

## Effect of Green and Degree of Roasted Arabic Coffee on Hyperlipidemia and Antioxidant Status in Diabetic Rats

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**Abstract:** This study aims to examine the effects of green and different roasted degree of Arabic coffee on alloxan induced diabetes in rats. Animals were allocated into five groups of six rats each: a normal control group, diabetic group, diabetic rats fed with green Arabic coffee, diabetic rats fed with light roasted coffee and diabetic rats fed with dark roasted coffee group. The results showed increasing roasting degrees led to a decrease in moisture, radical-scavenging activity and total phenols. The diabetic rats presented a significant increase in blood glucose, plasma lipid profile compared to the control group. In addition, plasma malonaldehyde levels significantly increased compared to normal control group. Antioxidant enzymes activities such as superoxide dismutase and reduced Glutathione (GSH) levels significantly decreased in the plasma of diabetic rats compared to normal controls. The results showed that the experimental rats supplemented by green and roasted Arabic coffee significant increased feed efficiency ratio than diabetic control group. At the end of the study period, the experimental rats were showed significant improvement in blood glucose. It is noted that green coffee bean group has the best effect in decreasing glucose level followed by light coffee group followed by dark coffee group which recorded 95.46, 119.17 and 201.46 mg/dL, respectively. Experimental rats supplemented by green and light roasted Arabic coffee were similar insulin concentration normal control group. All treated groups showed a significant decrease in TC, TL, TG and LDL-C, while a significant increase HDL compared with diabetic control group with the highest value for green coffee. The diabetic rat supplemented by dark coffee was lower effective against lipids profile than green and light coffee. Diet supplemented with green and roasted Arabic coffee in the diabetic rats ameliorated antioxidant enzymes activities and level of GSH in diabetic rats and significantly decreased MDA levels. The administration of Arabic coffee attenuated the increased levels of the plasma enzymes produced by the induction of diabetes and caused a subsequent recovery towards normalization comparable to the control group animals. Our results thus suggest that green and light roasted Arabic coffee supplemented may be helpful in preventing diabetic complications in adult rats.

**Keywords:** Antioxidant enzymes, arabic coffee, diabetes, lipid peroxidation, oxidative stress, roasted coffee

### INTRODUCTION

Coffee is among the most widely consumed pharmacologically active beverages in the world. Caffeine is the most widely consumed psychoactive substance. Coffee is rich in phenolic compounds with a strong antioxidant activity (Parliament, 2000). Phenolic compounds are secondary metabolites and generally involved in plant adaptation to environmental conditions (Vaast *et al.*, 2006). They are well recognised as potentially protective factors against human chronic degenerative diseases, such as cancer and cardiovascular disease (Nkondjock, 2009). Regular drinking of coffee can reduce the oxidation of human Low-Density Lipoprotein (LDL) and the oxidation of LDL, decreasing the risk of atherosclerosis (Delgado-Andrade and Morales, 2005). Roasting is an essential

step in coffee production for generating aroma, flavor and color of the coffee beans. The mode of heat transfer and the applied temperature profile are the most critical processing parameters that affect the physical and chemical properties of roasted coffee beans (Schenker *et al.*, 2002). The chemical reaction changes include Maillard reaction or nonenzymatic reaction, browning reaction and Strecker degradation of proteins, sugar, polysaccharides and other components. The degrees of roasting are controlled by roasting time and temperature and are necessary for the required chemical reactions without burning the beans and compromising the flavor of the beverage (Mendes, 2001). The degrees of roasting were qualitatively assessed from color and classified as a light, medium or dark roast (Clarke, 1985). However, over-roasted coffee could reduce antioxidant activity (Del Castillo *et al.*, 2002; Summa

*et al.*, 2007). Also Parliament (2000), found the major compositional changes occurring are the decrease of phenolic compounds and the formation of brown, water-soluble polymers called melanoidins, although decrease in protein, amino acids and other compounds is also described.

