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Different Preparations of Coffee Have Varied Effects on Body Weight and Blood Lipids in Experimental Rats

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Abstract

Coffee beverage is a globally consumed and is prepared in different ways. This study aims at finding out the effects of different preparations of coffee on body weight and blood lipids in experimental rats. Forty male albino rats (130 ± 2.3 g) were divided into five equal groups (n=8), control, Turkish coffee medium roasting (TMC), Turkish coffee dark roasting (TDC), instant coffee (IC), and Arabian coffee (AC). Each group received 2 ml oral dose of coffee (0, 4.3, 4.3, 14.3, 8.6 mg/100 g BW respectively). The experiment continued for 30 days, and by the end, rats were anesthetized and killed for collection of blood samples. Triglycerides, total cholesterol, HDLc, LDLc, VLDLc, and total lipids were determined in serum. Samples of liver, kidney, and heart were collected for histopathological examination. Results showed that rats fed different preparations of coffee had significantly smaller weight gain than control group. On the other hand, group fed instant coffee lost considerable amount of body weight. Among all kinds of studied coffee, the Turkish coffee dark roasting reduced significantly ($P<0.05$) serum triglycerides, total cholesterol, LDLc, VLDLc, and total lipids, whereas it elevated HDLc concentration. Moreover, group fed Instant coffee showed also lower blood lipids. In conclusion, moderate amounts of Turkish coffee dark roasting have desirable effects on serum total cholesterol, LDLc, and total blood lipids.

Keywords: Roasted; Coffee; Cholesterol; LDL; HDL; Rats; Body weight

Introduction

Coffee beverage is a globally consumed and is prepared in a wide variety of forms. People consume coffee in different forms (e.g., 37 °C – 88 °C; 0% – 80% milk; 0 g–16 g of sugar; 25 mL–880 mL in volume; with or without milk; foamed milk; cream; ice; flavorings; brew adjuncts or co-adjuncts) [1].

A method for preparing is the Turkish coffee (*Türk kahvesi*): coffee beans are roasted and then finely ground, the ground coffee beans are boiled in a pot, usually with sugar, and served in a cup where the grounds were allowed to settle.

Nescafé is a brand of coffee (instant coffee) made by Nestlé. It comes in many different product forms. The name is a mixture of the words "Nestlé" and "café". Nestlé first introduced their flagship powdered coffee brand in Switzerland on April 1, 1938 [www.nescafe.com].

Arabian coffee (Coffee Arabica) is a name that refers to Saudi coffee, or ''*Al-Qahwa*'' made from coffee beans roasted very lightly or heavily from 165 °C (329 °F) to 210 °C (410 °F) and cardamom [2]. Sometimes Arabian coffee is prepared with other spices like saffron (give a golden color), cloves, and cinnamon [3].

Some researchers observed that coffee might increase the risk of chronic diseases. For example, Jee et al. [4] found that coffee consumption for more than once a day led to a slight increase in blood pressure. Similarly, Chown et al. [5] and Keijzers et al. [6] observed that consumption of high amounts of coffee resulted in impaired glucose tolerance. In addition, de Roos et al. [7] showed that chronic consumption of boiled coffee has permanently elevated plasma cholesterol concentrations. Moreover, coffee consumption might increase the risk of acute myocardial infarction [8] and stroke [9].

On the other hand, epidemiologic studies indicated that drinking large amounts of coffee drastically reduced the incidence of type-2 diabetes [10,11].

Roasted coffee contains naturally antioxidants and other compounds that formed during the roasting process [12]. Caffeine and chlorogenic acids have been extensively studied because they may reduce the risk of insulin resistance [13,14], and development and progression of atherosclerosis [15] and they might decrease blood pressure (BP) [16,17].

