Food and Public Health 2013, 3(4): 228-233
DOI: 10.5923/j.fph.20130304.07

Different Doses of Arabic Coffee Improve Serum Lipid Profile, Uric Acid and Liver Enzymes of Experimental Rats

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Abstract Coffee mixed with cardamom is a traditional beverage in Arab countries. The study aimed at investigates the effect of consumption of different levels of Arabic coffee on blood parameters concentration. Thirty two male rats (130±2.3 g) were divided into four equal groups (n=8), control, single dose of coffee (ACS), double dose of coffee (ACD), triple dose of coffee (ACT) and received daily an oral dose of 2 ml normal saline, 8.6, 17.1 and 25.7 mg Arabic coffee mixture/100 g BW respectively. The experiment continued for thirty days, at the end, rats were anesthetized and killed by exsanguinations. Blood samples were collected and used for determination of triglycerides, total cholesterol, HDLc, LDLc and VLDLc, urea, uric acid, ALT, and AST. Samples of kidneys, heart, and liver were collected for histopathological examinations. The results revealed that there was a decrease in body weight, cholesterol, LDLc, HDLc, uric acid concentration, and liver enzymes in rats received coffee at all levels. Liver enzymes were found to be significantly ($P<0.05$) lower in ACT than control and other groups. Histopathological results indicated no evidence of kidneys dysfunction or pathological alterations in other organs. In conclusion, the consumption of Arabic coffee, at different levels, could be beneficial for patients with hypercholesterolemia or hyperuricemia.

Keywords Arabic Coffee, Cholesterol, LDLc, Uric Acid, Rats

1. Introduction

Arabic coffee (Coffee Arabica) is a name that refers to two types of coffee in some Arab countries: Turkish, and Saudi Coffee. The Saudi coffee ,or ''Al-Qahwa'' is made from coffee beans roasted very lightly or heavily between 165 -210°C and cardamom. Traditionally, it is roasted on the premises, ground, brewed and served in front of guests. This brewing method is common in Najd and Hijaz, and sometimes prepared with other spices like saffron (to give it a golden color), cloves, and cinnamon. It is often served with dates[1]

Viani, 1993[2] reported that coffee contains hundreds of biologically active compounds; phenolic polymers(8g/100g), polysaccharides (6g/100g), chlorogenic acids (4g/100g), minerals (3g/100g), organic acids (0.5g/100g), sugars (0.3g/100g) and lipids (0.2g/100g). Viani 1993[2] reported that of these compounds; chlorogenic acid in coffee mediates the anti-diabetic effects of coffee. Epidemiologic studies when examined coffee consumption indicated that drinking large amounts of coffee drastically reduced the incidence of type-2 diabetes[3,4]. Paradoxically,it was found that the consumption of caffeine (5mg/kg) in coffee results in impaired glucose tolerance in both healthy[5] and obese individuals[6].

Regarding the relationship between coffee consumption and health, a meta-analysis of several trials[7] reported that coffee consumption more than once per day lead to a slight increase in blood pressure. Similarly, coffee consumption may increase the risk of acute myocardial infarction[8] and stroke[9]. Chronic consumers of boiled coffee have been reported to elevate plasma cholesterol concentrations[10]. Intake of boiled coffee is associated with an increased risk of coronary heart disease[11]. Two constituents of ground coffee, cafestol and kahweol, were found to be responsible for the cholesterolemic action of boiled coffee[12]. The cholesterol-raising effect of diterpenes from coffee oil, present in boiled coffee, seems to be specific for human primates[13]. In Saudi Arabia, Shushan[14] found that consumption of Arabic coffee led to elevation of total cholesterol, LDLc, triglycerides, ALT, and AST, and decreased HDLc.

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Published online at http://journal.sapub.org/fph

Cross-sectional study investigated the link between coffee consumption and serum uric acid level and found a significant inverse association between coffee consumption and serum uric acid[15].The findings of Choi and Curhan[16] suggested that coffee consumption is associated with lower serum uric acid level. They added that serum uric acid decreased with the coffee intake of 4 to 5 and >6 cups daily. It has been speculated that non caffeine xanthines contained in coffee may inhibit xanthine oxidase, thus contributing to decrease serum uric acid levels[15].

With the widespread consumption of Arabic coffee in Saudia Arabia, the potential health effects of coffee are important for public health as well as for helping an individual make an informed choice regarding amounts of coffee consumption. To examine these issues, we conducted a study on experimental animals to find out the association between consumption of different doses of Arabic coffee and blood lipids, liver enzymes, urea, and uric acid concentrations.

