Original Article

A Comparative Study of the Effect Coffee and Tea on Some Parameters in the Rats Blood

Sadiq Al-Mansury, Suhad J. Hadi, Hawraa H. Naji, Adnan M. Jassim, Shatha M. Abbas, Nada Khazal K. Hindi[1]

Department of Physiology, Biochemistry and Pharmacology, Veterinary Medicine College, Al-Qasim Green University, Babylon, [1]Department of Basic and Medical Science, Nursing College, Babylon University, Pharmacy College, Al-Mustaqbal University, Hillah, Iraq

Abstract

Background: Tea and coffee have been consumed since decade ago and became a significant part of social life and cultural traditions. **Objective:** The study aims to evaluate the effects of coffee and black tea extract on some blood factors in adult albino rats. **Materials and Methods:** Eighteen adult male albino rats, classified into three groups of six: group 1 (control) received orally drinking water only, group 2 (tea) received orally extract tea, group 3 (coffee) treated orally with coffee. Coffee and tea extracts were prepared daily at a dose of 30 g/L and provided at all times to the rats for 30 day as drinking water. Iron and ferritin levels, cholesterol concentration of low-density lipoprotein (LDL) and high-density lipoprotein (HDL), superoxide dismutase (SOD), glutathione peroxidase (GPx) activities as well as the levels of tumor necrosis factor-alpha (TNF-α), and interleukin-6 (IL-6) were compared among these groups. **Results:** This study inferred, there was a significant decrease the amount of iron in the blood as well as the ferritin. Also consumption of coffee or tea lowers LDL cholesterol and HDL cholesterol and enhanced antioxidant activity by SOD and GPx enzymes more than control group. Furthermore, increased these levels in proinflammatory cytokines (IL-6 and TNF-α) in treated rats with tea and coffee extract more than their levels in the control group. **Conclusion:** The extracts of tea and coffee lowered the iron and ferritin concentration, have remarkable antioxidant activity, and increased the levels in proinflammatory cytokines (IL-6 and TNF-α) in the rat blood.

Keywords: Antioxidant activity, ferritin, superoxide dismutase, tea extract

Introduction

Coffee and tea are two of the most common hot drinks worldwide and contain significant amounts of caffeine, that leads to caffeine which is the most commonly consumed psychoactive factor.[1] A diversity of plants involves caffeine in their fruits, leaves, and seed. Also, baring tea and coffee, these plants include yerba matte leaves, cocoa beans and guarana berries. Caffeine can be manufactured and added to beverages and foods, including energy drinks, soft drinks, tablets and energy shots that are marketed to decrease tiredness,[2] whereas the increased consumption of energy drinks seriously harms the organs of body.[3] Caffeine is also widely used as a medicine for apnea of prematurity in infants,[4] caffeine used with analgesic factors are used together as pain relievers.[5] A gathering of adenosine in the brain prevents awakening and increases sleepiness. In modest doses (50-310 mg), caffeine can resist the influences of adenosine and decrease fatigue.[6]

According to recent studies in theUSA, about 85% of adults consume caffeine daily, and the average caffeine intake is estimated at 135 mg per day, which is equal to 1.5 standard cups of coffee or tea (standard cup set at 8 fluid ounces and equal to [235 mL]). Tea and soft drinks are more significant sources of caffeine ingested by teens, while coffee is the most source of caffeine intake by adults.[7]

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Address for correspondence: Dr. Sadiq Al-Mansury, Department of Physiology Biochemistry and Pharmacology, Veterinary Medicine College, Al-Qasim Green University, Babylon, Iraq. E-mail: sadiqaljubory1@gmail.com

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Iron has two known oxidation states: called ferric (Fe^{+3}) and ferrous (Fe^{+2}). These states help iron to participate in redox reactions which are necessary for energy metabolism by donating or accepting electrons. Moreover, free iron stimulates oxidative reactions and inhibition of free radicals.[8] Iron is also found in the blood to contribute to hemoglobin formation, a protein found in red cells that carry oxygen to body tissues whereas iron is included in the composition of myoglobin, which transports oxygen to muscle tissue. Excess iron can be saved as a reserve in the form of ferritin.[9] Iron deficiency anemia is one of the most common foods deficiencies in the world, according to the World Health Organization.[10] The first part of iron deficiency is described by the lack of stored iron (referred by ferritin) and it is known as iron depletion or the beginning of iron deficiency. Blood iron and the iron bearing serum protein concentration are normal at this case. When iron stocks are depleted (serum ferritin less than 12 µg/L), serum iron reduces and blood transferrin increases, which is generally measured by total iron-binding capacity.[11]

