

EFFECT OF BLACK TEA, COFFEE AND CAFFEINE ON SERUM LIPID PROFILES IN RATS

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ABSTRACT

The relationship between black tea, coffee and caffeine consumption and serum lipids was examined in eight comparison groups of albino rats of eight weeks old in a range weight between 128-131 g. Animals given diets for additional four weeks. It contained either black tea or coffee or caffeine (123.3 mg caffeine/100 g diet or 76.8 mg caffeine/100 g diet) without or plus cholesterol (1%), bile salts (0.2%) and tallow (20%). The mean final body weights gain were not changed ($P \leq 0.05$) in all groups exception of coffee group which decreased significantly. The rats fed on diet of coffee or caffeine (123.3 mg/100 g) had no significant elevation of total cholesterol, triglycerides, HDL-cholesterol and LDL-cholesterol compared to the negative control (without cholesterol). Whereas total cholesterol, triglycerides and LDL-cholesterol had significant increase in coffee plus cholesterol group and caffeine (123.3 mg/100 g diet) group compared the positive control (hepercholesterolemic) group. There was no significant differences in group 7 (black tea plus cholesterol) and group 8 (caffeine 76.8 mg/100 diet) compared to the positive control. The ratio of HDL-cholesterol to total cholesterol and HDL-cholesterol to LDL-cholesterol were lower ($P \leq 0.05$) in group: 5, 6, 4 (coffee plus cholesterol, caffeine 123.3mg/100 g diet plus cholesterol and positive control), respectively.

In conclusion, these findings suggest that black tea, coffee and caffeine did not increase the serum lipids in rats consumed the normal balanced diet. Whereas coffee and high concentrations of caffeine led to raise the serum lipids in rats fed on high fat and high cholesterol diet. Black tea had no effect on serum lipids in rats fed on normal or hyperlipidemic diet.

INTRODUCTION

Elevated concentration of plasma triglycerol and LDL-cholesterol are important risk factors for the development of cardiovascular diseases (De Roos et al., 2001). Many researcheres have studied the effect of various dietary components on the level and distribution of plasma lipids. Alcohol, high fat consumption, and heavy smoking are conflicting factors complicating the interpretation of the results of the studies (Trompson *et al.*, 1992).

Black tea is one of the most popular beverage all over the world. Some studies indicated that black tea cause no change in the level of serum lipids (Green and Harari, 1992 and Van-het-Hof *et al.*, 1997).

The widespread use of coffee has led some to suspect that the habit might have detrimental effects on health. Almost a decade ago an analysis of avialble data in favor of or against coffee use concluded that there were no strong arguments supporting advice to abandon drinking coffee. In recent years, however, the possibility that coffee use is associated with an increased incidence of cardiovascular disease has gained renewed interest. The most prominent compound in coffee is caffeine, and it has been suggested that some of the risks related to coffee results from the effects of this substance (Bak and Grobbee, 1991). There was a significant association between serum cholesterol and total caffeine consumption irrespective of source for females only (Shirlow and Mathers, 1984). Caffeine, which is one of the most

active substances in coffee, is also found not only in tea but also in cacao, chocolate, carbonated beverages and in medical drugs (Schreiber *et al.*, 1988). Coffee could not be considered alone as a risk factor for coronary heart disease (Rakicioglu *et al.*, 1998).

To derive more conclusive evidence for the effect of black tea, coffee and caffeine on serum lipids, conducted trial with eight groups of 56 male albino rats of eight weeks old and 128-131 g weight was arranged.

MATERIALS AND METHODS

Coffee (*Coffea arabica* Linn), black tea (*Camellia sinensis*) El-Arosa were obtained from local market in Giza.

Preparation of black tea water extract (BTWE)

The BTWE was prepared by adding 5 g black tea (BT) leaves to 250 ml of freshly boiled water. After 15 minutes, the infusion was filtered.

Animals

Fifty six male albino rats of eight weeks old and in a range weight between 128-131 g were used. They were divided into eight groups each of seven rats. The animals were housed individually in stainless steel cages in an animal room at 25°C with a 12 hours light/dark cycle. Fresh diets and fluid were given adlibitum every day. Food intake was measured daily and body weight was recorded twice a week.

