

Black and green tea improves lipid profile and lipid peroxidation parameters in Wistar rats fed a high-cholesterol diet

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Abstract In the present study, the efficacy of black tea (BT) and green tea (GT) was studied in relation to serum and hepatic oxidative abnormalities in hypercholesterolemic rats. Hypercholesterolemia was induced in male Wistar rats (8 week old) by feeding them with a high-cholesterol diet (HCD) for 35 days. The experimental rats were given BT and GT as a supplement (7 g/L) via drinking water. Increased hepatic and serum lipid profile along with abnormalities in oxidative marker, with a concomitant increase in the body weight, food intake, and food efficiency, were seen in hypercholesterolemic rats. Following the supplementation of BT and GT to rats fed with HCD, significantly lower levels of serum and hepatic cholesterol, triglycerides, serum low-density lipoprotein cholesterol, and increased high-density lipoprotein cholesterol levels were observed, when compared with hypercholesterolemic group.

Further, significantly lower levels in the serum and hepatic lipid peroxidation, body weight gain, and food efficiency were observed in BT and GT group when compared with control and HCD fed group. However, no such significant changes were observed in the food intake upon supplementation with BT and GT. These results suggest that supplementation of BT and GT may protect against the serum and hepatic abnormalities in hypercholesterolemic rats.

Keywords Hypercholesterolemia · Black tea · Green tea · Oxidative stress

Abbreviations

NR	Normal
HCD	High-cholesterol diet
BT	Black tea
GT	Green tea
LDL	Low-density lipoprotein
VLDL	Very low-density lipoprotein
HDL	High-density lipoprotein
LPO	Lipid peroxidation
CVD	Cardiovascular disease
MDA	Malondialdehyde
CV	Coefficients of variation

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Introduction

Hypercholesterolemia is one of the most important risk factors for atherosclerosis and subsequent cardio-

vascular disease (CVD) [46]. CVD is the leading cause of cardiovascular morbidity and mortality worldwide, and two thirds of all cardiovascular fatalities occur in developing countries [33]. It is expected that CVD will be the largest cause of disease burden worldwide by 2020 [53, 54]. Hypercholesterolemia causes malfunctioning of the liver, which apparently occurs through microvesicular stenosis due to the intracellular accumulation of lipids [3]. Cholesterol-rich diet causes hypercholesterolemia, increases free radical production, and thereby elevates lipid peroxides [7, 22].

Hypercholesterolemia is associated with increased production of oxygen radicals and increased oxidation of LDL cholesterol [10, 32]. This leads to endothelial injury and tissue injury by inducing oxidative modifications on lipid, protein, and DNA. Because of their intrinsic potential for free radical generation, erythrocytes might be a very suitable environment for cholesterol to exert its prooxidant action [26]. Oxidative stress, a state resulting from disruption of the delicate balance between oxidative and antioxidative processes, is believed to play an important role in the pathogenesis of hypercholesterolemic atherogenesis [45]. Thus, feeding experimental animals with cholesterol has often been used to elevate serum or tissue cholesterol levels to study the etiology of hypercholesterolemia-related metabolic disturbances [6].

Recently, there has been renewed interest in finding naturally occurring antioxidants for use in foods, cosmetics, or medicinal materials to replace synthetic antioxidants, which are being restricted due to their carcinogenicity [40]. The antioxidative phytochemicals, especially phenolic compounds found in vegetables, fruits, and medicinal plants, have received increasing attention for their potential role in prevention of human diseases [8, 37, 41]. Many medicinal plants contain large amounts of antioxidants, such as polyphenol, which play an important role in abstracting and neutralizing free radicals, quenching singlet and triplet oxygen, or in decomposing peroxides [2]. Grape seed polyphenols, proanthocyanidins, have a hypocholesterolemic effect on rats fed a high-cholesterol diet [49]. Supplementation of drinking water with dealcoholized wine, pomegranate juice, and quercetin reduced the size of these lesions in apolipoprotein E-deficient mice [23, 25], and these effects are associated with reduced uptake of LDL

cholesterol by macrophages and decreased susceptibility of LDL to aggregation.