On the other hand, earlier studies [18] found that Scandinavian boiled type of coffee, prepared by boiling coarsely ground coffee beans with water without filtration, elevated serum cholesterol and triglycerides. The diterpenoid alcohol cafestol, possibly together with kahweol, is responsible for these effects [19]. Hence, unfiltered coffee, like Scandinavian boiled and Turkish coffee, contains much higher concentrations of diterpenes than filtered coffee. While, espresso coffee contains intermediate amounts [20].

Importantly, the concentration of these compounds depends on how coffee is prepared. Boiled coffee has higher concentrations because diterpenes were extract from the coffee beans by prolonged contact with hot water. By comparison, brewed/filtered coffee, because of the much shorter contact with hot water and retention of diterpenes by filter paper, has a much lower concentration of cafestol and kahweol. A study used coffee that brewed by two common methods, filtering and boiling, observed a significant increase in total cholesterol and a non-significant increase in low-density lipoprotein (LDL) cholesterol

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in subjects consuming boiled coffee. While, there was no significant difference in the change in serum total or LDL cholesterol levels between the filtered-coffee group and the group who drank no coffee [18]. These results replicated in a meta-analysis of 14 randomized controlled trials in which the consumption of boiled coffee increased total and LDL cholesterol concentrations in serum, meanwhile the consumption of filtered coffee resulted in very little change in serum cholesterol [4]. Lopez-Garcia et al., [21] found in their large cohort study no impact of filtered coffee on total cholesterol, LDL and HDL levels.

However, most of cafestol and kahweol were retained by the paper filter or precipitated in the cup, which substantially reduces the cholesterol-raising effects potentially associated with coffee [22,23].

Data related to the effect of different preparations of coffee on blood lipids are insufficient; therefore, we carried out this study to find out the effects of different preparations of coffee (Turkish coffee - of both moderate or dark roasted, instant coffee, and Arabian coffee) on blood lipids in experimental rats.

Materials and Methods

Preparation of coffee

All coffee used in this study were obtained freshly from local markets in Cairo, Egypt, except for Arabian which obtained from local markets at Riyadh, Kingdom of Saudi Arabia. The coffee was prepared by traditional methods, and without adding sugar, sweeteners, creamer, or milk.

Turkish coffee (medium and dark roasting): In this experiment, we used two types of Turkish coffee; medium roasting (in which fresh coffee beans were roasted to brown color at temperature ranging from 210 °C to 220 °C) and dark roasting (in which fresh coffee beans were roasted to brown color at temperature ranging from 240 °C to 250 °C). After roasting, the beans are ground to the finest possible powder. The brew was prepared by immersing the coffee powder (5 g coffee / 100 ml water) in hot water, for just as the coffee comes to the boil, the pot is removed from the heat, then allowed to cool and the solution was separated and used in the experiments (The coffee foam were removed).

Instant coffee: In this experiment, we used the most common trademark in Egypt. It is instant coffee and ready to use, and the brew prepared by dissolving 5 grams of instant coffee in 100 ml of hot water.

Arabian coffee (Coffee Arabica): The coffee seeds were roasted for 10 minutes, then milled and turned into powder. The brew prepared by boiling 30 g of coffee powder in one liter of water for 20 min [2].

Coffee doses

The rat's dose of coffee was calculated according to the corresponding amounts consumed by the adult person. Researchers suggested that adult person who weighs 70 kg consumes an average:

1- Two small cups (beaker) of Turkish coffee daily (about 60 ml/ day), this amount contains about 3 grams of Turkish coffee powder. Therefore, the normal dose of coffee for human would be 3000 mg/ 70 kg of body weight and for rat would be 4.3 mg/ 100 gram of body weight per day.

2- One medium cup of Instant coffee daily (about 200 ml/day), this amount contains about 10 grams of Instant coffee. Therefore, the normal dose of Instant coffee for human would be 10000 mg/ 70 kg of body weight and for rat would be 14.3 mg/ 100 gram of body weight per day.

