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EFFECT OF BLACK TEA ON SOME BIOCHEMICAL PARAMETERS IN BROILER EXPOSED TO EXPERIMENTAL HYDROGEN PEROXIDE-INDUCED OXIDATIVE STRESS

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ABSTRACT

The effect of 3 weeks oral administration of 20% black tea infusion or decoction alone or combined with 0.5% H_2O_2 on body weight and certain blood biochemical constituents was studied in broiler chickens. The results indicated a significant decrease in body weight, Treatment with black tea decoction with or without H_2O_2 produced a significant decrease in both serum cholesterol and total lipid. Black tea infusion with H_2O_2 induced a significant decrease in serum cholesterol level. A significant increase in serum sodium concentration was observed in animals treated with black tea decoction. In addition, black tea decoction alone or with H_2O_2 caused a significant decrease in aspartate aminotransferase (AST) concentration. Moreover, different black tea treatments decreased liver glycogen concentration compared with its increased level with H_2O_2 treated groups. It is concluded that the use of widely available, inexpensive beverage, tea displayed valuable preventive properties on some biochemical parameters in broiler exposed to experimental hydrogen peroxide-induced oxidative stress.

تأثير الشاي الاسود على بعض التغييرات الكيميائية الحياتية للدم ومستوى كلايكوجين الكبد في افراخ الدواجن المعرضة للكرب التأكسدي المستحدث ببيروكسيد الهيدروجين

فدوى خالد توفيق

فرع الفسلجة، كلية الطب البيطري، جامعة الموصل، الموصل، العراق

الخلاصة

درس تاثير اعطاء مغلي ومنقوع الشاي الاسود (20%) لوحده او مع 0.5% بيروكسيد الهيدروجين مع ماء الشرب لمدة ثلاثة اسابيع على مكونات الدم الكيميائية الحياتية وكلايكوجين الكبد في فروج اللحم المعرض الى الكرب التأكسدي التجريبي. وقد اظهرت

النتائج ان اعطاء بيروكسيد الهيدروجين لوحده او مع نقيع او مغلي الشاي الاسود احدث انخفاضا معنويا في وزن الجسم مع عدم حدوث تغيرات معنوية في تركيز كلوكوز الدم. اما منقوع الشاي لوحده او مع بيروكسيد الهيدروجين فقد سبب انخفاضا معنويا في تركيز الكوليستيرول والدهون الكلية بينما ادت المعاملة بمغلي الشاي مع بيروكسيد الهيدروجين انخفاضا معنويا في تركيز الكوليستيرول. ومن ناحية اخرى ادت المعاملة بمنقوع الشاي الى زيادة معنوية في تركيز الصوديوم في الدم بينما ادت المعاملة بمنقوع الشاي لوحده او مع بيروكسيد الهيدروجين الى حدوث انخفاضا معنويا في فعالية خميرة الاسبرتيت نائلة الامين (Aspartate aminotransferase AST) فضلا عن ذلك احدثت المعاملات المختلفة للشاي انخفاضا في محتوى كلايكوجين الكبد مقارنة بمستواه المتزايد التي ظهرت في المجموعة المعاملة ببيروكسيد الهيدروجين. يستنتج من ذلك ان تناول الشاي المشروب المتوفر وغير المكلف قد يلعب دورا وقائيا" في بعض المتغيرات الكيميائية الحياتية في افراغ الدواجن المعرضة للكرب التأكسدي المستحدث ببيروكسيد الهيدروجين.

INTRODUCTION

Tea (from tea plants *Camellia sinensis*, family *Theaceae)* is one of the most popular beverages in the world because of its attractive flavor, aroma and taste and it is one of the safest beverages since it is made with boiling, sterile water. There are 3 general forms of tea: the unfermented green tea, the partially fermented Oolong tea and the fermented black tea which is the major form of tea consumed (1,2).Epidemiological observations and laboratory studies have indicated that tea may reduce the risk of a variety of illness including cancer and coronary heart disease (3). The most important chemicals in tea are the polyphenols which is a large diverse naturally occurring class of compounds (3) present in high quantities in dry tea leaves (4). The flavonoids are the largest and best studied group of polyphenols, Most of them have antioxidant effects (3). Black tea contains products of enzymatic polymerization of simple polyphenols particularly the aflavins and the arubigins (5). Also tea leaves contain several lipid soluble chemicals such as beta carotene and tocopherol (6). Although most of the research has focused on the beneficial effect of green tea, evidence is accumulating that black tea may share some of these properties, with the concept of preventive diseases, using naturally occurring substances is gaining increasing attention. There is currently much interest in the antioxidant role of flavonoids and other polyphenols found in tea (7). Studies *in vivo* and *in vitro* provide evidence of tea active ingredients with potential to lowering blood glucose level in diabetic rats (8), also with hypocholesterolemic effect (9, 10, 11, 12, 13). The present

study was undertaken to investigate the effect of black tea (decoction and infusion) on some blood biochemical parameters in broiler exposed to experimental hydrogen peroxide-induced oxidative stress.