Tea can be categorized into three types, depending on the level of fermentation, i.e., green (unfermented), oolong (partially fermented), and black (fermented) tea. Interestingly, the use of tea extracts as dietary supplements arises from the perception that some tea compounds have beneficial protective effects against chronic diseases [44]. The presence of polyphenols in tea may contribute to its antioxidant effect by inhibiting ROS-generating enzymes [38]. In general, green tea has been found to be superior to black tea in terms of antioxidant activity, owing to the higher content of epigallocatechin, epigallocatechin gallate. Catechins, which are tea flavonols, are well-known natural polyphenolic antioxidants, and approximately 80% of green tea consists of flavonoids [19, 38]. In contrast, the black tea (BT) contains considerable amount of the flavones and thearubigins, the oxidation products of quinones and flavols [30, 31]. Green tea (GT) polyphenols have shown strong chemopreventive and chemotherapeutic effects against various pathological conditions [9, 16, 28].

Although black tea is the most widely consumed beverage in Saudi Arabia, recently, green tea has received much attention as a protective agent against chronic diseases [27, 36, 37, 42]. Hence, the present study was designed to evaluate the effect of black or green tea consumption on lipid status and lipid peroxidation in Wistar rats fed a high-cholesterol diet.

Materials and methods

Chemicals

HIGH-pressure liquid chromatography (HPLC) standards [(−)-epicatechin (EC), (−)-epigallocatechin (EGC), (−)-epigallocatechin gallate (EGCG), (−)-epicatechin gallate (ECG), gallic acid (GA), and caffeine (CA)] were all purchased from Sigma Chemical Co. (St Louis, MO, USA). Commercially available green tea and black tea samples were purchased from Lipton outlet retailers, Riyadh, Kingdom of Saudi Arabia. Samples were stored at room temperature in a dark place. All the other chemicals and solvents used were of analytical grade.

Determination of gallic acid, catechins, and caffeine of green and black tea compounds by HPLC

Tea preparation

A strong brew of black tea was prepared following the instructions provided on the package by pouring 250 mL boiling water into a glass flask and dipping a tea bag for 3 min. A second, stronger brew of green tea was prepared by pouring 250 mL boiling water over 0.5 g tea for 10 min. After cooling, samples were mixed and filtered through a 0.45- μ m filter.

Standard preparation

Standards of five catechins (EC, EGC, ECG, and EGCG), GA, and CA standard were prepared by dissolving in a small volume of 5% (v/v) acetonitrile containing 0.05% (v/v) phosphoric acid (85%). The standards were finally filtered through a 0.45- μ m filter.

Chromatographic conditions

HPLC analysis was performed following the method of Goto et al. [18] in a Shimadzu model LC-10 (Shimadzu co, Tokyo, Japan), equipped with a binary pump, an autosampler, and photodiode array detector (PDA). The standard and samples were injected (10 μ L) into a reversed-phase C₁₈ column. All the solvents were filtered with 0.45- μ m filter. The following gradient elution was carried out: mobile phase A, water–acetonitrile–phosphoric acid (85%) (95.45:4.5:0.05, v/v/v); mobile phase B, water–acetonitrile–phosphoric acid (85%) (49.95:50.0:0.05, v/v/v). The mobile phase composition started at 90% mobile phase A and 10% mobile phase B, and was maintained for 12 min and then linearly increased to 15% mobile phase B during 12–18 min. This condition was maintained for 2 min followed by a linear increase of mobile phase B to 70% during 20–30 min. The final conditions were held for an additional 10 min. The mobile phase flow rate was 1.0 mL/min, and the temperature of the column oven was set at 35°C. The quantification of the gallic acid, catechin, and caffeine compounds by PDA was performed at 278 nm. Identification of compounds was carried out by comparing retention times and UV spectra of the unknown peaks to those of the standards.

Animal experiment

Male Wistar rats weighing approximately 150–200 g (2 month old) were housed under conditions of controlled temperature ($25\pm2^\circ\text{C}$) with a 12 h/12 h day–night cycle, during which time they had free access to food and water ad libitum. Animals were maintained per national guidelines and protocols approved by the graduate college animal care committee at Kind Saud University.