The overload of iron is related with an excess of non-protein bound iron result from the physiological ability to bind to iron. Damages of overloading are, for example, an increased risk of cardiomyopathy and bacterial infection. Overload can also product by more absorption of dietary iron for diverse reasons including chronic intake of more than sufficient quantity of dietary iron, particularly heme-iron.[12]

The aim of work was to study the effect of tea or coffee intake of tea or coffee with iron deficiency in a sample of albino rat's blood. In addition, to study the relationship between tea and coffee consumption with the concentration of high-density lipoprotein (HDL) and low-density lipoprotein (LDL) cholesterol (superoxide dismutase [SOD], glutathione peroxidase [GPx]) and (interleukin-6 [IL-6], tumor necrosis factor-alpha [TNF-α]) in a blood. Also, to determine which one (tea or coffee) is less effects and better for human consumption.

Materials and Methods

Chemicals

This study was conducted from December 2022 to January 2023 in the Physiology, Pharmacology and Chemistry Branch of the College of Veterinary Medicine, Al-Qasim Green University. Rats were purchased from Al-Daman drug repository in Al-Hilla city, in addition to the basic materials for the experiment, tea (apple tea) and coffee (Brazilian coffee) purchased from market. The other materials and kits were provided by the biochemistry laboratory of the above administration. They were given a standard diet, unlimited access to water (tea or coffee that had been extracted), housing that was kept at a typical humidity and temperature (25°C), exposure to 12 h of light and 12 h of darkness, and feeding on a standard diet.

Animals

Eighteen adult male rats, weighing between 190 and 200 g were purchased from Al-Daman drug repository in Babylon province. They were exposed passive preliminaries over a period of 14 days in order to cope themselves with the new environment. The black tea was prepared by dissolved 8 g tea in 250 mL of water. The mixture was left to infuse for 10 min then filtered and given to the animals. The coffee was prepared by adding 8 g powder to 250 mL boiling water.

Experimental design

Three equal major groups of rats were randomly and similarly divided; group I (also known as the control group) was not given any remedy whereas, group II was given tea orally at a dosage of 30 g/L. Group III: given a dosage of 30 g/L of coffee orally. Hot water was utilized for boiling the tea and coffee. After the end of the thirty-first day of the experiment, and under diethyl ether anesthesia, 5 mL of blood was drawn from each rat's heart. Test containers were used to hold the blood specimens. After centrifuging the blood samples at 3000 rpm for 5 min, they were allowed to remain at room temperature for 15 min.

Determination serum iron

Deionized water 0.5 mL was added to 5 mL test tube to make a blank tube. The standard tube: refill 5 mL EP tube with 0.5 mL of 2 mg/L iron standard working solution. Sample container: refill 5 mL EP tube with 0.5 mL of the specimen to be tested. Add 1.5 mL of the iron chromogenic an agent, mix well with a vortex blender, and then incubate for 5 min in a 100°C water bath. (Standard tube and blank tubes can be handled lacking a water bath at 100°C.) Tubes were centrifuged at 2300 g for 10 min after cooling them under water that was flowing. Optical density (OD) value was calculated of every tube by a spectrophotometer at 520 nm with a 0.5 cm light path quartz cuvette after calibrating it with water that has been deionized.

Determination of serum ferritin, cholesterol low-density lipoprotein, high-density lipoprotein, superoxide dismutase, glutathione peroxidase, tumor necrosis factor-alpha, and interleukin-6

This ELISA reagent utilizes the Sandwich-ELISA technique, kits supplied from Sunlong Biotech Co., Ltd. (Hangzhou, China).

Ethical approval

This research has been approved by the Ethics Commission of Veterinary Medicine according NO: 533FD2. Also, the procedure was approved by the institutional animal ethics council at Al-Qasim Green University.

Statistical analysis

Data were expressed as mean ± standard error (SE). Data were analyzed on a Statistical Package for the Social Sciences (SPSS, IBM Company, Chicago, IL, USA)

Table 1: The effect of tea and coffee extracts (30 g/L of water; oral) on iron, ferritin and some biochemical parameters in blood of male albino rats after 30 day of treatment

