OCTOBER 1998
VOLUME 46, NUMBER 10

JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Effect of Green Tea and Black Tea on the Blood Glucose, the Blood Triglycerides, and Antioxidation in Aged Rats

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During a 75 day feeding experiment of 108 12-month-old Sprague-Dawley rats in 13 groups respectively consuming diets with addinged green tea (GT), black tea (BT), and their water extracts (GTWE, BTWE), blood glucose was significantly decreased in all experimental groups, by averages of 23.9% in GT and GTWE and by 22.8% in BT and BTWE; the blood triglycerides were significantly reduced, by 33.3% in GT and GTWE and by 25.0% in BT and BTWE. The activity of superoxide dismutase was significantly increased, by averages of 117.0% in BT and BTWE and 90.8% in GT and GTWE. However, malondialdehyde was significantly decreased, by averages of 34.6% in BT and BTWE and 25.4% in GT and GTWE. This indicated that the ability of green tea to reduce blood glucose and blook triglycerides was higher than that of black tea in the aged rats but that the antioxidative ability of black tea was better than that of green tea in the aged rats.

Keywords: *Tea; BG; GTG; antioxidation*

INTRODUCTION

High blood glucose (BG) is one of the markers of the diabetes, and a high triglyceride level is one of the markers leading to chronic diseases such as cardiovascular disease and hypertension (Giugliano et al., 1995). Drinking tea has beneficial effects on serum lipids and blood pressure (Bozidar, 1994). Oxidative damage is thought to represent one of the mechanisms leading to chronic diseases such as atherosclerosis and cancer (Halliwell and Gutteridge, 1989). Oxidative stress is a major component in the development of late complications in diabetic patients (Hunt et al., 1990; Tsai et al., 1994). It is probably based on hyperglycemia. During the cellular metabolization of glucose, superoxide anions can be formed that shift the pro-/antioxidative balance in blood (Thornally, 1985).

Polyphenols are secondary plant metabolites occurring widely in plant food (Harborne, 1989). They possess outstanding antioxidant and free radical scavenging properties, suggesting a possible protective role in man (Laughton et al., 1991; Scott et al., 1993). Recent papers have provided the first evidence of an epidemiological link between polyphenol intake and risk of cardiovascular disease in man (Hertog et al., 1993). Evidence of an in vivo antioxidant effect in man after ingestion of polyphenol-rich beverages, such as tea (Serafini et al., 1994) and wine (Maxwell et al., 1994), has also been recently published. However, the mechanisms through which these compounds act are still incompletely understood.

Tea is a widely consumed beverage throughout the world. It has been calculated that the average consumption is 6 g per day for a 60 kg human (Chen, 1985; Deng et al., 1997). Tea leaves contain >35% of their dry weight in polyphenols, which differ depending on the manufacturing procedure (Balentine, 1992). Green tea, a nonfermented type of tea preferred in China, is rich in flavanols (mainly catechin, epicatechin, and epigallocatechin), flavandiols, and simple phenolic acids. Black tea, mostly consumed in the Western world, is obtained by fermentation of the green tea. During fermentation, simple polyphenols undergo an enzymatic polymerization that leads to the formation of complex compounds of condensation. The principal condensation

10.1021/jf980105+ CCC: $15.00 © 1998 American Chemical Society
Published on Web 09/18/1998

Table 1. Division of the Groups

group	no. of rats	GTWE (g/100 mL)	BTWE (g/100 mL)	GT in diet (g/kg)	BT in diet (g/kg)
control	12	0	0	0	0
1	8	0.6	0	0	0
2	8	1.2	0	0	0
3	8	2.4	0	0	0
4	8	0	0	5	0
5	8	0	0	10	0
6	8	0	0	20	0
7	8	0	0.6	0	0
8	8	0	1.2	0	0
9	8	0	2.4	0	0
10	8	0	0	0	5
11	8	0	0	0	10
12	8	0	0	0	20

compounds present in black tea are theaflavins and thearubigens, which have molecular weights of ∼500−3000 (Balentine, 1992).