The rats were divided into six groups of seven rats each. Group I rats served as vehicle control (normal diet). The normal diet consisted of 21.2% protein, 4.68% fat, 2.95% fiber (w/w), and 55% carbohydrate with adequate mineral and vitamin contents. Group II rats were fed a high-cholesterol diet (HCD) containing normal rat chow, supplemented with 1.5% cholesterol and 0.1% cholic acid for 35 days. Group III rats were fed a normal diet and supplemented with black tea (7 g/L) in 30 g/L sucrose via drinking water for 35 days. Group IV rats were fed HCD and supplemented with black tea (7 g/L) in 30 g/L sucrose via drinking water for 35 days. Group V rats were fed a normal diet and supplemented with green tea (7 g/L) in 30 g/L sucrose via drinking water for 35 days. Group VI rats were fed HCD and supplemented with green tea (7 g/L) in 30 g/L sucrose via drinking water for 35 days. The control group consumed 30 g/L sucrose. Sucrose was used to mask the bitterness of the tea. Drinking water containing the tea was prepared and replaced every evening. The amount of food ingested and fluid intake was measured daily in the experimental groups.

At the end of the experimental period, all the rats were killed by cervical decapitation. From each animal, blood samples were collected, and the hepatic tissue was dissected out and then washed thoroughly in physiological saline. The serum was separated from the blood, and the serum and hepatic tissue samples were stored at -80°C until analysis.

Prior to biochemical analysis, hepatic tissue (100 mg/mL) was homogenized in 50 mM phosphate buffer (pH 7.2); the homogenate was then centrifuged at $8,000\times g$ for 15 min at 4°C , and the supernatant was used for biochemical analysis.

Biochemical estimations

Lipids were extracted from the hepatic tissue according to the method of Folch et al. [12] using

chloroform–methanol mixture 2:1 v/v. After extraction of lipids, aliquots were used for the estimation of cholesterol and triglycerides. Total serum and liver cholesterol and serum high-density lipoprotein cholesterol (HDL-C) were determined by enzymatic colorimetric procedure of Richmond [39] (Randox cholesterol enzymatic kit No. 290 for cholesterol, and Randox HDL cholesterol precipitant kit No. 204 for HDL-C; Crumlin Co., Antrim, UK). Low-density lipoprotein cholesterol (LDL-C) was obtained by Friedewald formula [14]. Serum and liver total triglycerides were quantified by an enzymatic colorimetric procedure of Trinder [50] (Randox, triglycerides enzymatic kit No. TR 210, Crumlin Co., Antrim, UK).

Briefly, each serum sample 10 (μL) was pipetted into 1 mL of reagent in a test tube. A standard solution was prepared by mixing 10 μL of standard with 1.0 mL of reagent. A blank was prepared by adding 10 μL of distilled water into 1.0 mL of reagent. These serum, standard, and blank samples were then mixed and incubated for 10 min at 25°C. The absorbance of the blank reagent was subtracted from that of each sample and that of the standard. The concentrations of cholesterol/triglycerides in serum and liver samples were calculated from the corrected absorbance of both the sample and standard according to the manufacturer's recommendation. In the measurement of HDL-C, precipitations were prepared according to the manufacturer's Randox kit manual. A macro-precipitation was prepared by mixing of 0.5 mL of sample/standard with 1.0 mL of precipitant. Thereafter, a micro-precipitation was prepared by mixing of 0.2 mL of sample/standard with 0.5 mL of diluted precipitant. These tubes were mixed and allowed to sit for 10 min at room temperature. Then, tubes were centrifuged at 3,500 $\times g$ for 10 min. The clear supernatant was collected within 2 h for cholesterol determination as described above. In contrast, LDL-C was obtained by the Friedewald formula [14]. i.e., $\text{LDL-C} = \text{total cholesterol} - (\text{triglycerides}/5) - \text{HDL-C}$. The units were expressed as mg/dL.

