Academic Sciences International Journal of Pharmacy and Pharmaceutical Sciences

ISSN- 0975-1491 Vol 4, Issue 2, 2012

Research Article

BENEFICIAL EFFECT OF *CASSIA FISTULA (L)* FLOWER EXTRACT ON ANTIOXIDANT DEFENSE IN STREPTOZOTOCIN INDUCED DIABETIC RATS

JEYANTHI. K. A.

Professor and Head, Department of Biochemistry, Thanthai Hans Roever College, Perambalur, Tamilnadu, India.
Email: jeyanthi.ka@gmail.com

Received: 12 Nov 2011, Revised and Accepted: 3 Dec 2012

ABSTRACT

Diabetes mellitus is a metabolic disorder of multiple etiologies characterized by chronic hyperglycemia with disturbances in carbohydrate, fat and protein metabolism resulting from defects in insulin secretion, insulin action or both. The present study shows that increased oxidative stress is apparent in STZ induced diabetic animals. The ethanolic extract of *cassia fistula* flowers exhibits a significant antihyperglycemic as well as antioxidant as antioxidant activity in experimental rats. The presences of phytochemicals further strengthen the efficacy of cassia fistula flowers in protecting the tissue defense system against oxidative damage in STZ induced diabetes. The observed antioxidant potential of cassia fistula flowers may partially responsible for its antidiabetogenic properties.

Keywords: Diabetes mellitus, *Cassia fistula*, STZ, Antioxidant.

INTRODUCTION

Diabetes mellitus has been a medical problem for more than 2000 years. The effect of diabetes mellitus includes long term damage, dysfunction and failure of various organs. The total number of people under diabetes mellitus is projected to rise from 171 million in 2000 to 366 million in 2030[1]. Increased oxidative stress is a widely accepted. Insulin deficiency has many other metabolic consequences in carbohydrate, fat and protein metabolism. Cells must maintain a proper balance between the levels of free radicals, such as reactive oxygen species (ROS) and endogenous antioxidants to ensure the structural integrity cellular components. If there is an imbalance in favor of free radicals, sensitive biological structures such as DNA, lipids and proteins could be damaged. Such damage may play a role in the etiology of several degenerative diseases such as cancer, diabetes and arthritis [2, 3]. Free radicals are continuously produced in the body as a result of normal metabolic processes and interaction with environmental stimuli. Normal cells have a number of endogenous antioxidants, which eliminate toxic oxymetabolites under normal[4].

MATERIALS ANMD METHODS

Animals

Male albino rats of the wistar strain weighing around 160-180gm were purchased from the Tamilnadu vertinary and Animal Sciences University, Chennai. They were acclimatized to animal house condition, fed with commercial pellet, and had free access to water. The experiments were designed and conducted in accordance with the ethical norms approved by ministry of social justices and empowerment, government of India.

Plant material and preparation of plant extract

Fresh, mature *cassia fistula* flower were collected from a tree in Perambalur area, Tamlinadu, India. The flowers were first washed well with distilled water to remove the traces of contaminants. The flowers were dried at room temperature. One hundred grams of powdered was extracted with *petroleum ether* (60-80^{0}C) to remove lipids. It was then filtered; the residue was extracted with 95% *ethanol* by soxhlation. The *ethanol* was evaporated in a rotary evaporator at 40-50^{0}C under reduced pressure. All the extracts were dissolved in water and use.

Experimental induction of diabetes

The animals were fasted overnight and diabetes was induced by a single intraperitoneal injection of a freshly prepared solution of *streptozotocin* (55mg/kg body weight) in 0.1 M cold *citrate buffer* (pH 4.5).The animals were allowed to drink 5% *glucose solution* overnight. Control rats were injected with *citrate buffer* alone.

Experimental design

The rats were divided into four groups comprising of six animals in each group as follows:

Group1: Control rats receiving 0.1 M cold *citrate buffer.*

Group2: Diabetic control rats.