In this paper we have investigated the effects of green tea and black tea on the blood glucose (BG) and the blood triglyceride (BTG) levels and on the in vivo antioxidant action in old rats.

MATERIALS AND METHODS

Chemicals and Reagents. Thiobarbituric acid (TEA) and 1,1,3,3-tetraethoxypropane (TEP) were purchased from Sigma Chemical Co.; the others were from China Chemical Co.

Animals for Experiments. One hundred and eight Sprague-Dawley (SD) rats were selected for experiment. The rats were 12 months old, males and females were in equal numbers, and the average weight of the rats was 300 ± 12 g (303 ± 15 g after consumption of the experimental diet; no significant difference in each group, $p > 0.05$, not shown). Each rat was healthy and noninfected with emidemic virus or bacteria. The divison of the experimental rat groups was set as in Table 1. Animals were housed for 75 days in cages (3 rats per cage) and kept in a room at 22 ± 1 °C, with a 12 h light−dark cycle. The rats were fed with the complete diet.

Tea for Experiment and Sample Preparation. Green tea produced in Wuyuan and black tea in Qimen (in China) were ground and mixed with rat feed in designed dose (GT and BT) to feed the rats. Water extracts of green and black tea, boiled for 5 min, were filtered for random drinks (GTWE and BTWE). The contents of tea polyphenols and catechins in GT, BT, and their water extracts are shown in Table 2.

Tea Dose for Experiment. Each SD rat should consume 30 g of feed and 25 mL of water every day (National Research Council, 1972). The average tea consumption was estimated at 6 g/day for a 60 kg human (Chen, 1985). Tea doses in the feed and water extract were set at 5, 10, and 20 times the human dose; thus 5, 10, and 20 g of tea/kg of rat feed and 0.6, 1.2, and 2.4 g of tea/ per 100 mL of water extract were selected.

Collection and Analysis of Samples. After 75 days, the rats were fasted for 15 h and then anesthetized with phenobarbital; the blood was drawn in ice-cold heparin tubes at the abdominal aorta. Red cells were obtained by centrifuging a part of the blood at 4 °C for 10 min (4000 rpm). Livers were rinsed by ice-cold physiologic saline. Red cells and livers were immediately taken for the determination of their antioxidant effect. BG (GOD−POD method) and BTG levels (GPO−PAP method), Hb superoxide dismutase (SOD) activity (autoxidative method of triphenol), and liver malondialdehyde (MDA) (Bird et al., 1983) were determined.

Statistical Analysis. Results are expressed as means ± SD for observation of the indicated number (n). Statistical significance of differences between means was assessed by variance analysis (ANOVA). Values of $p < 0.05$ were considered to be significant and values of $p < 0.01$ very significant.

RESULTS

The contents of BG and BTG were significantly decreased by the addition of the green tea, the black tea, and their water extracts to the diets, but they were not dose-dependent. BG was decreased on average (control = 10.52 mg/100 mL) by 23.9% (7.8 mg /100 mL) in GT and GTWE and by 22.8% (7.9 mg/100 mL) in BT and BTWE; BTG was decreased on average (control = 1.32 mmol/L) by 33.3% (0.88 mmol/L) in GT and GTWE and by 25.0% (0.99 mmol/L) in BT and BTWE (Table 3).

GT, BT, and their water extracts had significant antioxidative activity. SOD was very significantly increased in all experimental groups ($p < 0.01$) (control = 449 ± 31 units/mg·Hb), on average by 117.0% (975.7 units/mg·Hb) in BT and BTWE and by 90.8% (857.8 units/mg·Hb) in GT and GTWE. Meanwhile, MDA in the liver was significantly decreased (control = 19.8 ± 2.2 nmol/g), on average by 34.6% (10.93 nmol/g) in BT and BTWE and by 25.4% (14.7 nmol/g) in GT and GTWE (Table 3). However, there was no significant difference in SOD and MDA between the groups of green tea and the groups of black tea.

