabetic condition (Aybar *et al*, 2002). Since a low dose 60 mg/kg BW of STZ was chosen for this study. Administration of aqueous extract of *Ficus benghalensis* bark at a dose of 250 mg/kg BW decreased the elevated blood glucose level and may be due to the insulin secretagogue effect of the active compound, leucopelargonin (Cherian *et al*, 1992) presents in the extract and prolonged administration may stimulate the β - cells of islets of Langerhans to produce insulin. The

antihyperglycemic effect of aqueous extract of *Ficus benghalensis* bark was compared with glibenclamide, a standard hypoglycemic drug.

Free radicals are generated in diabetes and leads to induction of lipid peroxidation during diabetes reported by several workers (Kaleem *et al*, 2006; Mano *et al*, 2000). Oxygen free radical are formed by stimulating H_2O_2 *in-vitro*, as well as *in-vivo* and in the pancreatic β – cells of Langerhans during diabetes mellitus reported by Halliwall and Gutteridge, 1989. In this investigation, the increased malondialdehyde (MDA) and hydroperoxides in liver and kidney tissues of STZ-induced diabetic rats served as an index of elevated lipid peroxidation in diabetes mellitus. When the concentration of endogenous peroxides increase it may initiate uncontrolled lipid peroxidation leading to cellular infiltration and islet cell damage in type I diabetes (Metz, 1984). The increased susceptibility of the tissues of diabetic animals to lipid peroxidation may be due to the observed increased concentration of TBARS and hydroperoxides in the liver and kidney of diabetic rats (Stanely *et al*, 2001). An increase in lipid peroxide concentration in the liver and kidney of diabetic animals has been observed (Nakakimura and Mizuno, 1980). Administration of aqueous extract of *Ficus benghalensis* bark and glibenclamide significantly decreased the levels of lipid peroxidation index significantly. The reduction of lipid peroxidation can be attributed to the antioxidant activity of various phytochemicals present in the *Ficus benghalensis* bark aqueous extract. Further, these results suggested that the major function of the aqueous extract is to protect vital tissues such as liver, kidney from damage and thereby reducing the after effects of diabetes.

GSH is an important source of reducing equivalents during oxidative stress generated by reactive oxygen species and can participate in the elimination of reactive intermediates by reducing hydroperoxides in the presence of GP_x (Meister, 1984; Nicotera and Orrenius, 1986). The decrease in the GSH level represents increased utilization due to oxidative stress (Anuradha and Selvam, 1993). In the present research work, we have observed the decreased level of glutathione in STZ induced diabetic rats. The GSH depletion in liver and kidney is considered the most important sensitizing mechanism in the pathogenesis of liver and kidney injury. Administration of aqueous extract of *Ficus benghalensis* bark and glibenclamide increased the content of GSH in the liver and kidney of diabetic rats.

Superoxide dismutase is the major attractive metalloprotein in the antioxidant family. The increased synthesis of superoxide dismutase against superoxide anion radical (O_2^-) production is an adaptive response of the cell to synthesis increased SOD through the stimulation of gene transcription (Das *et al.*, 1997). SOD is a defense enzyme that catalyses the dismutation of superoxide radicals (McCord and Fridovich 1969). The defensive antioxidant enzyme next to SOD is Catalase. Catalase is a hemoprotein that catalyses the reduction of hydrogen peroxides into water and oxygen and protects the tissues from highly reactive hydroxyl radicals (Chance *et al*, 1952). The activity of catalase was found to be decreased in diabetic rats. The inhibition of catalase activity during STZ induced diabetes may be due to the increased generation of reactive free radicals, which can create an oxidative stress in the cells. Therefore, the observed reduction in the activity of these enzymes (SOD, catalase) may result in a number of deleterious effects due to the accumulation of superoxide anion radicals and hydrogen peroxide. Administration of aqueous extract of *Ficus benghalensis* bark and glibenclamide increased the activities of SOD and Catalase in diabetic rats. The activities of GP_x and GST are observed to decrease significantly in diabetic rats. GP_x is a selenium dependent enzyme has high potency in scavenging reactive free radicals. In the present experiments,

the levels of glutathione peroxidase activity in liver and kidney was elevated during diabetes to compensate the free radical scavenging effect utilized by the GSH as the substrate (Rajashree *et al.*, 1998). GST catalyse the reduction of hydrogen peroxide and hydroperoxides to non-toxic products (Bruce *et al.*, 1982). The depletion in the activity of these enzymes may result in deleterious oxidative changes due to accumulation of toxic products. In this context, other workers also reported a decrease in the activities of these antioxidant enzymes (SOD, Catalase, GP_x and GST) in the liver and kidney of diabetic rats (Stanely *et al.*, 2001; Anuradha and Selvam, 1993). As the alterations produced in the antioxidant activities indicate the involvement of deleterious oxidative changes, increased activities of the components of this defense system would therefore be important in protection against radical damage. Administration of *Ficus benghalensis* aqueous fruit extract and glibenclamide increased the activities of GP_x and GST in the liver of diabetic rats. The over expression of these antioxidant enzymes in diabetic rats treated with aqueous extract of *Ficus benghalensis* bark implies that this potential oxidant defense is reactivated by the active principles of *Ficus benghalensis*. This results in an increase in the capacity of detoxification through enhanced scavenging of oxy radicals.

