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Chronic black tea administration protects plasma proteins, plasma, liver and kidney lipids against oxidation

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B Data Collection
C Statistical Analysis
D Data Interpretation
E Manuscript Preparation
F Literature Search
G Funds Collection

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Summary

Background: Black tea is known to have protective effects against plasma lipid and lipoprotein oxidation, but its influence on lipid peroxidation in tissue has been less studied. The effect of oral black tea consumption on protein oxidation has also not been demonstrated. The present study investigated the antioxidant effects of oral black tea consumption.

Material/Methods: Male Sprague-Dawley rats were fed a regular murine chow diet. The controls were supplied with water ad libitum, while the black tea group received aqueous black tea extract as the sole source of liquids. At the end of the ten-week experimental period, intestinal brush border, liver and kidney reduced-glutathione concentrations were evaluated as an index of cellular antioxidant defence. Plasma and tissue malondialdehyde concentrations and plasma protein carbonyl content were measured to evaluate lipid peroxidation and protein oxidation, respectively.

Results: The plasma malondialdehyde and protein carbonyl contents of rats consuming the black tea were significantly less than in controls. Similarly, liver and kidney malondialdehyde concentrations were significantly lower in the experimental group, while jejunoileal mucosa were not affected. Ten weeks of black tea administration caused significantly higher reduced-glutathione levels in the kidneys of black tea-administered rats, and a significant negative correlation was observed between kidney malondialdehyde and glutathione concentrations.

Conclusions: These findings provide evidence that long term black tea supplementation is capable of protecting both plasma proteins and plasma lipids from oxidative injury, and demonstrate that chronic black tea administration protects both liver and kidney tissues – but not the jejunoileal mucosa – against oxidation.

key words: **protein oxidation • plasma lipid peroxidation • tissue lipid peroxidation • antioxidants • glutathione**

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Background

Tea (*Camellia sinensis*) is the second most common beverage in the world after water. There are two major types of tea, namely green tea and black tea, both of which are reported to possess many beneficial health properties, including antipyretic, antineoplastic and antioxidant effects [1]. The antioxidant properties of tea extracts have been attributed to their content of polyphenols, which have been reported to inhibit oxidative processes in biological systems [1–3].

Although black tea is much more commonly consumed [4], studies on the antioxidant effect of tea extracts have been more focused on green tea. Nevertheless, studies on the antioxidant properties of black tea, which contains a higher quantity of catechin oxides, generally termed as thearubigens and theaflavins, and a lower quantity of free catechins, have recently been the object of scientific interest [5–8]. While studies on animal models and cell cultures have revealed that black tea has a preventive effect on oxidative DNA damage [9,10], *in vitro* studies indicate that black tea extracts are potent scavengers of reactive nitrogen and oxygen species [8], and point to stronger antioxidative activities for black tea than α-tocopherol in the erythrocyte ghost system [11]. On the other hand, while various animal studies demonstrate that black tea flavonoids inhibit plasma lipid and lipoprotein oxidation [12,13], and describe a rapid increase in plasma antioxidant capacity in *in vivo* black tea administration [14], there have been discrepant reports suggesting that black tea may not exhibit any protective effect on plasma and liver lipid peroxidation [15,16].

Although the protective effects of black tea on plasma lipid and lipoprotein oxidation, DNA oxidation and plasma antioxidant capacity have been better investigated, studies examining the influence of this beverage (which is thought to be well absorbed via the intestinal mucosa [5,17]) on tissue lipid peroxidation have been fewer and less conclusive [8,16,18,19]. Also, to our knowledge, the effects of oral black tea consumption on protein oxidation have not been demonstrated.

Accordingly, the present study was conducted to examine the possible protective effects of 10 weeks of oral black tea administration on plasma lipid and protein oxidation, intestinal brush border, liver and kidney lipid peroxidation and tissue reduced glutathione (GSH) concentrations, as an indicator of cellular antioxidant status, in rats.

Material and Methods

An aqueous black tea extract was prepared following the traditional Turkish method of tea preparation for human consumption: dry black tea leaves (Çaykur, Filiz™) (20 g) were added to 1000 ml of boiling water and brewed for 10 minutes, filtered into glass bottles and cooled before administration. The black tea extracts were renewed on alternating days during the experimental period.