Diets

Group 1 (Negative control):

The animals consumed negative control diet (g/100 g) included casein, 15g; corn starch, 73.5 g; corn oil, 2.5 g; salt mix., 4; vitamins mix., 1 g and cellulose, 4 g.

Group 2 (Coffee):

Coffee was added to the negative control diet in the amount of 4.2 g/100 g, which was estimated as equivalent to about 10 cups (one cup = 150 ml) for man. This was calculated in proportion to body weight of rat.

Group 3 (Caffeine):

Caffeine was added as 123.3 mg/100 g negative control diet. The amount of caffeine added to the diet was equivalent to about 10 cups of coffee content. Caffeine content of coffee used in this study was determined by high pressure liquid chromatography (Madison *et al.*, 1976). It was found to be 2.96 g/100 g coffee.

Group 4 (Positive control or cholesterol):

to have a hypercholesterolemic diet, cholesterol (1 g/100 g diet), bile salt (0.2 g/100 g diet) and tallow (20 g/100 g diet) were added to the negative control diet.

Group 5 (Cholesterol + Coffee):

Coffee 4.2 g/100 g was added to the positive control diet.

Group 6 (Cholesterol + 123.3 mg caffeine/100 g diet):

caffeine was added to the positive control diet. The amount of caffeine added to the diet was equivalent to about 10 cups of coffee content.

Group 7 (Cholesterol + black tea):

Positive control diet was used. This group received 70 g BTWE/L solution which equivalent to 10 glasses of tea (one glass 150 ml) for man calculated in proportion to body weight of rat.

Group 8 (Cholesterol + 76.8 mg caffeine/100 g diet):

Caffeine was added to the positive control diet. The amount of caffeine added to the diet was equivalent to about 10 glasses of black tea (one glass 150 ml) content. Caffeine content of black tea (El-Arosa) used in this study was determined by high pressure liquid chromatography (Madison *et al.*, 1976).

All the groups of rats received the water except group 7 which received black tea water extract. At the end of four weeks, all fluids were withdrawn and the distilled water was given instead. After food was withheld for 14 hours, the rats were killed and blood was collected from the eye plexuses by a fine heparinized capillary glass tubes. After clotting, the blood was centrifuged at 1300 xg for ten minutes, and serum was then collected.

Chemical analysis:

Serum cholesterol, triglycerides, HDL-cholesterol and LDL-cholesterol were measured using biochemical assay kits (Stanbio Laboratory TX, USA), as previously described by Allain *et al.* (1974), Fossati and Prencipe (1982), Lopez-Virella *et al.* (1977) and Glatter (1984), respectively.

Statistical analysis

Data for serum lipids, food consumption, weight gain were analyzed by student's t-test and ANOVA using SPSS Package (1990).

RESULTS

The results shown in Table (1) indicate that the rats were of similar weights, at the beginning of the feeding period. The mean final bodies weights gain were not significantly different in all groups within exception of coffee group and coffee plus cholesterol group ($P \leq 0.05$). Group 5 (coffee plus cholesterol) decreased significantly in final body weight than the other groups ($P \leq 0.05$). In the same Table, we may noticed that the mean food intake was significantly decreased in negative control group and coffee group. Table (2) and Fig. (1) summarized the serum lipid levels of grouped rats. It could be pointed out that the serum cholesterol and triglycerides levels increased significantly in groups 5, 6 (Coffee plus cholesterol and caffeine 123.3 mg/100 g diet plus cholesterol) compared to the positive control.

Table 1: Mean weight, weight gain and food consumption in rats (mean±SD).

Group of rats	Initial body weight (g)	Final body weight (g)	Total weight gain (g)	Food consumption (g/day)
1 Negative control	128.00b ±1.32	194.50a ±2.50	66.50a ±1.32	13.20c ±0.27
2 Coffee	130.67a ±1.53	169.33b ±4.26	38.67b ±5.66	13.67c ±0.42
3 Caffeine (123.3 mg/100 g diet)	128.33ab ±0.65	197.70a ±4.25	69.07a ±4.31	14.90ab ±0.36
4 Positive control	130.33ab ±1.53	197.37a ±3.49	67.03a ±4.77	15.20a ±0.40
5 Coffee + cholesterol	130.67a ±1.16	162.83c ±3.96	32.17b ±2.97	14.40b ±0.36
6 Caffeine (123.3 mg/100 g diet) + cholesterol	128.50ab ±1.71	198.40a ±3.62	69.90a ±2.55	15.13a ±0.40
7 Black tea + cholesterol	128.00b ±1.50	196.70a ±3.37	68.70a ±1.93	14.77ab ±0.21
8 Caffeine (76.8 mg/100 g diet) + cholesterol	129.00ab ±0.87	195.50a ±3.78	66.67a ±4.51	14.73ab ±0.12