2. Materials and Methods

2.1. Preparation of Arabic Coffee

Coffee seeds (*Coffee Arabica*) were obtained from local markets at Riyadh city. The coffee used in this study was prepared according to the traditional Saudi method. The coffee seeds were roasted for 10 min then milled and turned into powder. The coffee drink was prepared by boiling 30 g of coffee powder in one liter of water for 20 min.

The rats' dose was calculated according to the corresponding amount consumed by an adult person. The adult person who weigh 70kg consumes on average 5 small cups of coffee daily (about 150ml/day), this amount contain about 6 gram of Arabic coffee powder. Therefore, the normal dose for human would be 6000 mg/ 7 kg of body weight and for rat would be 8.6mg/100gram of body weight. The coffee powder was dissolved in 2 mL distilled water and administrationorally to experimental rats.

2.2. Animals

Thirty two male albino Sprague dawley rats weighing 125 – 135 grams were used. The animals were housed in standard cages and divided into four equals groups of 8 rats each. Each group was fed a defined basal diet plus water ad libitum. The standard diet is composed of casein (15%), sucrose (5%), fats (10%), vitamin mixture (1%), salt mixture (4%), fiber (4%), and starch up to 100%.

2.3. Experimental Feeding Groups

The control group was on the basal diet only. The first group (ACS) received single oral dose of Arabic coffee powder (8.6 mg/ 100 g). The second group (ACD) received double dose of Arabic coffee (17.1mg/ 100g). The third group (ACT) received orally triple dose of Arabic coffee (25.7 mg/ 100g).

The experimental continued for 30 days. The weights of the rats were measured at the beginning and end of the experimental period. At the end of the period, the rats were fasted for 8 hr then anesthetized with diethyl ether and killed by exsanguinations. Blood samples were collected in heparinzed tubes. All blood samples were immediately centrifuged (3,000 rpm, 20 min, and 4℃) for the separation of plasma. The plasma was stored at -20℃ until analysis. Kidneys, heart, and liver were removed, cleaned, and weighed. Organ samples were taken for histological examination.

2.4. Biochemical Analysis

The following parameters were determined in the plasma: Urea was determined according to the method of Tietz, [17] Uric acid was determined according to Buchanan et al.[18], alanine aminotransferase (ALT) and aspartate amino transferas (AST) were determined according to the method of Rej[19]; serum triglyceride was determined according to Stein and Myers[20]; total cholesterol according to the method of Trinder[21]; and High density lipoprotein cholesterol (HDLc) was determined according to Lopez et al.,[22]; and LDLc and VLDLc were calculated according to the method of Van Horn et al.,[23].

2.5. Histopathological Examination

Samples of kidneys, liver, heart were taken and fixed in 10% neutral buffered formalin for 24 hours. Paraffin sections 6 µm thick were prepared and stained with hematoxylin and eosin (H & E)[24] for the examination by light microscopy.

2.6. Statistical Analysis

All values were expressed as means±SD. Data were initially analyzed using the analysis of variance for each group (One Way ANOVA). When a significant F-value ($p < 0.05$) was obtained, LSD multiple test was performed for post hoc analysis.

Table 1. Initial weight, final weight, and food intake of rats fed Arabic coffee

	Control (n=8)	ACS (n=8)	ACD (n=8)	ACT (n=8)	ANOVA	
	Mean±SD	Mean±SD	Mean±SD	Mean±SD	F	P value
Initial body weight (g)	132.2±3.7	126.6±1.1	129.0±2.9	127.8±1.5	1.3	0.21
Final body weight (g)	155.6±24.8	133.0±14.0	130.3±38.2	124.6±21.9	4.1	0.02
Change in body weight (%)	+ 17.7%	+ 5.1%	+1.0%	- 2.5%		
Food intake (g/day)	12.8±2.8	10.8±1.7	11.8±1.3	11.6±2.1	0.7	0.55

ACS: Arabic coffee (Single dose); ACD: Arabic coffee (Double dose); and ACT: Arabic coffee (Triple dose. ANOVA: Analysis of variance. SD: Standard deviation. Mean values subscribed with different letters show significant differences between these values as calculated by ANOVA and LSD at $P<0.05$.

3. Results

Changes in body weights of experimental rats are presented in Table (1).

Table 2 show the relative liver and kidneys weight of groups fed different concentrations of Arabic coffee.