Assessment of lipid peroxidation

Lipid peroxidation in the hepatic tissue and serum samples were determined by the procedure of Hogberg et al. [24]. Briefly, to 0.2 mL of 8.1% sodium dodecyl

sulfate, 1.5 mL of 20% acetic acid (pH 3.5) and 1.5 mL of 0.81% thiobarbituric acid aqueous solution were added in succession. To this reaction mixture, 0.2 mL of the homogenate of hepatic tissue/serum was added. The mixture was then heated in boiling water bath for 30 min. Malondialdehyde (MDA), formed as an end product of peroxidation of lipids, served as an index of the intensity of oxidative stress. MDA reacts with thiobarbituric acid to generate a colored product which absorbs at 532 nm. The ferrous sulfate and ascorbate induced lipid peroxidation system contained 10 mM ferrous sulfate and 0.2 mM ascorbate as inducers [11]. Tetramethoxypropane was used as an external standard. The level of lipid peroxides was expressed as millimoles of MDA formed per liter per milligram protein.

Statistical analysis

The values are expressed as mean \pm standard deviation (SD) for seven animals in each group. Differences between groups were assessed by one-way analysis of variance using SPSS software package for Windows. Post hoc testing was performed for intergroup comparisons using the least significance difference test, and the Student's *t* test was applied wherever relevant. Values of $p < 0.001$, 0.01, and 0.05 have been denoted by distinct symbols in the tables and figures.

The percentage coefficients of variation (CV%) for lipid profiles in serum samples were calculated using the equation.

$$\text{CV (\%)} = \frac{\text{Standard deviation} \times 100}{\text{Mean}}$$

Results

Green and black teas were analyzed for the content of individual catechins, gallic acid, and caffeine by using the HPLC method. The results obtained are presented in Table 1. Our results have shown large amounts of catechins (EC, ECG, EGC, and EGCG) in green tea when compared with black tea, whereas the gallic acid and caffeine contents are significantly ($p < 0.001$) lower in green tea than black tea. This compound was then tested for its *in vivo* study.

Table 1 HPLC analysis of the resulting compounds of gallic acid, catechins, and caffeine in green and black teas

S. no.	Components (g/kg)	Green tea	Black tea
1	Gallic acid (GA)	1.21±0.01	3.10±0.04*
2	Epicatechin (EC)	2.15±0.02	2.04±0.02**
3	Epicatechin gallate (ECG)	2.10±0.01	0.046±0.01*
4	Epigallocatechin (EGC)	2.42±0.01	2.15±0.01**
5	Epigallocatechin gallate (EGCG)	1.89±0.01	0.049±0.01*
6	Caffeine (CA)	2.24±0.01	2.77±0.01*

Values are expressed as mean ± SD ($n=3$). Statistical analysis was performed by the Student t test

* $p<0.001$

** $p<0.01$

The role of black and green tea in countering the lipidemic–oxidative aberrations accompanying diet-induced hypercholesterolemia has been investigated here. The levels of serum cholesterol, triglycerides, and LDL cholesterol values were significantly ($p<0.001$) higher, and HDL cholesterol was lower in rats fed the high-cholesterol diet (group II) than those in rats fed with a normal diet (group I) (Table. 2).

However, groups III and V rats (normal diet consuming with black and green tea) showed a significant ($p<0.001$) decrease in serum cholesterol and LDL cholesterol; group V rats showed a significant ($p<0.01$) decrease in triglycerides values and a significant ($p<0.01$) increase in the level of HDL cholesterol (group V), when compared with values in group I rats. Group IV (HCD consuming

Table 2 Effect of consuming black and green tea on lipid profiles in serum samples of HCD-fed groups compared with the control animals

Parameters	Group I—NR	Group II—HCD	Group III—NR + BT	Group IV—HCD + BT	Group V—NR + GT	Group VI—HCD + GT
Cholesterol (mg/dL)	100±5	122±9 ^a	87±4 ^a	93±2 ^{b,c}	79±3 ^{a,d}	80±5 ^{a,c,e}
CV (%)	5	7	5	2	4	6
Triglycerides (mg/dL)	100±9	141±6 ^a	104±15	126±11 ^b	71±9 ^{b,f}	64±4 ^{a,c,e}
CV (%)	9	4	15	9	13	6
LDL cholesterol (mg/dL)	100±5	130±7 ^a	61±3 ^a	66±2 ^{a,c}	43±3 ^{a,d}	51±2 ^{a,c,g}
CV (%)	5	6	5	3	7	4
HDL cholesterol (mg/dL)	100±2	95±2	103±3	101±2	115±3 ^{b,d}	115±3 ^{h,i,j}
CV (%)	2	2	3	2	2	3