3- Five small cups of Arabian coffee daily (about 150 ml/day), this amount contains about 6 grams of Arabian coffee powder. Therefore, the normal dose of Arabian coffee for human would be 6000 mg/ 70 kg of body weight and for rat would be 8.6 mg/ 100 gram of body weight per day.

Animals

Forty male Albino rats weighing 115–135 grams (130 ± 2.3 g) were purchased from Animal Unit at Helwan, Ministry of Health, Egypt. The rats were housed individually in cages, and were kept at 22 C, 56% humidity (40 to 70%) and in a 12-h: 12-h light: dark cycle and were allowed free access to food and tap water.

After 7 days of acclimatization, rats were randomly allocated to five equal groups (8 rats for each). The study was conducted in the animal lab at Faculty of Home Economics, Minufiya University, Shibin El-Kom, Egypt.

Each group was fed a defined basal diet plus water ad libitum. The basal diet was composed of protein (20%), sucrose (5%), fats (10%), vitamin mixture (1%), salt mixture (4%), fiber (4%), and starch up to 100%.

Experimental Feeding Groups.

The control group kept on the basal diet only. The first group (TMC) received single oral dose of Turkish coffee medium roasting (4.3 mg/ 100 g/day). The second group (TDC) received single oral dose of Turkish coffee dark roasting (4.3 mg/ 100 g/day). The third group (IC) received single oral dose of Instant coffee (14.3 mg/ 100 g/day). The fourth group (AC) received single oral dose of Arabian coffee (8.6 mg/ 100 g/day). All coffee solutions were fed to animals orally on daily basis.

The experiment continued for 30 consecutive days. The weight of the rats was measured at the beginning and at the end of the experimental period. By the end, the rats were fasted for 8 hr, then anesthetized with diethyl ether and killed by exsanguinations. Blood samples were collected in heparinized tubes, and were immediately centrifuged (3,000 rpm, 20 min, and 4°C) for the separation of serum. The serum was stored at -20°C until analysis. Liver, kidney, and heart samples were taken for histological examination.

Biochemical analysis

The following parameters were determined in the serum: triglyceride [24]; total cholesterol [25]; HDLc [26]; and LDLc and VLDLc were calculate according to the method of Van Horn et al. [27].

Histopathological examination

Samples of kidneys, liver, heart were taken and fixed in 10% neutral buffered formalin for 24 hr. Paraffin sections 6 µm thick were prepared and stained with hematoxylin and eosin (H & E) 24 for the examination by light microscopy. The histopathology carried out in histology lab at Faculty of Veterinary Medicine, Cairo University, Egypt.

Statistical analysis

All values were expressed as means ± SD. Data were initially analyzed using the analysis of variance for each group (One Way ANOVA), and LSD multiple test was performed for post hoc analysis.

Results

The results of this study showed that body weight gain of control

group was significantly (P<0.05) the highest (+17.7%), while the IC group lost 4.2% of their weight (P<0.05). In parallel, the food intake of IC group was significantly lower than control group (P<0.05) (Table 1).

As shown in Table 2 the triglycerides and VLDLc values of AC and control group were significantly the highest, while the values of TDC group were significantly the lowest. It could be noticed from the same table that TDC and IC groups had significantly the lowest values of total cholesterol, LDLc, and total lipids while the TMC and control groups had significantly the highest values. The HDLc values of TMC were significantly the highest while the values of AC and control groups were significantly the lowest.

Histopathological changes associated with Turkish coffee (medium roasting), Turkish coffee (dark roasting), Instant coffee, and Arabian coffee treatment presented in Table 3.

For control group the examination revealed normal hepatic, kidney, and heart tissues. As for Turkish coffee (medium), the histopathological examination revealed mild degenerative changes in glomeruli and tubes, and minimal degenerative changes of hepatic cells in liver, while heart tissue was normal. For Turkish coffee dark roasting, the examination revealed mild degenerative changes of the renal tubules in renal tissue, while liver and heart were normal. For Instant coffee group it revealed moderate degenerative changes of hepatocytes in liver and mild degenerative changes of glomeruli and tubules, while the heart was normal. For Arabian coffee group the examination revealed no changes in kidney, and heart tissues while it revealed mild degenerative changes in hepatic tissue.