Parameters	Control	Tea	Coffee	LSD
Iron	252.7 ± 4.4 A	217.2 ± 7.21 B	206.6 ± 5.24 B	22.2
Ferritin	3.18 ± 0.32 A	1.92 ± 0.086 B	1.80 ± 0.19 B	0.76
HDL	1.4 ± 0.031 A	0.74 ± 0.092 B	1.2 ± 0.15 A	0.23
LDL	1.24 ± 0.037 A	0.86 ± 0.066 B	1.18 ± 0.066 A	0.22
SOD (IU/L)	10.33 ± 0.57 C	13.33 ± 2.08 A	11.33 ± 0.57 B	1.05
GPx (IU/L)	25 ± 0.59 B	28.3 ± 1.20 A	25.6 ± 0.88 B	1.34
TNF-α (pg/L)	0.20 ± 0.038 B	0.133 ± 0.08 A	0.27 ± 0.031 C	0.09
IL-6 (ng/L)	0.26 ± 0.025 A	0.31 ± 0.035 A	0.46 ± 0.059 B	0.11

LDL: low-density lipoprotein, HDL: high-density lipoprotein, SOD: superoxide dismutase, GPx: Glutathione Peroxidase, TNF-α: tumor necrosis factor-alpha, IL-6: interleukin-6

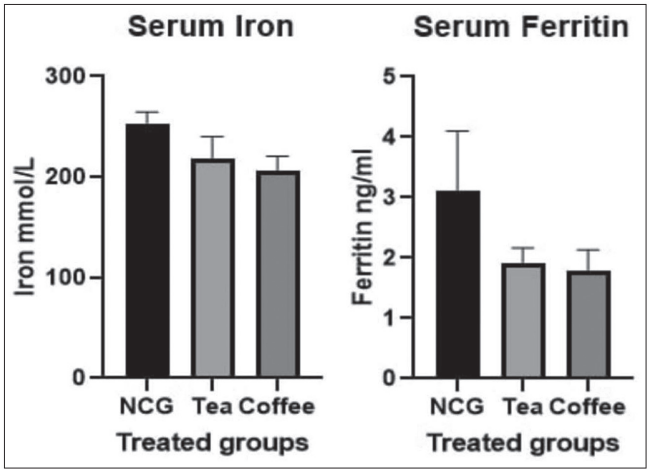

Figure 1: The effect of coffee and tea extract on serum iron and ferritin concentrations in treated rats. The first column is the control group (NCG), the second column is tea-treated and the third column is coffee-treated

software version 14.0 using one-way Analysis of Variance (ANOVA) and Student *t* test. *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$ were used as the criterion for significance.

Results

The current study showed that data of iron, ferritin levels appear a significant reduction of serum in both groups to recorded mean value 217.2 ± 7.21, 1.92 ± 0.086 for tea while 206.6 ± 5.24 and 1.80 ± 0.19 for coffee. Serum LDL value in Table 1 was a significant reduction in rats consumed tea to recorded 0.86 ± 0.066 as compared with coffee and negative control group, while HDL appear clear improvement in animal consumed coffee to recorded man value mimic to negative control group with no a significant between them (1.2 ± 0.15).

Discussion

A caffeoyl-quinic acid, or chlorogenic acid is main phenolic compound in coffee, that is, a phenolic acid.[13] whereas, herb teas have such peppermint, and include mainly the monomeric flavonoids. Tea flavonoids also include gallic acid esters and during fermentation to black tea complex polymers are formed containing both gallic acid esters and flavonoids.[13,14]

The obtained results were displayed in Table 1 and Figure 1, exhibited the tea consumption significantly lowered the iron concentration in blood by 217.2 ± 7.21 µmol/L compared with control 252.7 ± 4.4 A µmol/L. While coffee consumption significantly decreases the iron concentration in blood by 206.6 ± 5.24 C µmol/L, compared with control 252.7 ± 4.4 A µmol/L after 30 day of treatment.

Also this study revealed the concentration of ferritin decreased 1.92 ± 0.086 ng/mL of tea and 1.80 ± 0.19 of coffee compared with control 3.18 ± 0.32 ng/mL at the same duration. The results were explained on the basis that some of the natural compounds found in coffee or tea interfere with iron and prevent the absorption of iron in the small intestine. By forming an insoluble compound with iron, which leads to iron with animal waste. Then this causes some heart disease when the iron level in the body is imbalanced.[15,16]

The present study also estimated the relation between coffee and tea consuming and minimized in serum lipid concentration. The results propose that coffee or tea significantly decrease both HDL and LDL cholesterol level.[17]

In addition, the results are shown in Table 1 and Figure 2, exhibited the tea consumption significantly lowered the LDL concentration in serum by 0.86 ± 0.066. While coffee consumption significantly decrease the LDL concentration in serum by 1.18 ± 0.066 mmol/L, compared with control group 1.24 ± 0.037 A mmol/L after 30 day of treatment.

