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Antiperoxidative and antioxidant effect of ellagic acid on normal and streptozotocin induced diabetes in albino wistar rats

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ABSTRACT

The objective of the present study was to investigate the antioxidant effect of ellagic acid on normal and streptozotocin induced diabetes in albino wistar rats. Streptozotocin was used for the induction of diabetes in rats. The rats were divided into six groups: normal control rats, normal rats treated with ellagic acid (50mg/kg and 100mg/kg), diabetic control and diabetic rats treated with ellagic acid (50mg/kg and 100mg/kg). Diabetic rats were treated with ellagic acid for a period of 35 days. Streptozotocin induced diabetic rats showed significant elevation of thiobarbituric acid reactive substances (TBARS) and hydroperoxide (HP) in plasma and tissues and significant decrease in the levels of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GP$_X$), reduced glutathione (GSH), ascorbic acid (vitamin C) and tocopherol (vitamin E) in liver, kidney and pancreas. Treatment with ellagic acid counteracted the effect of lipid peroxidation in streptozotocin induced diabetes rats and also exhibited a significant increase in the antioxidants suggesting the potential antioxidant property of ellagic acid.
Key words: Ellagic acid, streptozotocin, diabetes, lipid peroxidation, antioxidants.

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INTRODUCTION

Despite the great efforts that have been made in the understanding and management of diabetes, the disease and disease related complications are increasing unabated [1]. Inspite of the presence of known antidiabetic medicine in the pharmaceutical market, remedies from medicinal plant compounds are used with success to treat this disease [2]. Diabetes mellitus is an endocrine disorder, characterized by a loss of glucose homeostasis resulting from defects in insulin secretion, insulin action or both resulting in impaired metabolism of glucose and other energy yield fuels such as lipid and protein [3].

Normal pancreatic β-cells can compensate for the insulin resistance by increasing insulin secretion; however, extensive exposure of pancreatic β-cells to high glucose levels causes β-cell dysfunction that is associated with impaired insulin secretion and biosynthesis. In living system, free radicals are generated as part of the body's normal metabolic process and the free radical chain reactions are usually produced in the mitochondrial respiratory chain, liver mixed function oxidases, through xanthine oxidase activity, drugs and xenobiotics [4].

According to World Health Organization projections, the diabetes population is likely to increase to 300 million or more by the year 2025 [5]. World Health Organization has suggested the evaluation of the potential of plants as effective therapeutic agents, especially in areas where we lack safe modern drugs [6]. In the ongoing search for more effective and safer drugs, attention is being paid to new and safe drugs [7].

Streptozotocin (STZ) was found to be selectively toxic to the beta cells of the pancreatic islets, the cells that normally regulate blood glucose levels by producing the hormone insulin. This suggested the drug's use as an animal model of diabetes [8-9]. STZ is synthesized by *Streptomycetes achromogenes* and is used to induce both insulin dependent and non-insulin dependent diabetes mellitus [10]. STZ enters the pancreatic β-cell via glucose transporter-$GLUT_2$ and causes alkylation of deoxyribonucleic acid. STZ induces activation of polyadenosine diphosphate ribosylation and nitric oxide release. As a result of STZ action, pancreatic β-cells are destroyed by necrosis [11]. Oxidative stress is reported to be increased in patients with diabetes mellitus [12]. Accumulating evidence suggests that oxidative cellular injury caused by free radicals contributes to the development of diabetes mellitus [13]. Moreover, diabetes also induces changes in the tissue content and activity of the antioxidant enzymes [14].

Most of the plants have been found to contain substances like glycosides, alkaloids, flavonoids and terpenoids which are frequently implicated as having antidiabetic effects [15]. Flavonoids, a large class of phenolic compounds widely distributed in plants and vegetables, have been reported to be strong antioxidants and radical scavengers [16]. Flavonoids are found in a wide range of foods. For example, flavanones are in citrus, isoflavones in soya products, anthocyanidins in wine and bilberry and flavans in apples and tea [17]. Ellagic acid is a polyphenol antioxidant found in numerous fruits and vegetables including blackberries, raspberries, strawberries, cranberries, walnuts, pecans, pomegranates, wolfberry and other

plant foods. The antiproliferative and antioxidant properties of ellagic acid have spurred preliminary research into the potential health benefits of ellagic acid consumption [18].