Discussion

In comparison with control group, the results showed that rats fed different preparations of coffee had significantly smaller weight gain than control group who gained more and significant body weight. On the other hand, group fed Instant coffee lost considerable amount of body weight, which in turn means that drinking Instant coffee reduces body weight. In addition, we noticed that the food intake by Instant coffee group was the lowest among studied groups, which may partially explain the loss of body weight. However, Lopez-Garcia et al. [21] found that coffee consumption had adverse effects on body weight, and attributed that effect to caffeine intakes. Some studies explained the relationship between coffee consumption and body weight. Some of those studies suggested that caffeine has several important metabolic effects and works as an adenosine-receptor antagonist [28], and all tissues with adenosine receptors can be affected by caffeine exposure. Astrup et al. [29] observed a dose-dependent increase in BMR with caffeine intake in healthy subjects who had moderate habitual caffeine consumption. The researchers attributed this effect to an increase in lactate and triacylglycerol production and increased vascular smooth muscle tone. Acheson et al. [30] suggested that caffeine might stimulate thermogenesis by increasing lipid turnover. All the above mechanisms suggested a beneficial effect of caffeine on energy metabolism.

The results of this study showed that among all studied coffee, the Turkish coffee dark roasting decreased significantly (P<0.05) serum triglycerides, total cholesterol, LDLc, VLDLc, and total lipids, while it elevated HDLc concentration. Moreover, Instant coffee had also favorable effects on blood lipids. In agreement with our results, Yukawa et al. [31] found that serum levels of total cholesterol and LDLc significantly decreased after coffee consumption. In addition, Kempf et al. [12] found that coffee increases HDL cholesterol, which may protect against the development of atherosclerosis.

It is well known that coffee contains cholesterol-raising compounds known as cafestol and kahweol [32-34]. However, our results showed that dark roasted coffee and instant coffee decreased serum cholesterol, we assume that most of cafestol and kahweol may be precipitated in dark roasted coffee or not fully extracted in Instant coffee. In addition, coffee contained besides caffeine hundreds of biologically active compounds e.g., phenolic polymers and chlorogenic acids. Several studies [35-38] reported that coffee polyphenols and chlorogenic acid reduce cholesterol levels. However, little has been reported about the mechanism of action of these substances: it has been reported only that chlorogenic acids inhibits cholesterol biosynthesis [36,37]. Moreover, Harumi et al. [39] found that phenolic acids of coffee enhance cholesterol efflux and decrease its blood concentrations. In addition, roasted coffee contain Quinides [40] that may have favorable effects on serum cholesterol and blood lipids.

Regarding the effect of roasting, medium roasted coffee in our study increased cholesterol, LDL, and total lipids, these findings were supported by findings obtained by Telma et al. [41] who found that

	Control (n=8)	TMC (n=8)	TDC (n=8)	IC (n=8)	AC (n=8)	ANOVA	
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	F	P value
Initial body weight (g)	132.2 ± 3.7a	123.0 ± 2.3b	121.0 ± 2.9b	117.2 ± 2.0b	126.6 ± 1.1b	109.8	0.001
Final body weight (g)	155.6 ± 24.8 a	132.4 ± 18.8b	127.7 ± 11.6b	112.3 ± 13.7c	133.0 ± 14.0b	6.0	0.042
% Change in body weight	+17.7%	+7.6%	+5.5%	-4.2%	+5.1%		
Food intake (g/day)	12.8 ± 2.8 a	11.3 ± 2.2 ab	10.8 ± 2.2 ab	9.0 ± 2.0 b	10.8 ± 1.7 ab	1.6	0.209

Table 1: Body weights and food intakes of experimental rats. TCM=Turkish coffee (medium roasting); TCD=Turkish coffee (dark roasting); IC=Instant coffee; AC=Arabian coffee. ANOVA=Analysis of variance. SD=Standard deviation. Mean values subscribed with different letters show significant differences between these values as calculated by one way ANOVA and LSD at P<0.05.