Twenty male Sprague-Dawley rats, 16 weeks old (body weight of 260–420 g) were used, with the institutional approval of the Uludag University Animal Care and Use Committee. The animals were randomly divided into two groups: control (n=10) and BT (n=10). All animals were fed regular murine chow diet ad libitum. The controls were supplied with water ad libitum, while the BT group received the aqueous black tea extract as a sole source of potable liquid.

After 10 weeks of treatment, the animals from both groups were anaesthetized with diethyl ether, the abdominal wall was opened, and blood was collected from the heart into heparinized tubes. Plasma was isolated by centrifugation at 4°C (1000 × g) for 20 min and used immediately for measurements. The livers and kidneys were harvested, washed in ice-cold saline, dried on filter paper, and stored at –40°C for subsequent analyses. An approximately 10-cm jejunoileal segment was excised, opened longitudinally, cleaned of fecal material with a gentle jet of saline, and blotted dry. The mucosa (brush-border) was stripped off from the freshly opened intestine and homogenized in 1.15 percent cold KCl, and used for tissue malondialdehyde (MDA) measurement on the day of the experiment. The remaining homogenate was stored at –40°C for evaluation of tissue GSH and protein concentrations. All biochemical assays were performed within 4 days of sample freezing.

As an index of plasma lipid peroxidation, plasma MDA concentrations were determined by measuring the thiobarbituric acid reactive substances according to the spectrophotometric method of Kamal et al. [20], using 1,1,3,3-tetraethoxypropane (Fluka, Switzerland) as the external standard, and expressed as nanomole MDA per millilitre plasma. Plasma protein carbonyl content (PCC) was determined to evaluate protein oxidation as described by Reznick et al. [21], and was expressed as nanomole carbonyls per mg protein. Tissue MDA and GSH levels were evaluated according to the methods of Ohkava et al. [22] and Sedlak et al. [23], respectively, and were expressed in terms of tissue protein content, measured according to the method of Lowry et al. [24].

The Mann-Whitney U test was used to test the differences in values between the two groups. Linear curve estimation regression analysis was used to calculate regression between tissue MDA and GSH values. The significance level was set at $p<0.05$. The data presented in figures are means (±SD), with n=10.

Results

The body weights of the rats in the control and BT groups were statistically similar at the beginning (301±43 g and 336±44 g, respectively) and at the end (389±26 g and 372±25 g, respectively) of the experimental period. Significant weight gains were recorded for both control ($p<0.001$) and BT ($p<0.05$) groups at the end of ten weeks. Plasma MDA concentrations and plasma PCC were significantly less in the BT group compared to the controls ($p<0.001$ and $p<0.05$, respectively) (Figure 1) While liver and kidney MDA concentrations were found to be significantly lower in the BT group ($p<0.01$ and $p<0.001$, respectively), brush-border MDA concentrations did not exhibit any statistically significant difference between groups (Figure 2).

Tissue GSH concentrations in the control and BT groups are shown in Figure 3. Ten weeks of black tea administration caused significantly higher GSH concentrations in the kidneys of the BT group ($p<0.001$); however, brush-border and liver tissue GSH contents did not differ. Kidney GSH

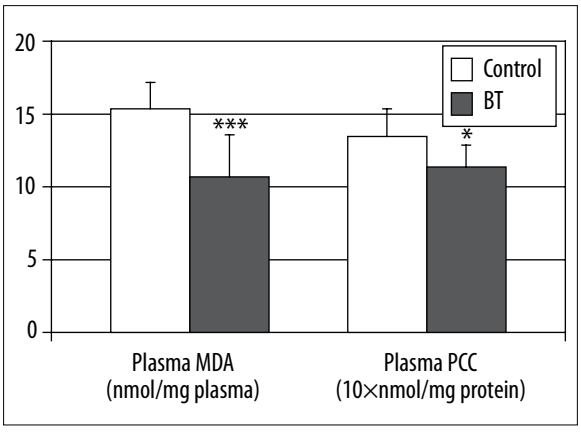

Figure 1. Plasma malondialdehyde (MDA) concentrations and protein carbonyl contents (PCC) in the control and black tea (BT) groups. * p<0.05 and *** p<0.001, compared to controls.