Each value is expressed as mean ± SD ($n=7$). In group I, rats fed a normal diet were assumed to be 100%

Group I—NR normal, Group II—HCD high-cholesterol diet, Group III—NR + BT normal + black tea, Group IV—HCD + BT high-cholesterol diet + black tea, Group V—NR + GT normal + green tea, Group VI—HCD + GT high-cholesterol diet + green tea

^a Comparisons between group I and groups II, III, IV, V, and VI; statistically significant at $p<0.001$

^b Comparisons between group I and groups IV and V; statistically significant at $p<0.01$

^c Comparisons between group II and groups IV and VI; statistically significant at $p<0.001$

^d Comparisons between group III and group V; statistically significant at $p<0.05$

^e Comparisons between group IV and group VI; statistically significant at $p<0.001$

^f Comparisons between group III and group V; statistically significant at $p<0.001$

^g Comparisons between group IV and group VI; statistically significant at $p<0.05$

^h Comparisons between group I and group VI; statistically significant at $p<0.01$

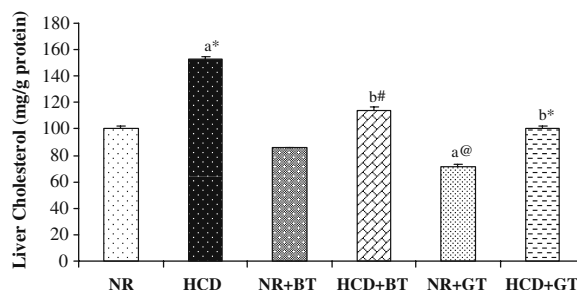
ⁱ Comparisons between group II and group VI; statistically significant at $p<0.01$

^j Comparisons between group IV and group VI; statistically significant at $p<0.01$

with black tea) rats showed a significant ($p < 0.001$ for cholesterol and LDL cholesterol) decrease values than those in group II rats. Similarly, group VI (HCD consuming with green tea) rats showed a significant ($p < 0.001$) decrease in cholesterol, triglycerides, and LDL cholesterol and a significant ($p < 0.01$) increase in HDL cholesterol, when compared with values in group II rats. Group V rats showed a significant ($p < 0.001$ for triglycerides; $p < 0.01$ for cholesterol and LDL cholesterol) decrease and significant ($p < 0.05$) increase in HDL cholesterol values than those in group III rats. Similarly, group VI rats showed a significant decrease in cholesterol and triglycerides ($p < 0.001$) and in LDL cholesterol ($p < 0.01$), and a significant increase in HDL cholesterol ($p < 0.05$), when compared with values in group IV rats.

The mean percentages of cholesterol and triglycerides in the hepatic tissue of group II rats were increased by 52.57% and 51.98%, respectively, when compared with group I rats. However, groups III and V rats (normal diet consuming with black and green tea) showed a significant decrease (14.20% and 28.50%, respectively) in cholesterol, when compared with percentages in group I rats. Similarly, a significant decrease (22.40% and 26.37%) in the hepatic tissue samples of triglycerides were observed in groups III and V rats, respectively, when compared with group I rats. The mean percentages of cholesterol in the hepatic tissue of groups IV and VI rats (HCD with black and green tea) significantly (25.01% and 34.39%, respectively) decreased compared with that of the group II rats (HCD). Interestingly, groups IV and VI rats exhibited a significant decrease (28.41% and 46.85%, respectively) in mean hepatic levels of triglycerides, when compared with group II rats (Figs. 1 and 2).

Figures 3 and 4 represent the abnormal elevation of lipid peroxidation (LPO) in the serum and hepatic tissue of HCD-fed rats. No significant differences were observed between the mean serum LPO concentration in rats of groups II and I. The mean concentration of LPO in the hepatic tissue samples from group II rats were significantly ($p < 0.001$) higher than that in group I rats. However, the mean LPO concentrations in serum samples of groups III and V ($p < 0.001$ and $p < 0.01$, respectively) rats were significantly lower than that in group I rats. Similarly, the mean LPO concentrations in hepatic tissue of groups III and V rats were also significantly lower ($p < 0.001$)



Each value is expressed as mean \pm SD (n=7).