Also this study showed the concentration of HDL decreased 0.74 ± 0.092 of tea group and 1.2 ± 0.15 of coffee group compared with control group 1.4 ± 0.031 A at the same duration. In addition to the above, the concentration of LDL and HDL cholesterol in the coffee group is higher than its concentration in the tea group; because of the coffee is a result of grinding coffee beans that contain a percentage of fat compared to the extracted tea which produced from fat-free leaves.

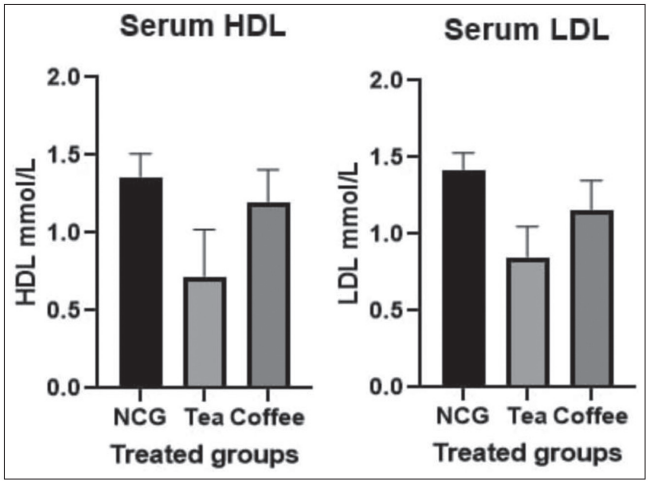

Figure 2: The relationship between tea and coffee consumption and the concentration of LDH and LDL in the serum of treated rats

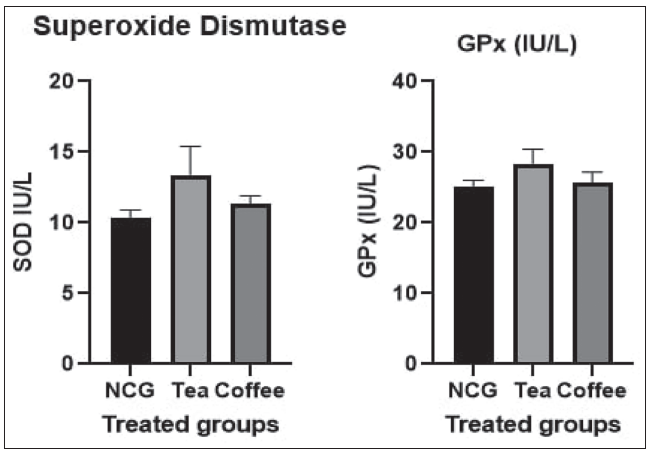

Figure 3: The effect of coffee and tea extracts on GPx (IU/L) and SOD (IU/L) enzymes levels in the serum of treated rats

Recent mechanistic studies that investigated at how drinking tea impacted cholesterol regulation provide additional support that these results are biologically plausible. According to our findings, multiple animal studies showed that adding tea to high-fat diet-induced rats' diets greatly improved their elevated cholesterol levels situation, including reducing HDL and LDL.[17]

In addition, a recent animal research showed that tea catechins could considerably reduce atherosclerosis as well as the accumulation of liver fat and raise HDL cholesterol in hyperlipidemic rats fed a diet high in fat and cholesterol.[17,18] Tea catechins are potent antioxidants capacity that mitigate LDL oxidation by attach their own into LDL particles in non-conjugated forms *in vitro*.[18] In a dose-dependent away tea consumption lead to up regulation in Hep G2 cells via regulating the SREBP-1 (sterol regulatory binding protein-1) pathway.[15,19]

High dose of caffeine had stronger anti-obesity effects than extract caffeine, which can lower body weight and lipid as

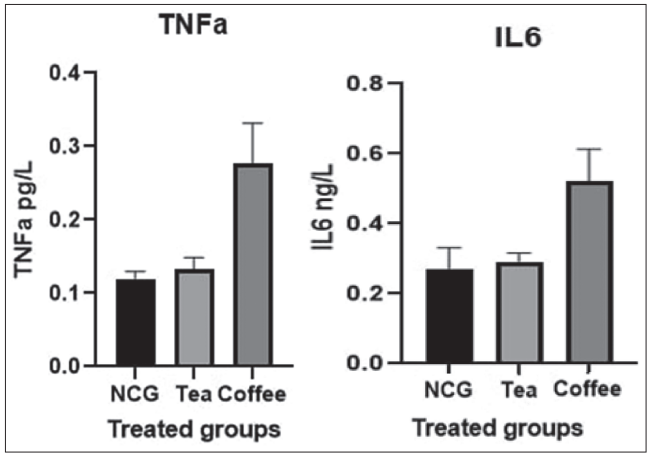

Figure 4: Effect of coffee and tea extract on TNF-α and IL-6 cytokines levels in serum of treated rats

well as inhibition of synthesis and the up-regulation of b-oxidation of fatty acids in the liver.[20] Tea can affect iron absorption in intestine by forming insoluble iron tenants.[21] In fact black tea is the main inhibitor of iron absorption in Westerners, coffee intake is associated with low serum ferritin levels in premenopausal women.[22]