DISCUSSION

Human beings, especially old men, have many chronic diseases such as diabetes, cardiovascular disease, and cancer (Christian, 1995). A high blood glucose level is one of the markers of diabetes, and a high triglyceride level is one of the markers of cardiovascular disease (Giugliano et al., 1995). The average life span of the rat is ∼18 months (Si, 1989). Compared with a human, a 12−14.5-month-old rat is equal to a 50−60-year-old man. The effects of green tea and black tea on the physiology of an old rat might represent their effects on an old man. In this experiment, green tea and black tea and their water extracts have significantly decreased BG and BTG. The decrease of BG and BTG in the normal ranges that are individually 7.6−10.6 mg/10 mL and 0.62−1.28 mmol/L (Si, 1989) might be applicable to humans. Sawsan et al. (1994) found that the extract of tea significantly reduced the mucosal uptake of both sodium and glucose. This supports the use of tea as a crude medicine in China for thousands of years. The mechanism of reducing BG is considered to be the diphenylamine and polysaccharides of tea (Karawya, 1984). Polysaccharides might inhibit the absorption of glucose, and diphenylamine might promote the metabolism of BG. The mechanism of reducing BTG is not clear. Some scientists (Hara and Honda, 1990; Thompson and Yoon, 1984) considered that the polyphenols bind proteins, decrease the activity of digestive enzymes, and finally reduce the digestibility and/or absorption of glucose and lipids.

Ample experimental and epidemiogical evidence supports the involvement of oxidative stress in the pathogenesis and progression of several chronic diseases (Halliwell et al., 1992). Lipid peroxidation is an autocatalytic free-radical-mediated destructive process whereby polyunsaturated fatty acids in cell membrances undergo degradation to form lipid hydroperoxides (Sevanian, 1985; Slater, 1984). These latter compounds decompose to form a wide variety of products, low molecular mass hydrocarbons, hydroxy aldehydes, fatty acids, ketones, alkenals, and alkanals, including malondialdehyde (MDA), whereas SOD can hinder the formation of these products. The reduction of MDA indicates the inhibition of lipid hydroperoxides, and the increase of SOD activity might represent antioxidative augmentation; these prove that green and black tea have an excellent antioxidation activity.

Table 2. Contents of Tea Polyphenols and Catechins in Green Tea, Black Tea, and Their Water Extracts[a] (Milligrams per Gram, Milligrams per 100 mL)

	polyphenols	L-EGC	D,L-GC	L-EC + D,L-C	L-EGCG	L-ECG
GT	226 ± 6	28.4 ± 1.3	8.1 ± 0.8	10.6 ± 0.8	82.7 ± 3.6	16.5 ± 1.0
BT	100 ± 4	6.0 ± 0.3	5.0 ± 0.4	9.3 ± 0.7	17.5 ± 1.2	2.9 ± 0.1
GTWE 0.6	67 ± 2	8.8 ± 0.4	2.6 ± 0.2	3.1 ± 0.4	24.8 ± 1.6	4.9 ± 0.3
GTWE 1.2	122 ± 3	15.6 ± 1.2	4.7 ± 0.3	5.8 ± 0.4	44.8 ± 2.3	8.0 ± 0.4
GTWE 2.4	228 ± 8	28.6 ± 1.9	8.1 ± 0.8	10.6 ± 0.9	83.9 ± 4.8	16.0 ± 1.0
BTWE 0.6	31 ± 2	1.8 ± 0.3	1.5 ± 0.1	2.8 ± 0.1	5.5 ± 0.2	0.9 ± 0.1
BTWE 1.2	58 ± 3	4.4 ± 0.1	3.4 ± 0.1	5.7 ± 0.2	10.6 ± 0.8	1.8 ± 0.1
BTWE 2.4	108 ± 4	6.4 ± 0.1	5.4 ± 0.1	9.9 ± 0.3	18.8 ± 1.3	3.7 ± 0.1

[a] EGC, epigallocatechin; GC, gallocatechin; EC, epicatechin; C, catechin; EGCG, epigallocatechin gallate; ECG, epicatechin gallate.