Plants produce ellagic acid and convert it to a form of tannin known as ellagitannins. These are glucosides which are readily hydrolyzed by water to regenerate ellagic acid when the plants are eaten. Ellagic acid has antiproliferative and antioxidant properties in a number of *in vitro* and small-animal models [19]. The antiproliferative properties of ellagic acid are due to its ability to directly inhibit the DNA binding of certain carcinogens, including nitrosamines [20-21]. As with other polyphenol antioxidants, ellagic acid has a chemoprotective effect in cellular models by reducing oxidative stress [22].

The aim of the present study was to evaluate the antioxidant effect of ellagic acid in streptozotocin induced diabetes in albino wistar rats.

MATERIALS AND METHODS

Experimental Animals

Female albino wistar rats (150-200 g) obtained from the Venkateswara Enterprises, Bangalore were used in this study. They were housed in polypropylene cages (47cm x 34cm x 20cm) lined with husk. It was renewed every 24 hours under a 12:12 hour light: dark cycle at around 22ºC and had free access to water and food. The rats were fed on a standard pellet diet (Pranav Agro Industries Limited., Maharashtra, India). The pellet diet consisted of 22.02% crude protein, 4.25% crude oil, 3.02% crude fiber, 7.5% ash, 1.38% sand silica, 0.8% calcium, 0.6% phosphorus, 2.46% glucose, 1.8% vitamins and 56.17% nitrogen free extract (carbohydrates). The diet provided metabolizable energy of 3600 kcal. The experiment was carried out in accordance with the guidelines of the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), New Delhi, India.

Drug and Chemicals

Streptozotocin (STZ) was purchased from Himedia Laboratories Private Limited, Mumbai. Ellagic acid was purchased from Sigma- Aldrich, St. Louis, USA. All other chemicals used in the study were of analytical grade.

Induction of Experimental Diabetes

Streptozotocin was used to induce diabetes mellitus in normoglycemic female albino wistar rats. A freshly prepared solution of STZ (45mg/kg body weight) in 0.1M citrate buffer, pH 4.5 was injected intraperitoneally in a volume of 1ml/kg body weight to overnight fasted rats. After 48 hours of STZ administration, rats with moderate diabetes having glycosuria and hyperglycemia were taken for the experiment [23].

Experimental Design

A total of 36 rats were used in the present investigation. The animals were randomly divided into 6 groups of 6 rats in each group.

Group 1: Normal control rats
Group 2: Normal rats + Ellagic acid (50 mg/kg)
Group 3: Normal rats + Ellagic acid (100 mg/kg)
Group 4: Diabetic control rats
Group 5: Diabetic + Ellagic acid (50 mg/kg)
Group 6: Diabetic + Ellagic acid (100 mg/kg)

Ellagic acid was dissolved in 0.2% dimethyl sulfoxide and administrated to rats orally using an intragastric tube daily for a period of 35 days.

Sample Collection

After 35 days of treatment, the animals were fasted for 12 hours, anaesthetized with pentobarbital sodium (35 mg/kg) and sacrificed by cervical decapitation. The blood was collected in tubes containing mixture of potassium oxalate and sodium fluoride as anticoagulant for the biochemical analysis. Liver, kidney and pancreas were dissected out, washed in ice-cold saline, and patted dry and weighed. 10% tissue homogenate prepared from liver, kidney and pancreas was used for various biochemical estimations.

Biochemical Estimations

Biochemical parameter such as plasma TBARS was estimated by the method of Yagi [24] and the concentration of TBARS in the tissues was estimated by the method of Nichans and Samuelson [25]. The levels of HP were estimated by the method of Jiang *et al* [26]. Superoxide dismutase was assayed by the method of Kakkar *et al* [27]. The activity of catalase was determined by the method of Sinha [28]. Glutathione peroxidase (GPx) was estimated by the method of Rotruck *et al* [29]. The level of GSH was estimated by the method of Ellman [30]. Vitamin C was estimated by the method of Omaye *et al* [31] and vitamin E by the method of Baker *et al* [32]. The level of ceruloplasmin in serum was estimated by the method of Ravin [33].

Statistical Analysis

Statistical analysis was performed by one-way analysis of variance (ANOVA) followed by Duncan's Multiple Range Test (DMRT) using Statistical Package from the Social Sciences (SPSS) software package version 9.05. Results were expressed as mean ± S.D for six rats in each group. *P*-values < 0.05 were considered as significant.

RESULTS

Thiobarbituric acid reactive substances (TBARS) and hydroperoxide (HP)

Table: 1 Effect of ellagic acid on the levels of thiobarbituric acid reactive substances (TBARS) in plasma and tissues of normal and streptozotocin induced diabetic rats.