Diabetes Mellitus (DM) is a chronic metabolic disorder characterized by high levels of glucose in the blood due to the non-secretion of insulin or insulin insensitivity (American Diabetes Association (ADA), 2005). DM affects approximately 4% of the population worldwide and is expected to increase by 5.4% in 2025 (Kim *et al.*, 2006). Although the underlying mechanisms of diabetes complications remain unclear, clinical and preclinical evidence suggests that diabetes is associated with oxidative stress, leading to an increased production of Reactive Oxygen Species (ROS), including superoxide radical ( $O_2\bullet$ ), Hydrogen peroxide ( $H_2O_2$ ) and hydroxyl radical ( $OH\bullet$ ) or a reduction in the antioxidant defense system (Ihara *et al.*, 1999; Rahimi *et al.*, 2005; Rudge *et al.*, 2007). The oxidant/antioxidant imbalance in favor of oxidants contributes to the pathogenesis of different diabetic complications which are considered to result from enhanced reactive oxygen species generation via nicotinamide adenine dinucleotide phosphate-oxidase (Baynes and Thorpe, 1999; Garg *et al.*, 1996; Ha and Kim, 1999).

Coffee is two species of coffee trees of commercial importance, *Coffea arabica* and *Coffea robusta*. The two species differ in chemical composition of the green coffee bean. Van Dam and Feskens (2002) reported that moderate daily consumption of coffee helped to reduce the risk of type 2 diabetes, while Fredholm and Lindgren (1984) found that caffeine promotes lipolysis in rat adipocytes. Human studies show that caffeine enhances energy expenditure (Arciero *et al.*, 1995) and improves the clinical conditions of diabetic patients (De Matteis *et al.*, 2002). Another study by Greer *et al.* (2001) revealed that caffeine ingestion promotes glucose consumption with an increase in blood epinephrine, while pre-exercise consumption promotes ventilation and enhances lipolysis (Ryu *et al.*, 2001). Chlorogenic acid, another main constituent of coffee beans, has recently been reported to selectively inhibit hepatic glucose-6-phosphatase Arion *et al.* (1997) which is a rate-limiting enzyme involved in gluconeogenesis. However, roasting of coffee beans has been shown to reduce the content of chlorogenic acid in coffee (Del Castillo *et al.*, 2002). Green coffee beans are rich in chlorogenic acid and its related compounds that show hypotensive effect (Suzuki *et al.*, 2002). Thus, the purpose of the present study was to assess in vivo some nutritional properties derived from regular consumption of green and roasting coffee bean especially its potential effect on diabetid and on antioxidant status in animal models.

## MATERIALS AND METHODS

**Roasting:** Coffee beans bean (GCB) weighing 1200 g for each replication was sampled and then, the beans were divided into three subsamples of 400 g each. The roasting process was carried out in a roaster, which could roast 400 g of coffee beans for each batch. The light roasting degrees were set up at 270°C for 3 min (LCB), the dark roasting degrees were set up at 300°C for 3 min (DCB) and 400 g of unroasted coffee for each replication was used as control. The different beans were supplied by Kraft Foods (Munich, Germany).

**Experimental design:** Male Wistar rats (weighing 190-210 g) were obtained from the Research Institute of Ophthalmology, Medical Analysis Department, Giza, Egypt. Rats (n = 30 rats) were housed individually in wire cages in a room maintained at 25±2°C and kept under normal healthy condition. All rats were fed on standard diet for one week before starting the experiment for acclimatization.

**Induction of diabetes:** After 2 weeks of acclimatization, diabetes was induced in male rats with a freshly prepared solution of alloxan monohydrate in normal saline at a dose of 120 mg/kg Body Weight (BW) injected intraperitoneally (Mansour *et al.*, 2002; Sheweita *et al.*, 2002). Because alloxan is capable of producing fatal hypoglycemia as a result of massive pancreatic insulin release, rats were orally treated with 20% glucose solution (5-10 mL) after 6 h. The rats were then kept for the next 24 h on 5% glucose water solution to prevent hypoglycemia. Rats with moderate diabetes that exhibited glycosuria and hyperglycemia (i.e., blood glucose concentration 200-300 mg/dL) were taken for the experimental tests.

Group Rats fed on basal diet only as the control negative, group 2 Diabetic rats fed on basal diet as the control positive, group 3 Diabetic rats fed on 5% green coffee bean, group 4 Diabetic rats fed on 5% light green coffee bean and group 5 Diabetic rats fed 5% dark coffee bean.

### Biochemical assays:

**Glucose levels:** Plasma glucose levels were assayed by enzymatic methods, using commercial reagent kits.

**Estimation of plasma insulin concentration:** Plasma insulin level was determined using rat Insulin enzymelinkedimmunosorbent assay kit ref.