Table 2. Relative liver, kidney, and heart weight (% BW) of rats fed Arabic coffee

	Control (n=8)	ACS (n=8)	ACD (n=8)	ACT (n=8)	ANOVA	
	Mean±SD	Mean±SD	Mean±SD	Mean±SD	F	P value
Relative liver weight (%BW)	3.3±0.9	3.2±0.3	3.9±0.1	3.6±0.5	1.2	0.35
Relative kidneys weight (%BW)	1.2±0.3	1.0±0.1	1.3±0.2	1.3±0.2	2.2	0.13
Relative heart weight (%BW)	1.4±0.5 a	1.1±0.2 a	1.2±0.3 a	0.9±0.1 b	5.2	0.01

ACS: Arabic coffee (Single dose); ACD: Arabic coffee (Double dose); and ACT: Arabic coffee (Triple dose). ANOVA: Analysis of variance. SD: Standard deviation. Mean values subscribed with different letters show significant differences between these values as calculated by ANOVA and LSD at P<0.05.

Concentration of serum uric acid, urea, AST, and ALT of rats fed different concentrations of Arabic coffee are presented in table 3.

Table 3. Serum uric acid, urea, AST, and ALT of rats fed Arabic coffee

	Control	ACS	ACD	ACT	ANOVA	
	Mean±SD	Mean±SD	Mean±SD	Mean±SD	F	P value
Uric acid (mg/dl)	2.3±1.2 a	1.7±0.6 b	1.5±0.3 bd	1.3±0.2 d	1.9	0.17
Urea (mg/dl)	57.0±3.5 a	82.4±15.9 b	81.8±26.5 b	58.0±14.1 a	3.5	0.04
AST (U/l)	251.6±54.9 a	267.6±29.3 a	244.2±32.9ab	228.4±68.4 b	0.6	0.65
ALT (U/l)	54.4±17.2 a	52.4±11.8 a	48.2±9.4 ab	41.0±10.8 b	1.1	0.37

ACS: Arabic coffee (Single dose); ACD: Arabic coffee (Double dose); and ACT: Arabic coffee (Triple dose). ANOVA: Analysis of variance. SD: Standard deviation. Mean values subscribed with different letters show significant differences between these values as calculated by ANOVA and LSD at P<0.05.

Results presented in table 4 showed the effect of arrabic coffee on serum triglycrides, total cholesterol, HDLc, LDLc, and VLDLc.

Table 4. Serum triglycerides, cholesterol, VLDLc, LDLc, HDLc, and total lipids of rats fed Arabic coffee

	Control	ACS	ACD	ACT	ANOVA	
	Mean±SD	Mean±SD	Mean±SD	Mean±SD	F	P value
TG (mg/dl)	142.2±33.2 a	152.0±19.1 a	209.0±29.4 b	144.0±12.7 a	10.5	P<0.001
TC (mg/dl)	126.0±7.0 a	109.0±10.9 b	105.3±8.3 b	103.2±10.7 b	8.6	0.001
HDL (mg/dl)	40.9±6.7 a	33.3±6.8 b	28.6±3.6 b	31.5±5.1 b	5.9	0.004
LDL (mg/dl)	56.7±12.3 a	45.3±8.3 ab	40.9±7.7 b	42.9±12.6 b	5.2	0.007
VLDL (mg/dl)	28.4±6.6a	30.4±3.8 a	41.8±5.9 b	28.8±2.5 a	10.5	P<0.001

ACS: Arabic coffee (Single dose); ACD: Arabic coffee (Double dose); and ACT: Arabic coffee (Triple dose). TG= Triglycerides, TC= Total Cholesterol. ANOVA: Analysis of variance. SD: Standard deviation. Mean values subscribed with different letters show significant differences between these values as calculated by ANOVA and LSD at P<0.05.

Histopathological changes associated with Arabic coffee treatment are presented in table (5).