Groups	Plasma TBARS (nmol/ml)	TBARS (mM/g tissue)		
		Liver	Kidney	Pancreas
Normal control	0.151 ± 0.03 [a]	1.748 ± 0.27 [a]	1.333 ± 0.07 [a]	44.27 ± 3.46 [a]
Normal + Ellagic acid (50mg/kg)	0.170 ± 0.03 [a]	1.736 ± 0.22 [a]	1.398 ± 0.09 [a]	46.33 ± 4.08 [a]
Normal + Ellagic acid (100mg/kg)	0.125 ± 0.04 [a]	1.736 ± 0.22 [a]	1.358 ± 0.05 [a]	44.08 ± 4.14 [a]
Diabetic control	0.418 ± 0.01 [b]	3.743 ± 0.42 [b]	3.515 ± 0.42 [b]	70.09 ± 5.26 [b]
Diabetic + Ellagic acid (50mg/kg)	0.286 ± 0.04 [c]	2.758 ± 0.26 [c]	2.858 ± 0.28 [c]	54.16 ± 4.86 [c]
Diabetic + Ellagic acid (100mg/kg)	0.250 ± 0.03 [d]	2.181 ± 0.21 [d]	2.115 ± 0.19 [d]	50.67 ± 3.43 [d]

Each value is mean $\pm$ S.D. for six rats in each group.
Values not sharing a common superscript (a-d) differ significantly with each other (P<0.05, DMRT).

Table: 2 Effect of ellagic acid on the levels of lipid hydroperoxide (HP) in plasma and tissues of normal and streptozotocin induced diabetic rats.

Groups	Plasma Hydroperoxides (mM/dl)	Lipid hydroperoxide (mM/g tissue)		
		Liver	Kidney	Pancreas
Normal control	9.54 ± 0.97 [a]	75.58 ± 4.49 [a]	58.15 ± 2.28 [a]	12.22 ± 0.83 [a]
Normal + Ellagic acid (50mg/kg)	8.73 ± 0.89 [a]	74.23 ± 3.25 [a]	57.36 ± 1.35 [a]	12.39 ± 1.1 [a]
Normal + Ellagic acid (100mg/kg)	9.25 ± 0.73 [a]	75.08 ± 2.79 [a]	59.17 ± 2.23 [a]	12.74 ± 0.81 [a]
Diabetic control	25.78 ± 1.08 [b]	97.16 ± 4.32 [b]	78.23 ± 2.38 [b]	19.03 ± 0.97 [b]
Diabetic + Ellagic acid (50mg/kg)	22.61 ± 0.76 [c]	85.83 ± 1.35 [c]	69.69 ± 3.24 [c]	15.99 ± 1.6 [c]
Diabetic + Ellagic acid (100mg/kg)	18.25 ± 2.0 [d]	83.34 ± 4.83 [d]	64.30 ± 3.07 [d]	14.21 ± 0.67 [d]

Each value is mean $\pm$ S.D. for six rats in each group.
Values not sharing a common superscript (a-d) differ significantly with each other (P<0.05, DMRT).

The effect of ellagic acid on the levels of thiobarbituric acid reactive substances (TBARS) and hydroperoxide (HP) in plasma and tissues (liver, kidney and pancreas) of normal and diabetic rats are presented in Table 1 and 2 respectively. STZ-induced diabetic control rats showed significant increase in the levels of TBARS and HP in plasma and tissues as compared to normal rats. Oral administration of ellagic acid (50 and 100 mg/kg) in STZ-induced diabetic rats significantly decreased the levels of TBARS and HP in plasma and tissues.

Enzymic antioxidants

Table: 3 Effect of ellagic acid on the activities of superoxide dismutase (SOD) and catalase in liver, kidney and pancreas of normal and streptozotocin induced diabetic rats.