**Analysis of lipids in plasma:** Plasma lipid parameters such as Total Cholesterol (TC), Triglycerides (TG) and High-Density Lipoprotein-Cholesterol (HDL-C) levels were determined by enzymatic methods, using

commercial kits. The Low-Density Lipoprotein-Cholesterol (LDL-C) fraction and LDL-C = TC-(Triglycerides/5 + HDL-C).

**Measurement of malondialdehyde:** Concentrations of MDA an index of lipid peroxidation, was determined spectrophotometrically according to Draper and Hadley (1990). An amount of 0.5 mL of each plasma sample was mixed with 1 mL of trichloroacetic acid solution and centrifuged at 2500 g for 10 min. A 1-mL solution containing 0.67% Thiobarbituric Acid (TBA) and 0.5 mL of supernatant were incubated for 15 min at 90°C and cooled. Absorbance of TBA-MDA complex was determined at 532 nm using a spectrophotometer (Bekman-USA-Du 7400). Lipid peroxidation was expressed as nanomoles of TBA reactive substances using 1, 1, 3, 3-tetraethoxypropane as standard.

**Antioxidant enzymes and glutathione assays in plasma:**

**Total Superoxide Dismutase (SOD) activity:** SOD activity was estimated according to Beauchamp and Fridovich (1971). The reaction mixture contained 50 mM of plasma, 0.1 mM EDTA, 13 mM L-methionine, 2 µM riboflavin and 75 µ Mnitrobluetetrazolium (NBT). The developed blue color in the reaction was measured at 560 nm. Units of SOD activity were expressed as the amount of enzyme required to inhibit the reduction of NBT by 50% and the activity was expressed as units per milligrams of protein.

**Glutathione levels (GSH):** GSH was determined by the method of Ellman (1959) modified by Jollow *et al.* (1974) based on the development of a yellow color when DTNB (5, 5-dithiobis-2 nitro benzoic acid) was added to compounds containing sulfhydryl groups; The absorbance was measured at 412 nm after 10 min. Total GSH content was expressed as milligrams per milliliter in plasma.

**Statistical analysis:** The data were analyzed using the statistical package program Stat View 5 Software for Windows (SAS Institute, Berkley, CA, USA). Statistical analysis between groups was performed with one-way analysis of variance followed by Student t test. All data were expressed as means±S.D. The results were considered significant if p≤0.05.

**RESULTS AND DISCUSSION**

**Total phenolic acid and DPPH radical-scavenging activity:** The changes in the moisture content of Arabic coffee beans during roasting at 270°C for 3 min (light) and 300°C for 3 min (dark) as a function of roasting conditions are presented in Table 1. Temperature and time significantly (p<0.05) affected the moisture removal during roasting process. As it was expected,

Table 1: Effect of different roasting degree of Arabic coffee beans on moisture, total phenolic content and DPPH radical-scavenging activity (%)

Samples	Moisture	Total phenolic acid	DPPH (% inhibition)
Green coffee	11.45±1.41 <sup>a</sup>	43.59±3.12 <sup>a</sup>	87.26±4.46 <sup>a</sup>
Roasted light coffee	2.13±0.23 <sup>b</sup>	41.64±2.76 <sup>a</sup>	84.39±4.28 <sup>a</sup>
Roasted dark coffee	1.32±0.18 <sup>c</sup>	25.15±2.18 <sup>b</sup>	55.23±3.55 <sup>b</sup>

Means in the same column with different superscripts are significantly different (p≤0.05)

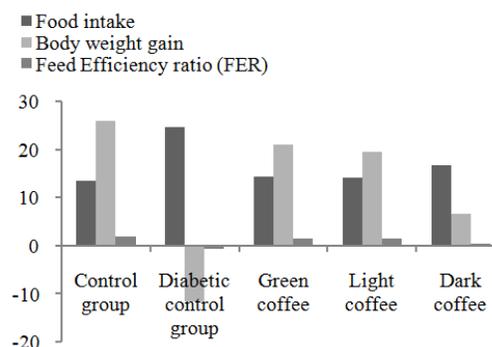


Fig. 1: Effect of supplemented by green, light and dark coffee bean on food intake, body weight and feed efficiency ratio

moisture loss occurred as roasting temperature increased, which varied from 11.45% in green beans to 2.13% in light roasted and from 11.45% in green beans to 1.32% in dark roasted, respectively. The Percentage of total phenolic acid and DPPH radical-scavenging activity for Arabic coffee bean samples after light roasted compared to green beans are indicated no significant difference. Meanwhile, the total phenolic content and DPPH radical-scavenging activity for Arabic coffee bean samples after dark roasted significantly decreased (p≤0.05) compared to green bean.