Means followed by the same letter(s) within the same column are not significantly different ($P \leq 0.05$).

There were no significant differences in group 8 (caffeine 76.8 mg/100 g diet plus cholesterol) and group 7 (black tea plus cholesterol) compared to the positive control ($P \leq 0.05$). HDL-cholesterol in the present study of groups 6, 8, 7 and 5 (caffeine 123.3 mg/100 g diet plus cholesterol, caffeine 76.8 mg/100 g diet plus cholesterol, black tea plus cholesterol and coffee plus cholesterol) respectively were significantly increased as compared to the positive control. LDL- cholesterol decreased significant in animals fed coffee plus cholesterol and animals fed caffeine 123.3 mg/100 g diet plus cholesterol, whereas there was no significant differences in group 7 (black tea plus cholesterol) and group 8 (caffeine 76.8 mg/100 g diet plus cholesterol) compared to the positive control. The ratios of HDL-cholesterol to total cholesterol and HDL-cholesterol to LDL-cholesterol were significantly lower ($P \leq 0.05$) in groups 5, 6 and 4 (coffee plus cholesterol, caffeine 123.3 mg/100 g diet plus cholesterol and positive control), respectively. However, it was noticed that the serum lipid levels of rats in group 2 (coffee) and group 3 (caffeine 123.3 mg/100 g diet) were not significantly different as compared to group 1 (negative control).

Table 2. Mean serum lipids in rats (mg/dL) (mean±SD).

Group of rats	Total cholesterol	Tri-glyceride	HDL-cholesterol	LDL-cholesterol	HDL/cholesterol	HDL/LDL cholesterol
1 Negative control	51.58c ±2.20	69.30c ±2.81	25.04d ±2.36	24.91c ±1.82	0.50a ±0.07	1.01a ±0.18
2 Coffee	52.80c ±3.52	70.81c ±2.57	25.17d ±1.71	24.55c ±2.12	0.48a ±0.04	1.02a ±0.16
3 Caffeine (123.3 mg/100 g diet)	55.50c ±4.69	71.06c ±4.45	27.48d ±2.23	25.41c ±1.94	0.51a ±0.00	1.09a ±0.17
4 Positive control	124.84b ±3.68	167.94b ±4.95	40.62c ±3.42	51.84b ±4.68	0.33b ±0.03	0.79b ±0.11
5 Coffee + cholesterol	184.74a ±3.53	213.17a ±4.73	50.80b ±4.42	81.85a ±7.25	0.30b ±0.03	0.68b ±0.12
6 Caffeine (123.3 mg/100 g diet) + cholesterol	186.89a ±2.76	209.90a ±7.30	56.93a ±3.72	83.13a ±6.31	0.32b ±0.02	0.69b ±0.03
7 Black tea + cholesterol	125.70b ±5.32b	168.39b ±7.28	52.41ab ±0.92	48.70b ±3.07	0.49a ±0.08	1.08a ±0.07
8 Caffeine (123.3 mg/100 g diet) + cholesterol	126.47 ±4.04	169.21b ±5.48	56.57a ±3.96	52.56b ±5.49	0.48a ±0.03	1.08a ±0.05

Means followed by the same letter(s) within the same column are not significantly different ($P \leq 0.05$).