Table 3. Effect of Green and Black Tea on SOD, MDA, BG, and BTG ($X \pm$ SD)

group	no. of rats	SOD (unit/mg·Hb)	MDA (nmol/g)	BTG (mmol/L)	BG (mg/10 mL)
control	12	450 ± 31	19.8 ± 2.2	1.32 ± 0.21	10.5 ± 0.2
1 GTWE 0.6	8	762 ± 36[b]	15.8 ± 3.3	0.61 ± 0.11[b]	7.60 ± 0.7[b]
2 GTWE 1.2	8	810 ± 48[b]	15.8 ± 3.5	1.22 ± 0.17	8.1 ± 0.6[a]
3 GTWE 2.4	8	826 ± 78[b]	16.3 ± 3.9	0.63 ± 0.05[b]	7.7 ± 0.5[b]
4 GT 5	8	502 ± 102	13.2 ± 4.3[a]	0.85 ± 0.05[a]	7.7 ± 0.6[b]
5 GT 10	7	1122 ± 135[b]	14.2 ± 3.7[a]	1.18 ± 0.14	7.9 ± 0.6[b]
6 GT 20	7	1124 ± 230[b]	13.3 ± 1.6[a]	0.79 ± 0.14[a]	7.9 ± 0.6[b]
7 BTWE 0.6	7	832 ± 62[b]	12.6 ± 2.5[b]	0.99 ± 0.14	8.5 ± 0.5[a]
8 BTWE 1.2	8	1200 ± 85[b]	12.2 ± 2.9[b]	0.92 ± 0.11[a]	7.8 ± 0.6[b]
9 BTWE 2.4	7	952 ± 61[b]	13.3 ± 2.9[a]	0.91 ± 0.15[a]	7.8 ± 0.8[b]
10 BT 5	7	867 ± 38[b]	12.9 ± 2.9[b]	0.89 ± 0.18[a]	8.2 ± 0.5[a]
11 BT 10	7	899 ± 105[b]	13.6 ± 3.7[a]	1.09 ± 0.10	7.7 ± 0.7[b]
12 BT 20	7	1105 ± 84[b]	12.9 ± 3.5[b]	1.11 ± 0.18	7.6 ± 0.5[b]
1−6[c]	46	857.8	14.7	0.88	7.8
7−12[c]	43	975.7	10.9	0.99	7.9

[a] Compared with control $p < 0.05$. [b] Compared with control $p < 0.01$. [c] 1−6 and 7−12 are individually the averages from group 1−6 and from 7−12.

Polyphenols possess powerful antioxidative properties (Serafini et al., 1994; Maxwell et al., 1994) and are now considered to be potentially important for the prevention of chronic diseases in man. Polyphenols are regarded as a class of semiessential food components.

Green tea and black tea represent an excellent source of antioxidant polyphenols. In vitro, the antioxidative capacity of green tea is much higher than that of black tea (Serafini et al., 1996; Gerhard et al., 1989). The same explanation might be used in the result of this experiment showing that the activities of SOD and MDA were increased with the higher dose of tea or tea water extract. The antioxidative properties of green and black tea in vivo were researched very little. Serafini et al. (1996) were surprised to find that black tea produced a response of the same antioxidative intensity of green tea in vivo. Mitsuaki et al. (1995) found that lipid peroxidation (MDA as a indicator) by $BrCC1_3$ in the liver of rats fed green and black tea was inhibited by 18 and 32% ($p < 0.05$), respectively. In this experiment we found that the antioxidative capacity of black tea was higher than that of green tea in vivo. The activity of SOD in the groups of black tea is higher than that in the groups of green tea ($p > 0.05$), whereas MDA was lower than that in the groups of green tea ($p > 0.05$). These results are similar to those of Mitsuaki et al. (1995).

Black tea is obtained by fermentation of green tea; during this process, simple polyphenols are polymerized to the condensed polyphenols, and the content of polyphenols was much higher in GTWE than in BTWE (Table 2). We therefore speculate that after ingestion either the condensed polyphenols of black tea rapidly break down and form different molecular structures having higher antioxidation capacity or some substances other than tea polyphenols, produced in the process of fermentation, are absorbed and decomposed in the body and play important roles in antioxidation, but further research should be done.

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Received for review February 4, 1998. Revised manuscript received June 30, 1998. Accepted July 8, 1998.

JF980105+