**Effect of Arabic coffee bean on body weight:**

Figure 1 shows the effect of supplemented by green, light and dark Arabic coffee bean on food intake; body weight and feed efficiency ratio were studied. Data indicated that the food intake of the diabetic control rats higher than the normal control and experimental rats fed on Arabic coffee bean. As shown in Fig. 1, body weight gain in the diabetic rats group was significantly lower than that in the control group (p<0.05). There were no significant differences between the green coffee group and light group (p>0.05). The results showed that the feed efficiency ratio in case of normal control rats was 1.91% while it was decreased to -0.67% when rats treated with alloxanas positive control group. On the other hand, no significant difference between the experimental rats supplemented by green and light coffee bean increased the feed efficiency ratio to 1.47 and 1.38%, respectively. Meanwhile, the feed efficiency ratio of experimental

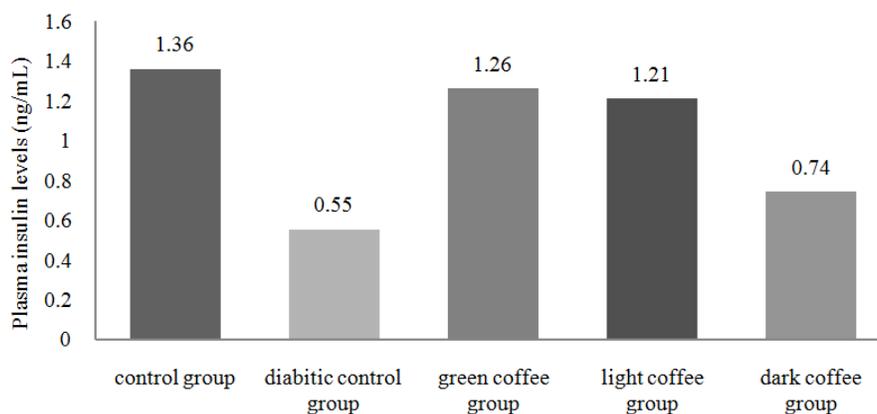


Fig. 2: Effect of different roasting degree of coffee beans on plasma insulin levels

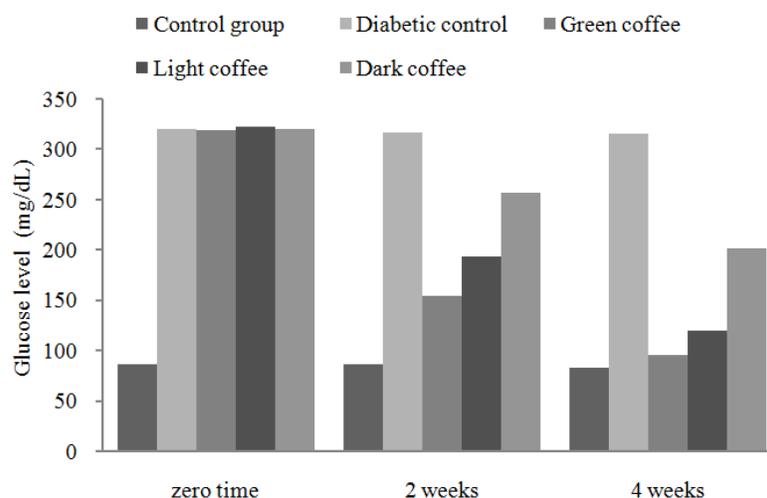


Fig. 3: Effect of different roasting degree of coffee beans on plasma glucose levels

rats feed on dark coffee bean had significantly higher than diabetic control group and lower than other experimental rats.