Group3: Diabetic rats treated with *Cassia fistula* flower extract (100mg/kg body weight/day) given orally for 30 days.

Group4: Diabetic rats treated with *glibenclamide* (600µg/kg body weight/day) given orally for 30 days.

At the end of the experimental period, the rats were anaesthetized and sacrificed by cervical dislocation. Blood was collected with anticoagulant and used for the preparation of plasma. Blood collected without anticoagulant was used for serum separation. The biochemical parameters such as Glucose, Insulin, Protein, Glycosylated hemoglobin, Ceruloplasmin, Vitamin E, Vitamin C, Reduced glutathione, Glutathione peroxidase, Super oxide dismutase and Catalase were analyzed in the serum sample.

RESULTS AND DISCUSSION

Table 1: List of phytochemicals in ethanolic extract of *cassia fistula*

Phytochemicals	Inference
Flavanoids	+
Alkaloids	+
Glycosides	+
Saponins	+
Phytosterols	+
Steroids	+
Triterpenoids	+
Tannins	+
Carbohydrates	+
Proteins	+
Amino acids	+
Anthroquinones	_

+ - Presence ; _ Absence

Table-1 shows the qualitative analysis of phytochemicals in the ethanolic extract of cassia fistula flowers. From preliminary phytochemical screening, it was found that the extract showed a positive response for the presence of flavanoids, alkaloids, glycosides, saponins, phytosterols, steroids, tannins, proteins and triterpenoids. Saponins possess hypercholesterolemic, antidiabetic,

anti tumor, antiviral, antioxidative and hepatoprotective properties[5]. Flavanoids extent the activity of Vitamin C act as antioxidants that protects LDL cholesterol from oxidation, inhibit plat let aggregation and act as anti inflammatory agents [6, 7] . Plant alkaloids have the tendency to release insulin from pancreatic ß cells and also have the potential to protect it from alloxan induced pancreatic damage in experimental animals[8].

Table 2 shows the levels of blood glucose, plasma insulin, glycosylated hemoglobin in control and experimental rats. There is an increase in the levels of serum glucose and HbA1c in STZ-NIC treated rats confirmed the induction of diabetes mellitus[9]. There was a significant increase in blood glucose, glycosylated hemoglobin and concomitant decrease in plasma insulin in STZ induced rats were reverted back to near normal in diabetic rats treated with cassia fistula and glibenclamide treated rats. Antihyperglycemic effect of medicinal plant extracts is generally depending upon the degree of ß cell destruction[10]. Increased level of glycosylated hemoglobin was observed in diabetic rats and this increase is directly proportional to blood glucose level [11].

Table 2: Effect of *cassia fistula* flower extract on blood glucose, plasma insulin, glycosylated hemoglobin levels in control and experimental rats

Group	Blood glucose (mg %)	Plasma insulin(µU ml^{-1})	Glycosylated hemoglobin(% Hb)
Control	89.38 + 6.16	15.64+ 0.82	5.9 + 0.33
Diabetic control	297.31+ 20.81[a]	5.21 + 0.26[a]	12.8 + 0.72[a]
Diabetic + C*assia fistula*	93.61 + 7.27[b]	13.63 + 0.79[b]	7.1 + 0.45[b]
Diabetic + glibenclamide	103.75 + 8.05[c]	12.43 + 0.70[c]	7.6 + 0.38[c]

Table 3: Effect of *cassia fistula* flower extract on Plasma TBARS, Vitamin C, Vitamin E, Ceruloplasmin levels in control and experimental rats.