	Control (n=8)	TMC (n=8)	TDC (n=8)	IC (n=8)	AC (n=8)	ANOVA	
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	F. value	P value
TG (mg/dL)	142.2 ± 33.2a	124.2 ± 19.9b	96.8 ± 43.2c	114.5 ± 22.0b	152.0 ± 19.1a	0.6	0. 008
TC (mg/dL)	126.0 ± 7.0a	140.0 ± 23.8b	99.4 ± 5.8c	96.8 ± 22.0c	109.0 ± 10.9a	2.0	0.141
HDLc (mg/dL)	40.9 ± 6.7a	61.6 ± 2.9b	54.2 ± 8.5c	50.8 ± 7.8c	33.3 ± 6.8a	3.9	0.019
LDLc (mg/dL)	56.7 ± 12.3a	53.6 ± 20.5a	25.8 ± 4.9b	23.1 ± 13.6b	45.3 ± 8.3a	0.6	0.685
VLDLc (mg/dL)	28.4 ± 6.6a	24.8 ± 4.0b	19.4 ± 8.6c	22.9 ± 4.4b	30.4 ± 3.8a	0.9	0.489
Total lipids (g/L)	4.3 ± 1.3a	4.3 ± 0.5a	3.6 ± 0.4b	3.8 ± 0.5b	4.2 ± 0.4a	1.1	0.395

Table 2: Concentration of blood lipids of experimental rats. TCM=Turkish coffee (medium roasting); TCD=Turkish coffee (dark roasting); IC=Instant coffee; AC=Arabian coffee. ANOVA=Analysis of variance. SD=Standard deviation. TG=Triglycerides; TC=Total Cholesterol. Mean values subscribed with different letters show significant differences between these values as calculated by one way ANOVA and LSD at P<0.05.

Organ	Histopathology	Study groups				
		Control (n=8)	TMC (n=8)	TDC (n=8)	IC (n=8)	AC (n=8)
Liver	Fatty degeneration.	N	+	+	++	+
	Necrotic cells.	N	+	+	++	+
	Congestion	N	+	+	++	+
	Inflammatory cell	N	+	+	++	+
	dilated of blood vessels	N	+	+	++	+
Kidney	Hydrophobic degeneration	N	+	N	+	N
	Necrotic cells	N	+	N	+	N
Heart	Cross striation.	N	N	N	+	N
	Necrotic cells.	N	N	N	+	N
	Cystic space between cardiac muscle fiber.	N	N	N	+	N

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

Table 3: Histopathological results for control and coffee groups.

both light and medium coffee roasts increase plasma total cholesterol and LDLc.

Although roasted Turkish coffee had slightly more caffeine [42] but we think that caffeine had no effect on blood lipids. The results of this study showed that Turkish dark coffee and Instant coffee had decreased the concentrations of cholesterol and other blood lipids, meanwhile the Turkish coffee contain approximately double fold of caffeine occurred in Instant coffee (approximately 84.7 mg vs. 37 mg of caffeine per 100 ml respectively) [42,43].

In comparison with other preparations of coffee, we assume that dark roasted coffee may contain biologically active compounds that are formed during the roasting process and these compounds are responsible for reduction of serum total cholesterol, LDLc and blood lipids. Meanwhile, we postulated that dark roasting decreased cholesterol-raising compounds in coffee.

Conclusion

In conclusion, moderate amounts of Turkish coffee dark roasting (without foam) have desirable effects and may reduce concentrations of serum total cholesterol, LDLc, and total blood lipids.

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