Group I, Rats fed a normal diet were assumed to be 100%

Comparisons are made as follows: a – between group I and groups II, V
b – between group II and groups IV, VI

*, # and @ represents statistical significance at $p < 0.001$, $p < 0.01$ and $p < 0.05$, respectively.

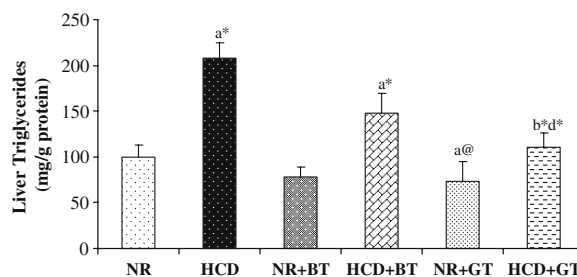
Abbreviations

G I - NR : Normal
G II - HCD : High cholesterol diet
G III - NR + BT : Normal + Black Tea
G IV - HCD + BT : High cholesterol diet + Black Tea
G V - NR + GT : Normal + Green Tea
G VI - HCD + GT : High cholesterol diet + GreenTea

Fig. 1 Effect of consuming black and green tea on the levels of cholesterol in hepatic tissue of control and experimental rats

than that in group I rats. No significant differences were noted between the mean LPO concentrations in serum samples of groups III and V, IV, and VI rats. The mean concentrations of LPO in the hepatic tissue of groups V and VI rats (HCD with black and green tea) were significantly ($p < 0.001$) higher than those in groups III and IV rats (normal with black and green tea), respectively.

Food efficiency is the calories consumed of a certain amount of food divided by weight gain. Foods with high food efficiency tend to add to



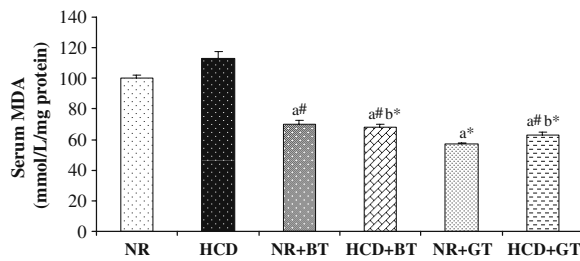
Each value is expressed as mean \pm SD (n=7).

Group I, Rats fed a normal diet were assumed to be 100%

Comparisons are made as follows: a – between group I and groups II, IV, V
b – between group II and groups VI
d – between group IV and group VI

*, # and @ represents statistical significance at $p < 0.001$, $p < 0.01$ and $p < 0.05$, respectively.

Fig. 2 Effect of consuming black and green tea on the levels of triglycerides in hepatic tissue of control and experimental rats



Each value is expressed as mean \pm SD (n=7).

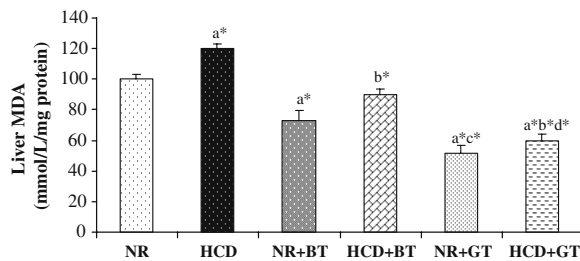
Group I, Rats fed a normal diet were assumed to be 100%

Comparisons are made as follows: a – between group I and groups III, IV, V, VI
b – between group II and groups IV, VI

* and # represents statistical significance at $p < 0.001$ and $p < 0.01$, respectively.

Fig. 3 Effect of consuming black and green tea on the levels of lipid peroxidation in serum of control and experimental rats

weight gain while foods with low food efficiency are more prone to be used as energy rather than stored as body weight. Figures 5, 6, and 7 show that HCD supplementation increased the body weight, food intake, and food efficiency (13.26%, 4.21%, and 10.31%, respectively), when compared with normal diet (group I). However, groups III and V rats (normal diet consuming with black and green tea) showed a significant decrease (18.10% and 23.25%, respectively) in the body weight, when compared with group I rats. No significant changes were observed in the food intake upon supplementation with black and green tea. Groups III and V rats showed a significant decrease (15.30% and 23.0%, respectively) in food efficiency than that of group I rats. Similarly, groups IV and VI rats showed a significant decrease (17.77% and 23.12%, respec-