Group	Plasma TBARS(nmol/ml)	Vitamin C (mg %)	Vitamin E (mg %)	Ceruloplasmin (mg %)
Control	3.09 + 0.14	1.63 + 0.07	1.94 + 0.08	15.27+ 0.68
Diabetic control	7.16 + 0.28[a]	0.98 + 0.03[a]	3.38 + 0.15[a]	25.35 + 1.29[a]
Diabetic + C*assia fistula*	3.64 + 0.134[b]	2.98 + 0.13[b]	2.98 + 0.13[b]	18.39 + 0.82[b]
Diabetic + glibenclamide	3.95 + 0.16[c]	2.84 + 0.12[c]	2.84 + 0.12[c]	19.04 + 0.90[c]

Table 3 depicts the levels of Plasma TBARS, Vitamin C, Vitamin E, Ceruloplasmin levels in control and experimental rats. There was a significant increase in the levels of TBARS. The oral administration of the extract and glibenclamide tended to bring these values back to the normal. The significant decrease in the levels of Vitamin C and a concomitant increase in Vitamin E, Ceruloplasmin in STZ induced diabetic rats. Treatment with *cassia fistula* flower extract reversed these levels to near normal. Increased lipid peroxidation impairs membrane function by decreasing membrane fluidity and changing the activity of membrane bound enzymes and receptors. The products of lipid peroxidation are harmful to most tissues in the body and are associated with a variety of diseases, such as atherosclerosis and brain damage [12].

The decreased level of Vitamin C may be due to either increased utilization as an antioxidant defense system against reactive oxygen species or to a decrease in glutathione level, since glutathione is required for the recycling of ascorbic acid [13,14]. Vitamin E is a lipophilic antioxidant and inhibits lipid peroxidaton[15]. The observed elevation in plasma ceruloplasmin in diabetic rats may be due to elevated lipid peroxides [16].

Table 4: Effect of *cassia fistula* flower extract on Plasma GSH, SOD, CAT, GPx levels in control and experimental rats.

Group	Plasma GSH (mg %)	SOD (mg %)	CAT (mg %)	GPx (mg %)
Control	26.38 + 1.37	3.86 + 0.16	14.98 + 0.79	6.08 + 0.27
Diabetic control	18.63 + 0.91[a]	2.08 + 0.07[a]	7.35 + 0.27[a]	3.02 + 0.11[a]
Diabetic + C*assia fistula*	24.16 + 1.30[b]	3.69 + 0.15[b]	12.94 + 0.54[b]	5.86+ 0.22[b]
Diabetic + glibenclamide	22.79 + 1.21[c]	3.51 + 0.14[c]	12.18 + 0.52[c]	5.74 + 0.21[c]

Values are given as mean ± S.D for groups of six animals each. Values are statistically significant at $P < 0.05$.

a - Diabetic control Vs control

b – Diabetic + Cassia fistula Vs diabetic control

Table 4 depicts the level of Plasma GSH, SOD, CAT, and GPx in control and experimental rats. A marked decrease in the level of reduced glutathione, SOD, CAT and GPx was observed in diabetic rats. The oral administration of *cassia fistula* extract glibenclamide to diabetic rats restored the activities of all the enzymes. Reduced glutathione normally plays the role of an intracellular radical scavenger and is the substrate of many xenobiotic elimination reactions[17]. Decreased levels of reduced glutathione are reported in the plasma and pancreas of the STZ induced diabetic condition [18].Our result is consistent with the results of [19,20,21] who reported a decrease in antioxidant enzymes such as SOD, CAT and glutathione peroxidase in diabetes mellitus.

CONCLUSION

Our results indicate that the preventive effects of *cassia fistula* flowers may be due to inhibition of lipid peroxidation and scavenging of free radicals by its antioxidant nature.

REFERENCES

1. Wild S, Roglic G, Green A, Sicree R and King H. Global prevalence of diabetes. Diabetes Care 27(5): 1047-1053 (2004).
2. Halliwell. B. Free radicals and antioxidants: a personal review. Nutr rev. 52: 253-265 (1994).
3. Collins. A.R. Oxidative DNA damage, antioxidants and cancer. Bioassays 21: 238-246 (1999).
4. Reilly P.M, Schiller H.J and Bulkley G.B. Pharmacogenic approach to tissue injury mediated by free radicals and other reactive oxygen metabolites. Am.J.Surg. 161(4): 488-503 (1991).
5. Rupasinghe H.P, Jackson .C.J, Poysa V, Di Berado C, Bewley J.D and Jenkinson J. Soyasapogenol. A and B distribution in soyabean in relation to seed physiology, genetic variability and growing location. J.Agri. Food Chem. 51(20): 5888-5894 (2003).