Antioxidant and anti-inflammatory parameters appear that tea shows significant increase of both GPx and SOD as compared with rats consumed coffee as shown in Table 1 and Figure 3, as well as negative control that due to potent scavenger activity of tea to remove some oxidative stress due to inducer some important cytochrome in hepatic cell special CYP3A4 and CYP3A3.[23] The recent study was confirmed that black and green tea has polyphenol content and antioxidant activity.[24] The present study agrees with[25] that regarding to polyphenol content and antioxidant potency between coffee and tea, different compounds such as gallic and ellagic acids, quercetin 3-O-glucoside, hyperoside, rutin, kaempferol 3-O-glucoside, catechin, epicatechin, vitexin, and epigallocatechin gallate were detected in teas than coffee. Reactive species composed of oxygen and nitrogen can be important in normal cell metabolism by assisting as signaling molecules that control gene expression, apoptosis and cell division. Nitric oxide (•NO) plays a vital role for the heart, blood vessels and nervous systems as a neurotransmitter and regulator of vascular tone, platelet activation, leukocyte adhesion and myocardial contractility that done by presence of superoxide anion (•O2−) and hydroxyl (•OH) radicals. Also, nitric oxide is critical for the immune system as part of macrophages' microbicidal mechanisms. In the presented experiments, coffees typically showed weak to modest antioxidant activity.[26]

The obtained results disagree with a single research paper[27] which indicated that coffee possessed greater antioxidant potential than wines and tea that return to phenolic acids and Coumarins.[28] Many published articles showed that

flavanols as the major carriers of antioxidant activity in tea. However, it is important to consider the contribution and synergistic effect of other substances, particularly those from the families of flavanols and phenolic acids.

On the other aspect, this study confirmed an enhancement of proinflammatory cytokines (IL-6 and TNF-α) in the coffee-treated rats (0.37 and 0.276 ng/L) higher than their tea-treated animals (0.32 and 0.135 ng/L), and this is higher than their levels in the control group animals (0.123 and 0.12 ng/L) as shown in Table 1 and Figure 4. The current study was compatible with Carito *et al*.[29] who noted men who consumed more than two hundred milliliters of coffee per day had 52% higher levels of IL-6, 31% higher levels of C-reactive protein, 15% higher levels of serum amyloid-A, 25% higher levels of TNF-α, and 5% higher levels of white blood cells compared to men who did not drink coffee (all $P = 0.05$).

The current study disagrees with a number of studies, noted that tea contain sufficient quantity of polyphenols' has ability to prevent the production of proinflammatory cytokines such as interferon (IFN), chemokines and TNF- in different cell types is what mainly impacts the course of inflammation.[29,30] Polyphenols have anti-inflammatory activity on numerous levels, primarily by blocking NF-B, controlling mitogen-activated protein kinase, inducible nitric oxide synthase, cyclooxygenase-2, arachidonic acid, and lipoxygenase in addition, decreasing reactive oxygen species (ROS) synthesis in comparison to reactive nitrogen species.[31] Many studies[32,33] confirmed that tea flowers may be used as a functional natural diet, as well as antibacterial and anti-inflammatory effect on immunological inflammation and acute *in vivo*.

Conclusions

In conclusion, the iron and ferritin levels in animal of tea group were decreased by increasing the consumption of black tea extract compared to the control group. This study also showed the levels of iron and ferritin in serum of coffee group were less than the tea group and this group was lower compared to the control group.

The results refer that consumption tea and coffee lower the levels of LDL and HDL in the rat's blood. While, the consumption tea decreasing these levels of (LDL and HDL) were more than the coffee group at same conditions. In other side, the antioxidant activities by SOD and GPx enzyme of tea and coffee more than control group. Furthermore, increased the levels in proinflammatory cytokines (IL-6, and TNF-α) in treated rats with tea and coffee extract more than their levels in the control group.

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Conflicts of interest

They have no conflicts of interest.

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