Groups	SOD (Units[a] / mg protein)			Catalase (Units[b] / mg protein)		
	Liver	Kidney	Pancreas	Liver	Kidney	Pancreas
Normal control	9.81 ± 0.49 [a]	14.85 ± 0.82[a]	14.52 ± 1.62 [a]	75.08 ± 4.49 [a]	36.58 ± 2.94 [a]	17.33 ± 1.45[a]
Normal + Ellagic acid (50mg/kg)	9.75 ± 0.78 [a]	14.41 ± 0.72[a]	14.56 ± 1.69 [a]	75.45 ± 2.79 [a]	36.84 ± 1.45 [a]	17.49 ± 1.7 [a]
Normal + Ellagic acid (100mg/kg)	9.63 ± 0.48 [a]	14.56 ± 0.17[a]	14.70 ± 1.55 [a]	74.98 ± 4.86[a]	37.11 ± 2.37[a]	17.29 ± 1.0[a]
Diabetic control	5.43 ± 1.27 [b]	6.84 ± 0.39 [b]	7.55 ± 0.61 [b]	40.93 ± 2.96[b]	18.73 ± 2.20 [b]	7.87 ± 0.28[b]
Diabetic + Ellagic acid (50mg/kg)	7.63 ± 1.32 [c]	11.62 ± 1.45[c]	9.37 ± 0.74 [c]	54.12 ± 3.31[c]	26.44 ± 2.54 [c]	9.81 ± 0.42 [c]
Diabetic + Ellagic acid (100mg/kg)	8.51 ± 1.12 [d]	12.64 ± 0.92[d]	12.01 ± 0.94 [d]	67.55 ± 3.38[d]	32.65 ± 2.77 [d]	12.81 ± 0.65 [d]

U[a] – Enzyme concentration required to inhibit the chromogen produced by 50% in one minute.

U[b] – µmol of hydrogen peroxide consumed per minute.

Each value is mean ± S.D. for six rats in each group.

Values not sharing a common superscript (a-d) differ significantly with each other (P<0.05, DMRT).

Table: 4 Effect of ellagic acid on the activity of glutathione peroxidase (GPx) in liver, kidney and pancreas of normal and streptozotocin induced diabetic rats.

Groups	GPx (Units[a] /min/mg protein)		
	Liver	Kidney	Pancreas
Normal control	7.47 ± 2.40 [a]	5.64 ± 1.82 [a]	32.50 ± 2.8 [a]
Normal + Ellagic acid (50mg/kg)	7.75 ± 0.92[a]	5.56 ± 0.73 [a]	32.45 ± 2.5 [a]
Normal + Ellagic acid (100mg/kg)	7.73 ± 0.8 [a]	5.58 ± 0.74[a]	32.68 ± 3.0[a]
Diabetic control	3.82 ± 0.20[b]	4.52 ± 0.35[b]	18.08 ± 1.32[b]
Diabetic + Ellagic acid (50mg/kg)	5.70 ± 1.84[c]	5.04 ± 1.49[c]	21.88 ± 2.01[c]
Diabetic + Ellagic acid (100mg/kg)	6.03 ± 0.25[d]	5.59 ± 0.80[d]	28.29 ± 2.65[d]

U[a] – µg of glutathione consumed.

Each value is mean ± S.D. for six rats in each group.

Values not sharing a common superscript (a-d) differ significantly with each other (P<0.05, DMRT).

Table 3 and 4 depicts the effect of ellagic acid on the activities of superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPx) in tissues (liver, kidney and pancreas) of normal and diabetic rats. STZ-induced diabetic control rats showed significant decrease in the activities of the antioxidant enzymes in tissues. Diabetic rats treated with ellagic acid (50 and 100 mg/kg) significantly increased the levels of antioxidant enzymes in tissues as compared to diabetic control rats.

Non-enzymic antioxidants

The effect of ellagic acid on the levels of vitamin C and vitamin E in plasma and tissues, serum ceruloplasmin and reduced glutathione (GSH) in tissues (liver, kidney and pancreas) in

normal and STZ-induced diabetic rats are presented in Table 5 and 6. STZ-induced diabetic rats showed significant decrease in the levels of all the non-enzymic antioxidants as compared to normal rats. Rats treated with ellagic acid (50 and 100 mg/kg) significantly increased the levels of these antioxidants in tissues and serum when compared with diabetic control rats.

Table: 5 Effect of ellagic acid on the levels of vitamin C and vitamin E in plasma and tissues of normal and streptozotocin induced diabetic rats.