CONCLUSIONS

The present investigation shows that there were significant variations in the observed biochemical parameters. The administration of aqueous extract of *Ficus benghalensis* bark possesses an antioxidant activity that may contribute to its protective action on lipid peroxidation and to enhancing its effect on cellular antioxidant defense. Therefore, therapeutic administrations of aqueous extract of *Ficus benghalensis* bark greatly change the biochemical parameters in the STZ induced diabetic induced rats and maintained well to the normal level. These results clearly suggest that, the *Ficus benghalensis* have enormous antidiabetic potential value. Further, this study creates a hope on new drug discovery in diabetic controlling by using *Ficus benghalensis* as precursor. Moreover, it is very important to study the specific phytochemical compounds responsible for contributes to the protection against oxidative damage in streptozotocin induced diabetes.

REFERENCES:

- Anuradha, C.V. and Selvam, R. (1993): Effect of oral methionine on tissue lipid peroxidation and antioxidants in alloxan induced diabetic rats. *J. Nutr. Biochem.*, 4: 212.
- Aybar, M., Sanchez, Riera, A.N., Grau, A., and Sanchez, S.S. (2002): Hypoglycemic effect of the water extract of *Smilax glabra* (yacon) leaves in normal and in diabetic rats. *J. Ethnopharmacol.*, 74: 125 – 32.
- Baynes, J. W. (1991): Role of oxidative stress in development of complications in diabetes. *Diabetes.*, 40: 405.
- Bergmayer, H. U. (1983): UV method of catalase assay. In *Methods of Enzymatic Analysis*, Weheim Deer field Beach, Florida, Bansal., 3: 273.
- Bruce, A., Freeman, D. and James, C. (1982): Biology of disease – free radicals and tissue injury. *Lab. Invest.*, 47: 412.
- Chance, B., Greenstein, D.S. and Roughton, R.J.W. (1952): The mechanism of catalase action I – steady state analysis. *Arch. Biochem. Biophys.*, 37: 301.
- Chaude, M.A., Orisakwe, O.E., Afonne, O.J., Gamenial, K.S., Vongtau, O.H. and Ob, E. (2001): Hypoglycemic effect of the aqueous extract of *Boerhavia diffusa* leaves. *Ind. J. Pharmacol.*, 33: 215 – 16.