Table 5. Histopathological results for control and coffee groups

Organ	Histopathology	Study groups			
		Control	ACS	ACD	ACT
Liver	Fatty degeneration.	N	+	+	++
	Necrotic cells.	N	N	+	N
	Congestion	N	N	+	N
	Inflammatory cell	N	N	+	+
	dilated of blood vessels	N	N	+	+
Kidney	Hydrophobic degeneration	N	N	+	+
	Necrotic cells	N	N	+	N
Heart	Cross striation.	N	N	+	+
	Necrotic cells.	N	N	+	++
	Cystic space between cardiac muscle fiber.	N	N	+	+

Normal= N, Mild = +, Moderate= ++, and Severe = +++

4. Discussion

The results showed that rats of control group gained more and significant body weight than rats fed different concentration of Arabic coffee, while there was a slight increase in body weight in rats fed single or double dose. On the other hand, group fed triple dose of Arabic coffee lost considerable amount of body weight, which in turn mean that drinking high amounts of Arabic coffee might have negative effect on body weight. Similar finding was reported by Eun-Young et al.[25] and Vinson et al.[26]. However, this loss in body weight accompanying high intake of Arabic coffee may be due to caffeine where Lopez-Garcia et al.,[27] concluded that increases in caffeine intake might lead to a small reduction in long-term weight gain. Several studies tried to explain the mechanism by which caffeine decreased body weight like Acheson et al.[28] who suggested that caffeine might stimulate thermogenesis by increasing lipid turnover. Other studies observed association between decaffeinated coffee and smaller weight gain which may indicate that the effect of coffee could be due to compounds other than caffeine. For example, chlorogenic acid in coffee is able to attenuate glucose absorption in the digestive track, which could help control weight[29]. In addition, the results of Shimoda et al.[30] suggested that green coffee is possibly effective against weight gain and fat accumulation by inhibition of fat absorption and activation of fat metabolism in the liver.

The results of this study showed that the higher the intake of Arabic coffee the lower the serum uric acid concentration. Shushan[13] reported similar finding. However, in their studies on Japanese and American population. Two studies Kiyohara et al.[31] and[15] found that individuals who consumed 5 cups of coffee daily had significantly low serum uric acid than individuals who consumed 1 cup daily. Some studies tried to find out the mechanism by which coffee decreased serum uric acid, however, because there is a strong positive relationship between serum insulin resistance and elevated serum uric acid[32] and it is well authenticated that insulin reduces the renal excretion of urate[33- 35], decreased insulin resistance, and insulin levels associated with coffee consumption may lead to lower uric acid levels. Another study speculated that non-caffeine xanthines contained in coffee may inhibit xanthine oxidase, thus contributing to lowering serum uric acid levels[34].

In this study it was found that the triple dose of Arabic coffee reduced serum urea but the results were inconsistent because of the mean values of single and double dose were significantly ($P<0.05$) higher than control and triple dose group. Although, the elevated blood urea is known to be related to increased protein breakdown or kidney dysfunction. According to organs weight and histopathological results there is no evidence that protein breakdown increased or kidneys dysfunction.

In comparison with control and rats fed single or double dose, the triple dose of Arabic coffee decreased significantly the serum levels of major liver enzymes (AST and ALT). this favorable effect of high intakes of coffee on liver enzymes in this study was in agreement with results obtained by Honjo et al,[36] and Tanaka et al.[37] who investigated the potential relationship between coffee consumption and alanine (ALT) and aspartate (AST) aminotransferase, and found that coffee intake was significantly related to decreased serum concentrations of both enzymes., Ruhl and Everhart[38], found that drinking was related to a lower prevalence of high AST and ALT levels and this can explain the results obtained in this study and the reports presented by Honjo et al.[36] and Tanaka et al.[37]

Our results revealed that Arabic coffee had favorable effects on serum total cholesterol and LDLc, where it decreased their concentration significantly. These results seemed to be opposite to that obtained by Shushan[13] who found that Arabic coffee alone or with other ingredients like cardamom increased the levels of total cholesterol and LDLc. On the other hand, other studies carried out in Saudi Arabia showed that Arabic coffee consumption have no significant effect on serum cholesterol or LDLc levels[39,40]. However, Eun-Young et al.[25], in their study found that Coffee intake increased total cholesterol and LDLc levels in serum of rats fed coffee when compared with control group.

However, Arabic coffee had negative effects on serum HDLc; it decreased its concentration significantly. Similar results were obtained by Eun-Young et al.[25] who found that feeding rats coffee resulted in significant decrease of HDLc in serum. However, this may be due in part to the reduction of total cholesterol and hence reduction of its

concentration in different lipoproteins including HDLc.

5. Conclusions

Arabic coffee was found to reduce body weight and may be beneficial for obese persons, it also decreases liver enzymes and this may helpful in improving health status of hepatic patients. Finally coffee consumption reduced total cholesterol and LDLc and therefore it may be consumed in considerable amounts for reducing cholesterol problems.

ACKOWLEDGEMENTS

The authors thank Professor H.M.Mousa for revising the manuscript and Mr.Ali Yousif for the technical support in Histopathology.

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