Groups	Vitamin C			Vitamin E		
	Plasma (mg/dl)	Liver (µmoles/mg protein)	Kidney (µmoles/mg protein)	Plasma (mg/dl)	Liver (µmoles/mg protein)	Kidney (µmoles/mg protein)
Normal control	2.42 ± 0.54^a	0.67 ± 0.1^a	0.83 ± 0.19^a	8.66 ± 0.93^a	4.60 ± 0.5^a	4.09 ± 0.35^a
Normal + Ellagic acid (50mg/kg)	2.38 ± 0.21^a	0.67 ± 0.1^a	0.84 ± 0.13^a	8.66 ± 0.40^a	4.63 ± 0.58^a	4.11 ± 1.0^a
Normal + Ellagic acid (100mg/kg)	2.48 ± 0.56^a	0.69 ± 0.2^a	0.83 ± 0.20^a	9.08 ± 0.21^a	4.68 ± 0.65^a	4.0 ± 0.65^a
Diabetic control	0.80 ± 0.10^b	0.23 ± 0.05^b	0.31 ± 0.12^b	5.29 ± 0.29^b	1.35 ± 0.14^b	2.01 ± 0.12^b
Diabetic + Ellagic acid (50mg/kg)	1.32 ± 0.3^c	0.50 ± 0.12^c	0.46 ± 0.11^c	6.52 ± 0.27^c	2.80 ± 0.17^c	2.53 ± 0.15^c
Diabetic + Ellagic acid (100mg/kg)	1.92 ± 0.2^d	0.60 ± 0.1^d	0.65 ± 0.16^d	7.64 ± 0.5^d	3.60 ± 0.25^d	3.54 ± 0.37^d

Each value is mean $\pm$ S.D. for six rats in each group.
Values not sharing a common superscript (a-d) differ significantly with each other (P<0.05, DMRT).

Table: 6 Effect of ellagic acid on serum ceruloplasmin and reduced glutathione (GSH) in tissues of normal and streptozotocin induced diabetic rats.

Groups	Ceruloplasmin (mg/dl)	GSH (mM/g wet tissue)		
		Liver	Kidney	Pancreas
Normal control	17.38 ± 3.0^a	8.61 ± 1.22^a	8.57 ± 1.25^a	25.36 ± 1.5^a
Normal + Ellagic acid (50mg/kg)	17.94 ± 2.4^a	8.63 ± 1.12^a	8.37 ± 2.40^a	25.45 ± 1.61^a
Normal + Ellagic acid (100mg/kg)	17.54 ± 2.5^a	8.65 ± 0.77^a	8.44 ± 0.31^a	25.32 ± 1.46^a
Diabetic control	15.25 ± 2.8^b	4.67 ± 0.67^b	4.60 ± 0.67^b	15.78 ± 0.18^b
Diabetic + Ellagic acid (50mg/kg)	16.55 ± 2.4^c	5.60 ± 0.38^c	5.56 ± 0.38^c	20.01 ± 0.68^c
Diabetic + Ellagic acid (100mg/kg)	17.22 ± 1.9^d	6.47 ± 0.39^d	6.36 ± 1.03^d	21.91 ± 1.47^d

Each value is mean $\pm$ S.D. for six rats in each group.
Values not sharing a common superscript (a-d) differ significantly with each other (P<0.05, DMRT).

DISCUSSION

Lipid peroxidation is a free radical mediated process leading to oxidative deterioration of polyunsaturated lipids. Under normal physiological conditions, low concentrations of lipid peroxide are found in plasma and tissues. Oxygen derived free radicals generated in excess in response to various stimuli could be cytotoxic to several tissues. Most of the tissue damage is considered to be mediated by these free radicals by attacking membranes through peroxidation

of polyunsaturated fatty acids. The increase in oxygen free radicals in diabetes could be primarily due to increase in blood glucose levels, which upon auto-oxidation generate free radicals [34].

In the present study, the levels of TBARS and HP were significantly increased in plasma, liver, kidney and pancreas of STZ-induced diabetic rats which might be due to an increase in the generation of free radicals by STZ. Auto-oxidation of glucose and the increased susceptibility of the tissues of the diabetic animals may be due to the activation of the lipid peroxidation system and observed remarkable increase in the concentration of TBARS and HP [35].

A marked increase in the concentration of TBARS has been observed in STZ induced diabetic rats [36]. The increased susceptibility of the tissues of the diabetic animals may be due to the activation of the lipid peroxidation system. The possible source of oxidative stress in diabetes includes shifts in redox balance resulting from altered carbohydrate and lipid metabolism, increased generation of reactive oxygen species and decreased level of antioxidant defenses such as GSH and ascorbic acid.

Oral administration of ellagic acid in STZ-induced diabetic rats significantly decreased the levels of TBARS and HP in plasma and tissues of STZ-induced diabetic rats when compared with diabetic control rats. The decrease in the level of TBARS and HP observed in the treated animals may be due to the inactivation of the lipid peroxidation system by ellagic acid.