**Effect of Arabic coffee bean on plasma insulin levels:** The concentration of plasma insulin (Fig. 2) of diabetic rats decreased by -59.56% in comparison to the control group. Green and light coffee bean supplemented to the diet of experimental diabetic groups significantly increased the insulin concentration in plasma compared to the diabetic group and the same with normal control group. The green and light roasted Arabic coffee supplemented to the diet of diabetic rats increased the insulin concentration in plasma by 56.43 and 54.54% in comparison to the diabetic group. The diabetic rats fed on diet content dark coffee bean was significant lower insulin concentration than diabetic rats fed on diet content green and light groups; while significant higher by 34.5% compared to diabetic control group.

**Effect of Arabic coffee bean on plasma glucose level concentration:** The determination of plasma glucose concentrations were carried at the initial time, 2 week and at the final time are shown Fig. 3. At initial time, the diabetic groups showed significant differences concerning plasma glucose evaluation compared with the control. In the control group, no significant differences in the levels of the glucose level indices were noted between initial and final time. In contrast, diabetic rats fed on diet content coffee showed a significant decrease in serum glucose after two weeks. At the end of the study period, the experimental rats were showed significant improvement in blood glucose. It is noted that green coffee bean group has the best effect in decreasing glucose level followed by light coffee group followed by dark coffee group which recorded 95.46, 119.17 and 201.46 mg/dL, respectively.

**Effect of Arabic coffee bean lipids profiles:** Data in Table 2 revealed that diabetic control group showed a significant increase in the values of TC, TL, TG and a

Table 2: Effect of different roasting degree of coffee beans on lipid profiles

Groups	TC (mg/dL)	TL (mg/mL)	TG (mg/dL)	LDL (mg/dL)	HDL (mg/dL)
Control	102.36±3.45 <sup>d</sup>	213.48±6.35 <sup>d</sup>	96.25±2.36 <sup>d</sup>	36.23±1.45 <sup>e</sup>	46.88±2.56 <sup>f</sup>
Diabetic control	166.25±5.39 <sup>a</sup>	291.37±8.26 <sup>a</sup>	152.37±5.74 <sup>a</sup>	99.36±2.67 <sup>a</sup>	36.42±1.83 <sup>b</sup>
Green coffee	124.87±4.26 <sup>c</sup>	222.41±7.21 <sup>d</sup>	103.48±4.26 <sup>b</sup>	55.23±3.40 <sup>d</sup>	48.94±2.92 <sup>a</sup>
Light coffee	129.65±4.18 <sup>c</sup>	234.35±6.10 <sup>c</sup>	109.59±3.42 <sup>b</sup>	59.57±2.91 <sup>c</sup>	47.56±3.07 <sup>a</sup>
Dark coffee	146.23±3.97 <sup>b</sup>	276.16±6.54 <sup>b</sup>	143.42±4.25 <sup>c</sup>	78.34±3.78 <sup>b</sup>	39.21±2.33 <sup>b</sup>
LSD	5.67	9.34	4.06	2.85	3.09

Values are means±S.D. of 6 rats from each group; Means in the same column with different superscripts are significantly different (p≤0.05)

Table 3: Effect of Arabic coffee bean on MDA, GSH levels and enzymes activities SOD

Groups	MAD (nmol/mL)	SOD (mg/mL)	GSH (mg/mL)
Control (C)	6.25±0.46 <sup>f</sup>	19.85±1.06 <sup>a</sup>	9.37±0.76 <sup>a</sup>
Diabetic Control (DC)	13.36±0.89 <sup>a</sup>	8.34±0.85 <sup>e</sup>	2.64±0.13 <sup>d</sup>
Green Coffee (GC)	7.57±0.37 <sup>cd</sup>	13.89±1.04 <sup>c</sup>	7.36±0.53 <sup>b</sup>
Light Coffee (LC)	8.34±0.42 <sup>c</sup>	11.26±0.96 <sup>d</sup>	6.85±0.62 <sup>bc</sup>
Dark Coffee (DC)	10.62±0.56 <sup>b</sup>	15.37±1.18 <sup>b</sup>	4.56±0.41 <sup>c</sup>

Values are means±S.D. of 6 rats from each group; Means in the same column with different superscripts are significantly different (p≤0.05)

significant increase (p<0.05) in LDL when compared with normal control group. All treated groups showed a significant decrease in TC, TL and TG and a significant increase HDL compared with control group. The best reduction in the lipids profile was recorded for the Arabic green coffee supplement; the levels of total lipids, total cholesterol, triglycerides and LDL were decreased by 23.66, 24.89, 32.08 and 44.41%, respectively. A significant increase in the HDL level was observed for the Arabic green and light coffee bean supplement; and no significant difference in the HDL-C level was observed for normal control. The results show that Arabic dark coffee supplemented diets are lower effective against diabetic than green and light coffee.