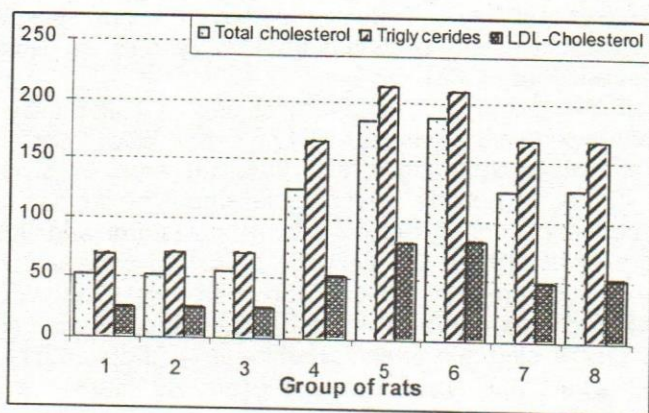


Fig. 1. Serum cholesterol, triglycerides and LDL-cholesterol (mg/dL) of rats given black tea, coffee and caffeine.

DISCUSSION

The present study showed that the addition of 10 cups of coffee or the amount of caffeine (equivalent to 10 cups of coffee content) to the negative control diet had no effect on serum lipids ($P < 0.05$). However, the lipoproteins profile of the control is in agreement with those examined before for rat (Abd El-Maksoud *et al.*, 1996; Bashandy *et al.*, 1996 and Yang *et al.*,

1999). Also, addition of 10 glasses of black tea or the amount of caffeine (equivalent to 10 glasses of black tea content) to the positive control diet (hypercholesterolemic) had no significant change in total cholesterol, triglycerides and LDL-cholesterol, whereas the ratios of HDL-cholesterol to total cholesterol and HDL-cholesterol to LDL-cholesterol were increased ($P < 0.05$). Animals which consumed coffee plus hypercholesterolemic diet or caffeine (123.3 mg/100 g diet) plus hypercholesterolemic diet had significant increasing in total cholesterol, triglycerides, HDL-cholesterol and LDL-cholesterol as compared to the positive control group. Our results were in agreement with those obtained by Rakicioglu *et al.* (1988). They observed that addition of coffee (equivalent to 12 cups of coffee per person) to the rats fed on diet included cholesterol (1 g/100 g diet) and cholic acid (0.2 g/100 g diet) had raised the levels of serum total cholesterol, triglycerides and VLDL-cholesterol ($P < 0.05$).

The same authors also revealed that coffee, tea and caffeine addition to the standard rat chow diet, not including atherogenic substances such as cholesterol, animal fat and bile salt as used in the present study, had no effect on the serum lipid parameters. It was also noted that increases in serum lipids were found to be related to the concentration of caffeine in the diet. As tea contains half the caffeine compared to the same amount of coffee, it is tempting to think that tea and decaffeinated coffee may cause very little or even no changes in the levels of plasma cholesterol. Green and Harari (1992) and Van-Het-Hof *et al.* (1997) indicated that there was non significant negative association between black tea consumption and serum cholesterol or serum lipids.

The amount of caffeine and composition of diet are the important factors for the effect of caffeine. Caffeine may have a suppressing effect on the release of VLDL-cholesterol and sterol from liver and cause accumulation in tissues (Hostmark *et al.*, 1988).

Many of the recent researches concluded that in healthy people, consumption of brewed coffee had no effect on serum lipids in balanced diet (Lancaster *et al.*, 1994; Wahrburg *et al.*, 1994; Sanguigni *et al.*, 1995 and Miyake *et al.*, 1999). In healthy adults, replacement of regular coffee by decaffeinated coffee has no effect on serum cholesterol and lipoproteins (Van-Dusseldore *et al.*, 1990). Consumption of 75 mg caffeine/day did not cause any change in serum lipids (Bak and Grobbee, 1991).

However, persons who had smoking habit and consumed coffee had clear effects on increasing serum cholesterol and lipoproteins (D'Avanzo *et al.*, 1993; Miyake *et al.*, 1999 and Christensen *et al.*, 2001).

Increases in serum lipids were greater in studies of patients with hyperlipidemia and in trials of caffeinated or boiled coffee (Jee *et al.*, 2001). Some studies suggested that besides caffeine there were other substances may be lipids materials in unfiltered coffee such as cafestol and Kahweol (Urgert and Katan, 1996; Van-Tol *et al.*, 1997; Urgert *et al.*, 1997 and de-Roos *et al.*, 2001).

Recently, number of studies indicated that drinking large quantities of filtered coffee 1 L/d (not contained any lipids, Roos *et al.*, 2001) had raised

plasma or serum lipids in humans (Urgert *et al.*, 2000 and Christensen *et al.*, 2001).