Several studies have reported elevations in specific oxidant stress markers in both experimental STZ and human diabetes mellitus, together with reduced total antioxidant defense and depletion in individual antioxidants [37]. The major antioxidant enzymes including SOD, GPx and CAT are regarded as the first line of the antioxidant defense system against reactive oxygen species generated *in vivo* during oxidative stress. SOD dismutates superoxide radicals to form hydrogen peroxide, which in turn is decomposed to water and oxygen by GPx and CAT, thereby preventing the formation of hydroxyl radicals [38, 39].

In diabetes, the oxidative stress damages the pancreatic tissue thereby further reducing insulin secretion. In the present study, the activities of SOD, CAT and GPx were significantly reduced in liver, kidney and pancreas of diabetes induced rats. Reports have shown that the activities of SOD, CAT and GPx were lowered in tissues of diabetic rats [40]. The observed decrease may be due to the utilization of non protein thiols by increased oxygen free radicals produced in hyperglycemia conditions.

Glutathione peroxidase and glutathione -S- transferase (GST) work together with glutathione in the decomposition of hydrogen peroxide to non-toxic products at the expense of the GSH [41]. Reduced activity of GPx may result from radical-induced inactivation as well as diabetes-induced glycation [42].

GSH plays an important role in the detoxification and metabolism as a cofactor or a substrate for enzymes GPx and GST. It is generally recognized that the detoxification function is impaired in diabetic animals. Further, insufficient availability of GSH may also reduce the activity of GPx. Decreased activities of GPx and GST in the liver and kidney have been observed during diabetes and this may result in a number of deleterious effects due to the accumulation of toxic products [35]. Glutathione (GSH) is a metabolic regulator and putative indicator of health. Lower level of plasma GSH observed in STZ induced diabetic rats in the present study is in accordance with those reported earlier. It appears that generation of oxygen radicals by increased levels of glucose causes increased utilization of GSH. [43].

GSH also functions as free radical scavenger and in the repair of free radical caused biological damage [44]. The important mechanism implicated in the diabetogenic action of STZ is by increased generation of oxygen free radicals, which causes a decrease in plasma GSH concentration, and plasma GSH/GSSG ratio [45]. The reduced level of blood GSH observed in diabetic rats could be due to the destruction of pancreatic ß cells by STZ reinforcing the view that STZ induces diabetes probably through the generation of oxygen free radicals [46].

Vitamin E is a lipophilic chain breaking antioxidant that scavenges lipid peroxyl radicals [47]. Antioxidants such as N-acetylcysteine and dietary antioxidants such as vitamin C and E have shown to be beneficial in protecting the beta cells from glucose toxicity in diabetes [48]. Vitamin E interrupts the chain reaction of lipid peroxidation by reacting with lipid peroxyl radicals, thus protecting the cell structures against damage [49]. The decreased level of vitamin E found in the diabetic rats as compared with control rats could be due to the increased oxidative stress, which accompanies the decrease in the level of antioxidants, and may be related to the causation of diabetes mellitus. In this context, Garg *et al* [43] also observed decreased level of plasma vitamin E in STZ induced diabetic rats.

Vitamin C is one of the most powerful natural antioxidants [50]. Vitamin C is an excellent water-soluble antioxidant that primarily scavenges oxygen radicals. Vitamin C has been reported to contribute to up to 24% of the total peroxyl radical-trapping antioxidant activity [51]. A decreased level of plasma vitamin C observed in the diabetic rats could be due to the increased utilization of vitamin C in deactivation of the increased levels of reactive oxygen species or to the decrease in the GSH level, since the GSH is required for the recycling of vitamin C [52].

The antioxidant defense system is significantly altered in diabetes. Ceruloplasmin is a copper containing oxidase, which serves to transport copper in tissues. Ceruloplasmin has been established as chain-breaking antioxidant with a potential to scavenge peroxyl radicals [53]. The level of ceruloplasmin was found to be significantly decreased in diabetic rats when compared to control rats.

Rats treated with ellagic acid significantly increased the levels of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx), reduced glutathione (GSH), vitamin C and

vitamin E in tissues and serum ceruloplasmin in STZ-induced diabetic rats suggesting the antioxidant property of ellagic acid.

In conclusion, our study demonstrates that ellagic cid exhibits antiperoxidative and antioxidant activities in STZ-induced diabetic rats by decreasing the levels if lipid peroxidation products and increasing the levels and activities of antioxidants. The possible mechanism of action of ellagic acid responsible for antioxidant effect needs further investigation.

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