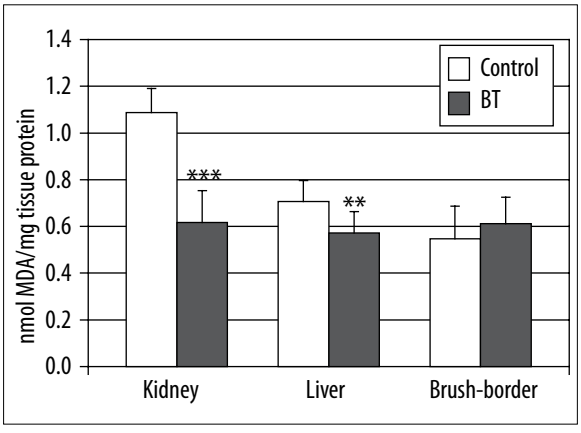

Figure 2. Kidney, liver and brush-border tissue malondialdehyde (MDA) concentrations in the control and black tea (BT) groups. ** p<0.01 and *** p<0.001, compared to controls.

and MDA concentrations were found to be significantly correlated (r=–0.815; p<0.001) (Figure 4).

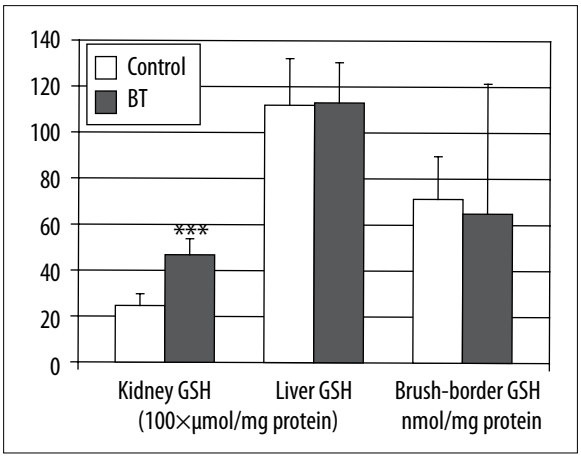

Figure 3. Kidney, liver and brush-border tissue glutathion (GSH) concentrations in the control and black tea (BT) groups. *** p<0.001, compared to controls.

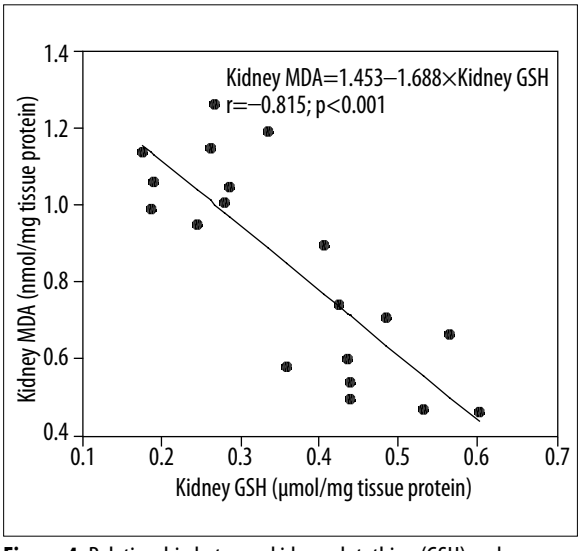

Figure 4. Relationship between kidney glutathion (GSH) and malondialdehyde (MDA) concentrations.

Discussion

The results of the present study indicate that black tea extract administration for 10 weeks protects rat plasma from lipid peroxidation (Figure 1). Previous reports regarding the effects of black tea on plasma lipid and lipoprotein oxidation indices in various animal and human models support the findings of our study [12,13]. However, using a similar experimental design and methods, Cherubini et al. [15] and Da Silva et al. [12] reported contradictory results on the protective effect of black tea extracts on plasma lipid peroxidation, the former stating that black tea administration did not increase the resistance of plasma to lipid peroxidation. The authors [15] suggested that the discrepancy between the two studies was due to the different doses of the extracts used, and stated that the dose in the study by Da Silva et al. [12] cannot be achieved in humans by tea drinking. The black tea extracts used in the present model were prepared in the traditional way of Turkish tea preparation for human consumption, and administered chronically, contrary to the single dose administration applied by Cherubini et al. [15]. Therefore, we suggest that the duration of consumption may be as important for the antioxidant effect of black tea as the dose administered.