**Effect of Arabic coffee bean on MDA, GSH levels and enzymes activities SOD:** MDA levels in plasma are illustrated in Table 3. A significant increase in MDA levels in plasma (132%) was observed in the diabetic rats group compared to those of the normal control. Diet supplemented with Arabic coffee beans induced a significant decrease of MDA levels in plasma compared to the diabetic control group. Antioxidant enzyme activities SOD and GSH levels in the plasma of control and experimental groups are shown in Table 3. In diabetic group, a significant decrease of GSH levels (-71.8%) and SOD activities was observed in plasma (-58.0%), respectively, as compared to the control group. Diet supplemented with Arabic coffee beans improved, GSH levels and SOD activities in plasma as compared to those of the diabetic group.

## DISSUASION

Roasting is an essential step in coffee production for generating aroma, flavor and color of the coffee beans. The mode of heat transfer and the applied temperature profile are the most critical processing parameters that affect the physical and chemical properties of roasted coffee beans (Schenker *et al.*, 2002). The moisture content is sensitive to the

temperature and time used during roasting. During coffee roasting, there are two major phases: dehydration and pyrolysis. Most of the water is lost during dehydration, at the beginning of the roasting process, reaching very low levels. During pyrolysis, there is still water loss, along with CO<sub>2</sub> and CO, however at a very slow rate (Montavon *et al.*, 2003).

Phenolic compounds are widely distributed in fruits and vegetables (Li *et al.*, 2006), which have received considerable attention because of their potential antioxidant activities and free radical-scavenging abilities, which potentially have beneficial implications in human health (Lopez-Velez *et al.*, 2003; Govindarajan *et al.*, 2007). At higher roasting degrees, damage to sensory characteristics and radical-scavenging activity of coffee beans is described as the main disadvantages of dark roasting. Therefore, degradation of polyphenol compounds by thermal process may result in releasing antioxidant compounds that have different chemical and biological properties (Tsai *et al.*, 2002). Phenolic compounds contributed directly to antioxidant activity and therefore, it was necessary to investigate the total phenolic content. These values are expressed as milligrams of GAE per gram of coffee beans grown under shaded conditions (Table 1). As mentioned, the content of phenolic acid in roasted coffee beans was influenced by the roasting time and temperature. Similar observations were made by Baggenstoss *et al.* (2008). Thermal processing has been reported to have both adverse and favorable effects on total phenolic acid. Roy *et al.* (2007) and Chan *et al.* (2009) found that in vegetables, phenolic content was reduced by thermal processing. Therefore, degradation of polyphenol compounds by thermal processing may result in releasing antioxidant compounds that have different chemical and biological properties (Tsai *et al.*, 2002).

The DPPH assay is a widely used assay to investigate the antioxidant potential of extracts in a preliminary test (Montavon and Bortlik, 2004). DPPH is a free radical compound with a stable free radical with a characteristic absorption at 517 nm. As antioxidants donate protons to this radical, the absorption decreases. The antioxidant activity of the natural plant extracts is generally attributed to their hydrogen donating ability (Klein *et al.*, 1991). It is well known that free radicals cause auto-oxidation of unsaturated lipids in food (Kaur and Perkins, 1991). Antioxidants are believed to intercept the free radical

chain of oxidation by donating hydrogen from the phenolic hydroxyl groups, thereby forming a stable end product, which does not initiate or propagate oxidation of the lipids (Sherwin, 1978). The results indicated that roasting degree (light and dark) significantly influenced the phenolic compositions as well as antioxidant activity of the coffee beans. The light-roasted coffee gave higher antioxidant activity and total phenolic content of coffee beans than dark roasted coffee but less than unroasted (green) bean. Meanwhile, Del Castillo *et al.* (2002) reported that maximum antioxidant activity was observed in Colombian Arabica coffee beans for the medium-roasted coffee (233°C 3 min), while the dark coffee (240°C 3 min) had a lower antioxidant activity.