From the above discussion, it seems that the consumption of coffee, black tea and caffeine had no effect on serum lipids in rats fed on balanced diet that not contained any atherogenic substances such as cholesterol, bile salts and high animal fats. Whereas, the consumption of coffee or its equivalent caffeine with hypercholesterolemic diet were increased the serum lipids. Consumption of black tea or its equivalent caffeine with hypercholesterolemic diet had no effect on serum lipids.

This result indicated that consumption of high concentration of caffeine (found in coffee not in black tea) with hypercholesterolemic diet most probably increased the serum lipids in rats.

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تأثير الشاي الأسود والقهوة والكافيين على صورة دهون السيرم فى الفئران عزة أحمد بكرى ١ ، سمير محمد شلبى ٢ ١معهد بحوث تكنولوجيا الأغذية - مركز البحوث الزراعية - الجيزة - مصر. ٢كلية التربية النوعية - جامعة طنطا.

تم اختبار تأثير الشاي الأسود والقهوة والكافيين على دهون السيرم فى ثمانية مجاميع فئران أمهق عمرها ثمانية أسابيع ومتوسط وزن الفأر من ١٢٨-١٣١ جرام. وتم تغذية الفئران على وجبات محددة لمدة أربعة أسابيع وأحتوت الوجبات على الشاي الأسود أو القهوة أو الكافيين (١٢٣,٣ مجم كافيين / ١٠٠ جم وجبة أو ٧٦,٨ مجم كافيين/١٠٠ جم وجبة) وذلك بدون أو بإضافة الكوليسترول ١% واملاح الصفراء ٠,٢% و ٢٠% دهن ليه. ولوحظ انه ليس هناك تغيير معنوى فى متوسط الزيادة فى وزن الجسم والزيادة النهائية فى وزن الفئران لكل المجاميع ماعدا مجموعة القهوة ومجموعة القهوة + الكوليسترول حيث قل وزنها معنوياً.

كما لوحظ انه ليس هناك زيادة معنوية فى الكوليسترول الكلى والجليسيريدات الثلاثية والكوليسترول مرتفع الكثافة والكوليسترول منخفض الكثافة فى الفئران التى تتغذى على وجبة القهوة ووجبة الكافيين (١٢٣,٣/١٠٠ جم) وذلك بالمقارنة بالمجموعة الضابطة (بدون كوليسترول). بينما ارتفع معنوياً الكوليسترول الكلى والجليسيريدات الثلاثية والكوليسترول منخفض الكثافة فى مجموعة القهوة + كوليسترول ومجموعة الكافيين (١٢٣,٣ مجم/١٠٠ جم وجبة) بالمقارنة بالمجموعة الضابطة (عالية الكوليسترول). ووجد انه ليست هناك فروق معنوية فى مجموعة رقم ٧ (مجموعة الشاي الأسود + كوليسترول) ومجموعة رقم ٨ (٧٦,٨ مجم كافيين/١٠٠ جم وجبة) بالمقارنة بالمجموعة الضابطة (عالية الكوليسترول). وجدير بالذكر أن النسبة بين الكوليسترول مرتفع الكثافة إلى الكوليسترول الكلى والنسبة بين الكوليسترول مرتفع الكثافة إلى الكوليسترول منخفض الكثافة كان أقل معنوياً فى مجاميع رقم ٥ ، ٦ ، ٤ (القهوة + كوليسترول ، ١٢٣,٣ مجم كافيين/١٠٠ جم وجبة + كوليسترول والمجموعة الضابطة مرتفعة الكوليسترول) على التوالى.

ويمكن أن نستنتج من نتائج البحث أن الشاي الأسود والقهوة والكافيين لاتزيد دهون السيرم فى الفئران التى تتغذى على وجبة متوازنة بينما القهوة والتركيز العالى من الكافيين يودى إلى رفع لييدات السيرم فى الفئران التى تتغذى على وجبة عالية فى الدهن وعالية فى الكوليسترول. الشاي الأسود ليس له تأثير على لييدات السيرم فى الفئران التى تتغذى على وجبة عادية أو وجبة عالية الكوليسترول.