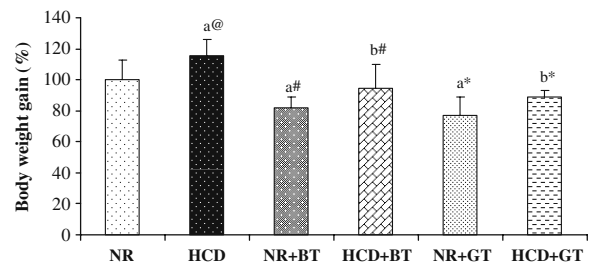
Each value is expressed as mean \pm SD (n=7).

Group I, Rats fed a normal diet were assumed to be 100%

Comparisons are made as follows: a – between group I and groups II, III, IV, V, VI
b – between group II and groups IV, VI
c – between group III and group V
d – between group IV and group VI

* represent statistical significance at $p < 0.001$.

Fig. 4 Effect of consuming black and green tea on the levels of lipid peroxidation in hepatic tissue of control and experimental rats



Each value is expressed as mean \pm SD (n=7).

Group I, Rats fed a normal diet were assumed to be 100%

Comparisons are made as follows: a – between group I and groups II, III, V
b – between group II and groups IV, VI

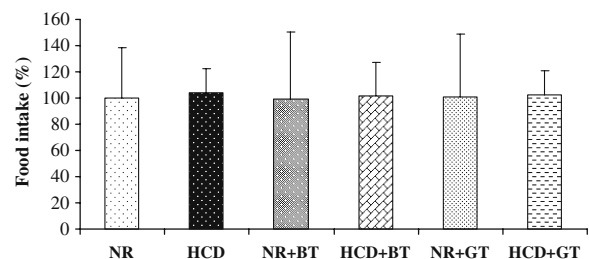
*, # and @ represents statistical significance at $p < 0.001$, $p < 0.01$ and $p < 0.05$, respectively.

Fig. 5 Effect of consuming black and green tea on body weight gain of control and experimental rats

tively) in the body weight, when compared with HCD (group II). Interestingly, food efficiency was decreased in rats of groups IV and VI (17.77% and 23.12%, respectively) compared with that in group II rats.

Discussion

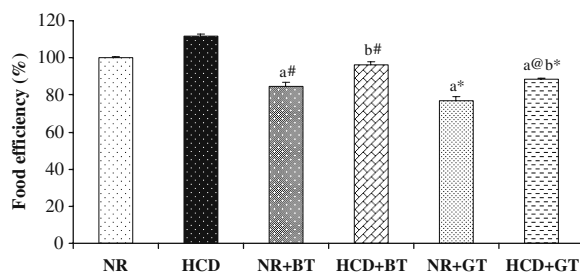
In general, green tea contains higher levels of catechins than black tea; the catechin contents of black tea are very low, which is in agreement with the degree of fermentation during manufacturing process [5]. Green tea is derived directly from drying and steaming the fresh tea leaves, and, thus, no fermentation (i.e., oxidation) occurs. During the fermentation, tea catechins are oxidized or condensed to other large polyphenolic molecules such as theaflavins and thearubigins [5].



Each value is expressed as mean \pm SD (n=7).

Group I, Rats fed a normal diet were assumed to be 100%

Fig. 6 Effect of consuming black and green tea on food intake of control and experimental rats



Each value is expressed as mean \pm SD (n=7).

Group I, Rats fed a normal diet were assumed to be 100%

The food efficiency was measured using the formula:

Food efficiency = Food intake/body weight gain [(g/d)/(g/wk)]

Comparisons are made as follows: a – between group I and groups III, V, VI

b – between group II and groups IV, VI

*, # and @ represents statistical significance at $p < 0.001$, $p < 0.01$ and $p < 0.05$, respectively.

Fig. 7 Effect of consuming black and green tea on food efficiency of control and experimental rats

The drugs available in the modern medicinal system are definitely good enough to provide symptomatic relief, but are associated with undesirable side effects. In recent years, there has been considerable emphasis in rediscovering biomolecules with hypolipidemic property from natural sources [48]. The results of the present work revealed collapse of oxidative defense system in hepatic tissue and lipoprotein oxidation in the hypercholesterolemic condition, while supplementation with black and green tea afforded considerable protection.