6. Manach C, Regreat F and Texier O. (1996). Bioavailability, metabolism and physiological impact of 4.oxo-flavanoids. Nutr. Res. 16: 517-524.
7. Cook. N.C. and Samman.S. Flavanoids chemistry, metabolism, cardio protective effects and dietary sources. J.Biol Chem. 255(10): 4758-4762 (1996).
8. Ignachimuthu S and Amalraj T. Effect of leaf extract of *Zizyphus jujube* on diabetic rats. Indian. J. Pharmacol 30: 107 (1998).
9. V.chitra,Venkata.K.R.CH.H.Varma.P, M.V.R.Krishna Raju, K. Jeya Prakash. Study of antidiabetic and free radical scavenging activity of the seed extract of strychnos nuxvomica. International journal of pharmacy and pharmaceutical sciences, vol 2, suppl 1: 106-110 (2010)
10. Grover J.K, Vats V and Rathi S.S. Antihyperglycemic effect of *Eugenia jambolana* and *Tinospora cordifolia* in experimental diabetes and their effects on key metabolic enzymes involved in carbohydrate metabolism. J. Ethnopharmacol. 73: 461-470 (2000).
11. Al-Yassin. D, Ibrahim K.A. Minor hemoglobin fraction and the level of fasting blood glucose J.Fac.Med.Univ. Baghdad. 23: 373-380 (1981).
12. Acworth I.N., Mc Cabe D.R and Maher Y. The analysis of free radicals, their reaction products, and antioxidants. In: S.I.Baskin, H.Salem (Eds). Oxidants antioxidants and free radicals, Taylor and Francis, Washington, DC. (Chapter 2) (1997).
13. Sajithlal G.B, Chitra P, Chandrasekaran G. Effect of curcumin on the advanced glycation and cross-linking of collagen in diabetic rats Biochem. Pharmacol. 56: 1606-1614 (1998).
14. Jin .X.L, Shao .Y, Wang M.J, Chen L.J and Jin.G.Z. Tetrahydro protoberberines inhibit lipid peroxidation and scavenge hydroxyl free radical. Acta Pharmacol. Sin 21: 477-480 (2000).
15. Stahl.W, Sies H. Antioxidant defense vitamin E and C and carotenoids. Diabetes 46:14-18 (1997).
16. Ravi. K, Ramachandran B and Subramanian S. Effect of *Eugenia jambolana* seed kernel on antioxidant defense system in STZ induced diabetes in rats. Life Sci.75: 2717-2731 (2004).
17. Gregus Z, Fekete T, Halaszi E, Klassen C.D. Lipoic acid impairs glycine conjugation of benzoic acid and renal excretion of benzoylglycine. Drug Metab. 24: 682-688 (1996).
18. Sathishsekar .D, Subramanian S. Beneficial effect of *Momordica charantia* seeds in the treatment of STZ induced diabetes in experimental rats. Biol. Pharm. Bull. 28: 978-983 (2000).
19. Wolff S.P. Diabetes mellitus and free radicals. Free radicals, transition metals and oxidative stress in the etiology of diabetes mellitus and complications. Br.Med. Bull. 49: 642-652 (1993).
20. El-Missiry, M.A, El-Guindy. A.M. Amelioration of alloxan induced diabetes mellitus and oxidative stress in rats by oil of Erica sativa seeds. Ann. Nutr. Metab. 44:97-100 (2000).
21. Venkidesh R., Subhash C. Mandal, Dilipkumar Pal, Mohana Lakshmi S, Saravanakumar A. Antidiabetic activity of smilax Chinensis (L.) in alloxan induced diabetic rats. Vol 2, Suppl 2 : 51-54 (2010)