- Cherian, S., Vinod Kumar, R., August, K.T., and Kidwai, K.R. (1992): Antidiabetic effect of a glycoside of pelarginidin isolated from the bark of *Ficus bengalensis* Linn. *Ind. J. Biochem. BioPh.*, 29: 380 – 82.
- Ellman, G.L. (1959): Tissue sulphhydryl groups. *Arch. Biochem. Biophys.*, 82 : 70.
- Fraga, C. G., Leibovitz, B. E., Toppel, A. L. (1988): Lipid peroxidation measured as TBARS in tissue slices. Characterization and comparison with homogenates and microsomes. *Free Radic. Biol. Med.*, 4: 155.
- FRLHT. (2002). Vidya Venkatesh (Ed.). Medicinal plants scenario in India. Unpublished report of the Foundation for Revitalization of Local Health Traditions (FRLHT), Bangalore, India.
- Habig, W. H., Pabst, M. S. and Jekpoly, W. B. (1974): Glutathione transferase; a first enzymatic step in mercapturic acid formation. *J. Biol. Chem.*, 249: 7130.
- Halliwall, B. and Gutteridge, J.M.C. (1989): Free radicals in biology and medicine 2nd ed. *Clarendon Press., Oxford*.
- Halliwell, B. and Gutteridge, J. M. C. (1994): Lipid peroxidation, oxygen radicals, cell damage and antioxidant therapy. *Lancet.*, 1:1396.
- Jeyaprakash, K., Ayyanar, M., Geetha, K.N. and Sekar, T. (2011): Traditional uses of medicinal plants among the tribal people in Theni District (Western Ghats), Southern India. *Asian Pac J Tropic Biomed.* S-20-S25.
- Jiang, Z. Y., Hunt, J. V. and Wolff, S. (1992): Ferrous sulphate oxidation in the presence of xylenol orange for detection of lipid hydroperoxide in low-density lipoprotein. *Anal. Biochem.*, 202: 384 – 389.
- Kaleem, M., Asif, M., Ahmed., and Bano, B. (2006): Antidiabetic and antioxidant activity of *Annona squamosa* extract in streptozotocin – induced diabetic rats. *Singapore Med. J.*, 47: 670 – 75.
- Lange, D. (2004). Medicinal and aromatic plants: trade, production, and management of botanical resources. *Acta Horticulturae.* 629: 177-197.
- Lowry, O. H., Rosebrough, N. J., Farr, A. L. and Randall, R. J. (1951): Protein measurement with the Folin phenol reagent. *J. Biol. Chem.*, 193: 265-275.
- Madhesh, M., and Balasubramanian, K. A. (1998): Microtitre plate assay for superoxide dismutase using MTT reduction by superoxide. *Indian Journal of Biochemistry and Biophysics.*, 35: 184-188.
- Mc Coll, A. J., Kong, C., Nimmo, L., Collins, J., Eikeles, R. and Richmond, W. (1997): Total antioxidant status, Protein glycolation, lipid hydroperoxides in non-insulin dependent diabetes mellitus. *Biochem. Soc. Trans.*, 25: 132.
- McCord, J. M. and Fridovich, I. (1969): Superoxide dismutase. An enzymic function for erythrocyte hemocuprein. *J. Biol Chem.* 244:6049–6055.
- Mano, T., Shinohara, R., Nagasaka, A., Nagasaka, H., Uchimura, K., and Hayashi, R. (2000): Scavenging effect of nicrandil on free radicals and lipid peroxide in streptozotocin – induced diabetic rats. *Metabol.*, 49: 427 – 31.
- Meister, A. (1984): New aspects of glutathione biochemistry and transport selective alterations of glutathione metabolism. *Nutr. Rev.*, 42: 397.
- Metz, S.A. (1984): Oxygenation products of arachidonic acid: Third messenger for insulin release. *Prostaglandins.*, 27: 147.
- Nakakimura, H. and Mizuno, K. (1980): Studies on lipid peroxidation in biological systems II. Hyperlipoperoxidemia in mice induced by alloxan. *Chem. Pharmacol. Bull.*, 28: 2207.
- Nichans, W. G., Samuelsson, D. (1968): Formation of malondialdehyde from phospholipid arachidonate during microsomal lipid peroxidation. *Eur J Biochem.*, 6: 126 – 30.
- Nicotera, P. and Orrenius, S. (1986): Role of thiols in protection against biological reactive intermediates. *Adv. Exp. Med. Biol.*, 197: 41.
- Pari, L., Uma Maheswari, J. (2000): Antihyperglycemic activity of *Musa sapientum* flowers: Effect on lipid peroxidation in alloxan diabetic rats. *J. Ethnopharmacol.*, 14: 136.
- Roman-Ramos, R., Flores Sanoz, J. L., Alarcon Aquilar, F. J. (1995): Anti hyperglycemic effect of some edible plants. *J.*

Ethnopharmacol., 48:25.

Rotruck, J. T., Pope, A. L., Ganther, H. E., and Swanson, A. B. (1973): Selenium: Biochemical role as a component of glutathione peroxidases. *Science.*, 179: 588.

Saganuwan, A.S. (2010). Some medicinal plants of Arabian Peninsula. *J. Med. Plants Res.* 4(9): 766-788.

Sasaki, T., Matsy, S., Sonae, A. (1972): Effect of acetic acid concentration on the colour reaction on the o-toluidine boric acid method for blood glucose. *Rinsho Kagaku.*, 1: 346.

Shinwari, Z.K. (2010). Medicinal Plants Research in Pakistan. *Journ. Med. Pl. Res.* 4(3): 161-176.

Siddique, O., Sun, Y., Lin, J. C., Chien, Y. W. (1987): Facilitated transdermal transport of insulin. *J. Pharm Sci.*, 76: 341.

Simmons, K. J. (1984): Defense against free radicals has therapeutic implications. *JAMA.*, 251: 2187.

Stanely, P., Prince, M. and Menon, V.P. (2001): Antioxidant action of *Tinospora cardifolia* root extract in alloxan diabetic rats. *Phytother Res.*, 15: 213.

Wohaieb, S.A. and Godin, D.V. (1987): Alterations in free radical tissue defense mechanism in streptozotocin-induced diabetes in rat Effects of insulin treatment. *Diabetes.*, 36: 1014.

Das, S., Bandyopadhyay, D., Bhattacharjee, M & Banerjee, R.K. (1997). Hydroxyl radical is the major curative factor in stress induced gastric ulceration. *Free. Rad. Biol. Med.*, 23: 8.

Rajashree, C.R., Rajmohan, T and Augusti, K.T. (1998). Antiperoxidative effect of garlic in alcohol fed rats. *Indian J. Exp. Biol.*, 36: 60 – 64.

Tappel, A. L. (1973): Lipid peroxidation damage to cell components. *Federation proceedings.*, 32 (8): 1870-1874.