The importance of serum levels of cholesterol and lipoproteins in atherogenesis have been observed in a number of studies [35, 43]. Cellular cholesterol homeostasis is important for the prevention of cardiovascular diseases. Increased levels of serum cholesterol and LDL are associated with increased risk of developing coronary diseases [13, 47]. In the present study, rats fed high-cholesterol diet showed a significant increase in serum and hepatic cholesterol, triglycerides (TG), and serum LDL, and decrease in HDL. These findings are consistent with earlier reports [4, 35, 51]. In the present investigation, supplementation with BT and GT resulted in marked decrease in serum and hepatic cholesterol, TG, and serum LDL, and increased HDL level, when compared with the hypercholesterolemic group. This is indicative of the hypolipidemic effect of both BT and GT tested in this study, however; the recovery was more pronounced in GT consumption than in BT. Earlier studies have shown that polyphenols in tea may block the intestinal absorption of cholesterol and

enhance its fecal excretion from the body [19, 29]. Our previous studies have shown that the supplementation of tomato powder and lycopene to hydrogen peroxide-induced rats resulted in markedly lowered serum and liver cholesterol, TG, and serum LDL concentrations, while the serum HDL concentration was lowered [1]. The data presented herein show that BT and GT improves the serum and hepatic levels of lipid profile.

Lipid peroxidation of biological membranes can cause alterations in fluidity, reduction in membrane potential, increased permeability to H^+ and other ions, and eventual membrane rupture, leading to the release of cell and organelle contents. Cytotoxic aldehydes resulting from LPO can block macrophage action, inhibit protein synthesis, inactivate enzymes, and cross-link proteins, and can lead to the generation of thrombin [21]. Hence, lipid peroxidation can play a crucial role in inflammation, cancer, and cardiac diseases [15]. Further, a cholesterol-rich diet results in increased lipid peroxidation by the induction of free radical production and followed by hypercholesterolemia [52]. The significantly increased LPO in the high-cholesterol-fed rats of the present investigation corroborates with the earlier report of Gokkusu [17], who showed the correlation between lipid peroxidation and hypercholesterolemia. However, supplementation of BT and GT caused a significant decrease in lipid peroxidation level. Earlier studies have shown that tomato powder and lycopene possess potent inhibitory effects on the intensity of LPO in serum and hepatic samples from hydrogen peroxide-induced rats [1]. In addition, Sudhakar et al. [48] reported that rats fed with HCD showed a reduction in the LPO in hepatic tissue following luteol and luteol linoleate treatment. Earlier studies have shown that green tea polyphenols is a good scavenger of hydroxyl radicals and peroxynitrite radicals [20, 34]. Thus, the present study demonstrates that the supplementation of BT and GT results in near normal values of LPO in rats fed with HCD.

In the present study, the body mass examination of rats fed HCD revealed an increase in the body weight gain, food intake, and food efficiency, which are compatible with the process of hypercholesterolemia. These changes of the body mass and food efficiency correlated with the biochemical parameters observed in rats fed with HCD. No such significant changes were observed in the food intake upon supplementa-

tion with black and green tea. Further, supplementation of BT and GT to rats that had been fed with HCD prevented such alterations of the body weight gain and food efficiency. Similarly, an improvement in the body mass following the administration of piperine, an alkaloid constituent of black and long pepper to rats fed a high fat diet, and to hypercholesterolemic rats, has been reported [45]. These results suggest that BT and GT confer protection against the development of hypercholesterolemia in rats.

In conclusion, the present study emphasizes that the supplementation of BT and GT plays a positive role in high-cholesterol diet rats, possibly by suppressing the production of free radicals. However, it may be due to both antioxidative and hypolipidemic properties or by protecting the hepatic tissue from lipidemic–oxidative stress. The results of the present study suggest that BT and GT could serve as an easily accessible item of beverage/food rich in natural antioxidants, as a possible beverage supplement with food or even as a pharmaceutical agent. However, of the both tea tested, green tea appeared to be more effective than black tea.

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