Elevated serum glucose and insulin in diabetic control group as compared to normal control group confirm uncontrolled hyperglycemia, whereas green, roasted and dark coffee decreased serum glucose but increased insulin ( $p < 0.05$ ) with the lowest glucose value observed with green coffee and with light coffee for insulin. Also, the most important result of the present study was that rats, fed on green and light Arabic coffee enriched diet, were able to partly recover from alloxan-induced diabetes within a short time compared with rats fed control diet. These results are with the line of Van Dam (2008) who found that frequent consumption of coffee may reduce risk of type 2 diabetes and liver cancer. Several plausible mechanisms for a beneficial effect of coffee on glucose metabolism exist. Coffee has been shown to be a major contributor to the total in vitro antioxidant capacity of the diet (Pulido *et al.*, 2003) which may be relevant as oxidative stress can contribute to the development of type 2 diabetes. Coffee is the major source of the phenol chlorogenic acid. Intake of chlorogenic acid has been shown to reduce glucose concentrations in rats (Andrade-Cetto and Wiedenfeld, 2001; Rodriguez de Sotillo and Hadley, 2002) and intake of quinides, degradation products of chlorogenic acids, increased insulin sensitivity in rats (Shearer *et al.*, 2003). Chlorogenic acid contributes to the antioxidant effects of coffee, (Clifford, 1999) may reduce hepatic glucose output through inhibition of glucose-6-phosphatase, (Arion *et al.*, 1997) and may improve tissue mineral distribution through its action as a metal chelator (Rodriguez de Sotillo and Hadley, 2002). In addition, chlorogenic acid acts as a competitive inhibitor of glucose absorption in the intestine.

Green, light roasted and dark coffee resulted in a significant decrease ( $p < 0.05$ ) in triglycerides (TAG); LDL-C; and total cholesterol. On the other hand a significant increase ( $p > 0.05$ ) in serum HDL-C is observed in green, roasted and dark coffee groups compared to diabetic rats compared to normal control with the highest value for green coffee and light roasted coffee. Our results are in agreement with those of Kempf *et al.* (2010) who reported that coffee consumption led to an increase in coffee-derived

compounds, mainly serum caffeine, chlorogenic acid and caffeic acid metabolites. Significant changes were also observed for serum concentrations of interleukin-18, 8-isoprostane and adiponectin (8 compared with 0 cups coffee/d). These indicate that coffee consumption appears to have beneficial effects on subclinical inflammation and HDL cholesterol. It has been hypothesized that one of the principal causes of diabetes-induced injury is the formation of lipid peroxides by free radical derivatives. Thus, the antioxidant activity or the inhibition of the generation of free radicals is important in the protection against diabetes-induced hepatopathy (Castro *et al.*, 1974). The body has an effective defense mechanism to prevent and neutralize the free radical-induced damage. This is proficient by a set of endogenous antioxidant enzymes such as SOD and CAT. These enzymes constitute a mutually supportive team of defense against ROS (Amresh *et al.*, 2007). In diabetes, the balance between ROS production and these antioxidant defenses may be lost, resulting in oxidative stress which, through a series of events, deregulates the cellular functions leading to hepatic necrosis, for example. The reduced activities of SOD point out the tissues damage in the diabetic rats. Diabetic rats group showed a significant increase in the level of these enzymes as compared to experimental groups, which indicates the antioxidant activity of the seed. Regarding non enzymic antioxidants, GSH is a critical determinant of tissue susceptibility to oxidative damage and the depletion of GSH has been shown to be associated with an enhanced toxicity to chemicals (Hewawasam *et al.*, 2003), including diabetic status. In the present study, a decrease in plasma GSH level was observed in diabetic group. The increase in plasma GSH level in the diabetic rats fed on green and light coffee may be due to the novo GSH synthesis or GSH regeneration.

The level of lipid peroxide (MDA) is a measure of membrane damage and alterations in the structure and function of cellular membranes. In the present study, the elevation of lipid peroxidation in the plasma of diabetic rats was observed. The increase in MDA levels suggests an enhanced lipid peroxidation leading to tissue damage and the failure of antioxidant defense mechanisms to prevent the formation of excessive free radicals (Amresh *et al.*, 2007). Supplementation of green and light roasted Arabic coffee significantly reversed these changes. Hence, it is possible that the mechanism of hepatoprotection may be due to its antioxidant activity.

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