The significantly lower levels of PCC observed in BT group in the present study show that black tea administration is as efficient in protecting plasma proteins from oxidation as in protecting plasma lipids. To our knowledge, the influence of black tea on protein oxidation is not documented in the literature, and this is the first report showing the antioxidant effect of black tea on plasma protein oxidation (Figure 1). The fact that black tea improves plasma antioxidant capacity has been documented by several authors: Serafini et al. [14] have shown that black tea supplementation significantly improves plasma antioxidant capacity, and Da Silva et al. [12] found that black tea supplementation delays α-tocopherol depletion in Cu-oxidized plasma. The significantly lower levels of plasma PCC and MDA observed in the present model suggest that, either by directly protecting plasma lipids and proteins from oxidation or by improving the existing anti-

oxidant defenses in plasma, chronic black tea administration exerts an antioxidant effect on plasma molecules.

Since black tea is reported to be absorbed via the gastrointestinal tract and function in circulation [25], we plan to examine its possible antioxidant effects on tissue lipid peroxidation and tissue antioxidant defense systems in the same animal model. GSH is one of the main parts of the cellular endogenous antioxidant system. It exerts its antioxidant function by donating electrons to radicals and changing to its oxidized form, which is subsequently reduced by the enzyme glutathione reductase [26]. In the present study, kidney, liver and jejunoileal brush-border MDA levels and their relation to tissue GSH concentrations were examined. According to the results, black tea administration significantly protected kidney and liver tissues from lipid peroxidation (Figure 2). The results of recent work by Sava et al. [7] demonstrating the liver-protecting activity of melanin-like pigment derived from black tea, are in line with our findings. Ghiselli et al. [25] have stated that polyphenols from the tea extracts are absorbed in the higher regions of the gastrointestinal tract, probably starting from the stomach. The reason why jejunoileal mucosa were not protected in the present model may be the earlier absorption of the antioxidant molecules of tea extract from the alimentary canal, before they reached the site examined. Further studies examining the effects of black tea on each level of the gastrointestinal tract will better illuminate this topic.

Significantly higher GSH concentrations were detected only in kidney tissues after the experimental period (Figure 3). In addition, the significant negative correlation observed between kidney MDA and GSH levels (Figure 4) suggests that the improvement of the GSH defense system by chronic black tea supplementation is efficient in protecting this tissue against lipid peroxidation. The higher levels of GSH measured in kidney tissues may be either because of the induction of GSH synthesis or slower oxidation of GSH molecules. Liver and brush border GSH concentrations in the BT group were not different from those of the controls (in accordance with published data [27]). This could be interpreted as kidney tissue being better protected by black tea; however, further studies on the metabolism of black tea flavonoids, their absorption and excretion, and their influence on the GSH system should be conducted to make a precise judgment on this matter.

Conclusions

In conclusion, the present study provides evidence that long term black tea supplementation is capable of protecting both plasma proteins and plasma lipids from oxidative injury. Also, based on the present model, we suggest that chronic black tea administration protects both liver and kidney tissues – but not the jejunoileal mucosa – against oxidation. Further studies examining the changes in tissue protein oxidation and black tea flavonoid concentrations in plasma and tissues are needed to better define the cellular mechanisms underlying the antioxidant effects of this beverage.

References:

1. Dufrense CJ, Farnworth ER: A review of latest research findings on the health promotion properties of tea. J Nutr Biochem, 2001; 12: 404–21
2. Auger C, Al-Awwadi N, Bornet A et al: Catechins and procyanidins in Mediterranean diets. Food Res Int, 2004; 37: 233–45
3. Gou Q, Zhao B, Shen S et al: ESR study on the structure-antioxidant activity relationship of tea catechins and their epimers. Biochim Biophys Acta, 1999; 1427: 13–23
4. Kuroda Y, Hara Y: Antimutagenic and anticarcinogenic activity of tea polyphenols. Mutat Res, 1999; 436: 69–97
5. Hollman PCH, Tijburg LBM, Yang CS: Bioavailability of flavonoids from tea. Crit Rev Food Sci Nurt, 1997; 37: 719–38
6. McAngelis GT, McEneny J, Pearce J, Young IS: Black tea consumption does not protect low density lipoprotein from oxidative modification. Eur J Clin Nutr, 1998; 52: 202–6
7. Sava VM, Hung YC, Blagodarsky VA et al: The liver-protecting activity of melanin-like pigment derived from black tea. Food Res Int, 2003; 36: 505–11
8. Maity S, Ukil A, Karmakar S et al: Thearubigin, the major polyphenol of black tea, ameliorates mucosal injury in trinitrobenzene sulfonic acid-induced colitis. 2003; 470: 103–12
9. Lodovici M, Casalini C, De Filippo C et al: Inhibition of 1,2-dimetylhydrazine-induced oxidative DNA damage in rat colon mucosa by black tea complex polyphenols. Food Chem Toxicol, 2000; 38: 1085–88
10. Feng Q, Torri Y, Uchida K et al: Black tea polyphenols, theaflavins, prevent cellular DNA damage by inhibiting oxidative stress and suppressing cytochrome P450 1A1 in cell cultures. J Agric Food Chem, 2002; 50: 213–20
11. Shiraki M, Hara Y, Osawa T et al: Antioxidative and antimutagenic effects of theaflavins from black teas. Mutat Res, 1994; 323: 29–43
12. Da Silva EL, Piskula M, Terao J: Enhancement of antioxidative ability of rat plasma by oral administration of (–) – epicatechin. Free Radic Biol Med, 1998; 24: 1209–16
13. Vinson JA, Dabbagh YA: Effect of green and black tea supplementation on lipids, lipid oxidation and fibrinogen in the hamster: mechanisms for the epidemiological benefits of tea drinking. FEBS Lett, 1998; 433: 44–46
14. Serafini M, Ghiselli A: *In vivo* antioxidant effect of green and black tea in man. Eur J Clin Nutr, 1996; 50: 28–32
15. Cherubini A, Beal MF, Frei B: Black tea increases the resistance of human plasma to lipid peroxidation *in vitro*, but not *ex vivo*. Free Radic Biol Med, 1999; 27: 381–87
16. De Vos S, De Schrijver R: Lipid metabolism, intestinal fermentation and mineral absorption in rats consuming black tea. Nurt Res, 2003; 23: 527–37
17. Dreosti IE: Bioactive ingredients: antioxidants and polyphenols in tea. Nutr Rev, 1996; 54: 51–54
18. Bu-Abbas A, Dobrota M, Copeland E et al: Proliferation of hepatic peroxisomes in rats following the intake of green or black tea. Toxicol Lett, 1999; 109: 69–76
19. Fadhel ZA, Amran S: Effects of black tea extract on carbon tetrachloride-induced lipid peroxidation in liver, kidneys, and testes of rats. Phytotherapy Res, 2002; 16: 28–32
20. Kamal A, Gomaa A, Khafif M, Hammad A: Plasma lipid peroxides among workers exposed to silica or asbestos dust. Environ Res, 1989; 49: 173–80
21. Reznick AZ, Packer L: Oxidative damage to proteins: spectrophotometric method for carbonyl assay. Methods Enzymol, 1994; 233: 357–63
22. Ohkawa H, Ohishi N, Yagi K: Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. Anal Biochem, 1979; 95: 351–58
23. Sedlak J, Lindsay RH: Estimation of total protein-bound and non protein sulfhydryl groups in tissue with Ellman's reagent. Anal Biochem, 1968; 25: 192–205
24. Lowry OH, Rosebrough NJ, Farr AL, Randall RJ: Protein measurement with the Folin phenol reagent. J Biol Chem, 1951; 193: 265–75
25. Ghiselli A, Serafini M, Natella F, Scaccini C: Total antioxidant capacity as a tool to assess redox status: critical view and experimental data. Free Radic Biol Med, 2000; 29: 1106–14
26. Rice-Evans C, Burdon R: Free radical-lipid interactions and their pathological consequences. Prog Lipid Res, 1993; 32: 71–110
27. Marnewick JL, Joubert E, Swat P et al: Modulation of hepatic drug metabolizing enzymes and oxidative status by rooibos (Aspalathus linearis) and Honeybush (Cyclopia intermedia), green and black (Camellia sinensis) teas in rats. J Agric Food Chem, 2003; 27: 8113–19