MATERIALS AND METHODS

Animals

Broiler chickens weighing 221 - 250 gram, 21 days old, were housed and maintained under controlled conditions of temperature (25 - 29 °C) and light (14 hr/day). They were allowed free access of commercial poultry ration, black tea decoction or infusion (as a sole source of drinking fluid) during three weeks experimental period.

Experimental Design

Animals were randomly divided into six groups (7 chickens/group). Group 1 (control) received tap water. Group 2 received 0.5% hydrogen peroxide (Al-Shaheed Factory, Baghdad) with drinking water (14). Group 3 received 0.5% H_2O_2 with 20% black tea infusion (15). Group 4 received 20% black tea as a sole source of drinking water. Group 5 received 0.5% H_2O_2 with 20% black tea decoction (15) and Group 6 received 20% black tea decoction as a sole source of drinking fluid during the three weeks experimental period.

Samples Collection and Analysis

Individual body weights of control and treated groups were recorded before and after the three weeks experimental period. Blood samples were collected at slaughtering time. Sera were separated immediately and stored at (-20 °C) until assayed for glucose, cholesterol, total lipids, sodium and potassium ions, aspartate aminotransferase (AST) and alanine aminotransferase(ALT).

Serum glucose and cholesterol were measured using calorimetric assay kits (Bicon, Diagnostic GmbH, Burbach, Germany and BioMericux, France, respectively).

Serum total lipids, Sodium and potassium ions concentrations were determined according to Toro and Ackermann (16). AST and ALT were measured using calorimetric assay kits (RANDOX, England). Liver glycogen was determined using Anthron method (17).

Statistical Analysis

The data were analyzed statistically using one-way analysis of variance. The specific group differences were determined using Duncan multiple range test. The accepted level of significance was at $P<0.05$ (18).

RESULTS

Oral administration of H_2O_2 alone or with different forms of black tea for the three weeks experimental period resulted in a significant decrease in body weight as compared with the control group (Table 1). Glucose concentration was not affected by H_2O_2 treatment. Furthermore, different black tea treatments with or without H_2O_2 did not show significant changes in serum glucose concentration (Fig. 1-a). Treatment with H_2O_2 produced no change in serum cholesterol and total lipids concentration as compared with control group while groups treated with black tea decoction with or without H_2O_2 produced a significant decrease in both serum cholesterol and total lipids compared with both control and H_2O_2 treated groups. Black tea infusion with H_2O_2 caused a significant decrease in serum cholesterol level compared with both control and H_2O_2 treated groups (Fig. 1-b, c).

Table 1: Effect of 3 weeks oral administration of 0.5% H_2O_2, 20% black tea infusion, 20% black tea infusion with 0.5% H_2O_2, 20% black tea decoction and 20% black tea decoction with 0.5% H_2O_2 on body weight of broiler chickens.

Treatments	Initial weight (g)	Final weight (g)	Differences in Weight (g)
Control	257.1 ± 14.1 a	392.9 ± 20.2 a	135.7 ± 12 a
H_2O_2	250 ± 18.9 a	200 ± 18.9 b	- 50 ± 0.0000 b
Black tea infusion	232.1 ± 10.5 a	182.1 ± 10.5 b	- 50 ± 0.0000 b
Black tea infusion + H_2O_2	221.4 ± 8.5 a	171.4 ± 8.5 b	- 50 ± 0.0000 b
Black tea decoction	228.6 ± 17.6 a	178.6 ± 17.6 b	- 50 ± 0.0000 b
Black tea decoction + H_2O_2	248.6 ± 19.6 a	198.6 ± 19.6 b	- 50 ± 0.0000 b

Values represent mean ± SE. Number of animals (7 chickens / group)
Different small letters indicate significant difference at $P \leq 0.05$.

Induction of oxidative stress by 0.5% H_2O_2 treatment revealed no significant change in serum sodium and potassium ions concentration compared with the control group. In addition, different treatments with black tea alone or with H_2O_2 caused no significant change in sodium and potassium concentration except the significant increase in serum sodium concentration in group treated with black tea

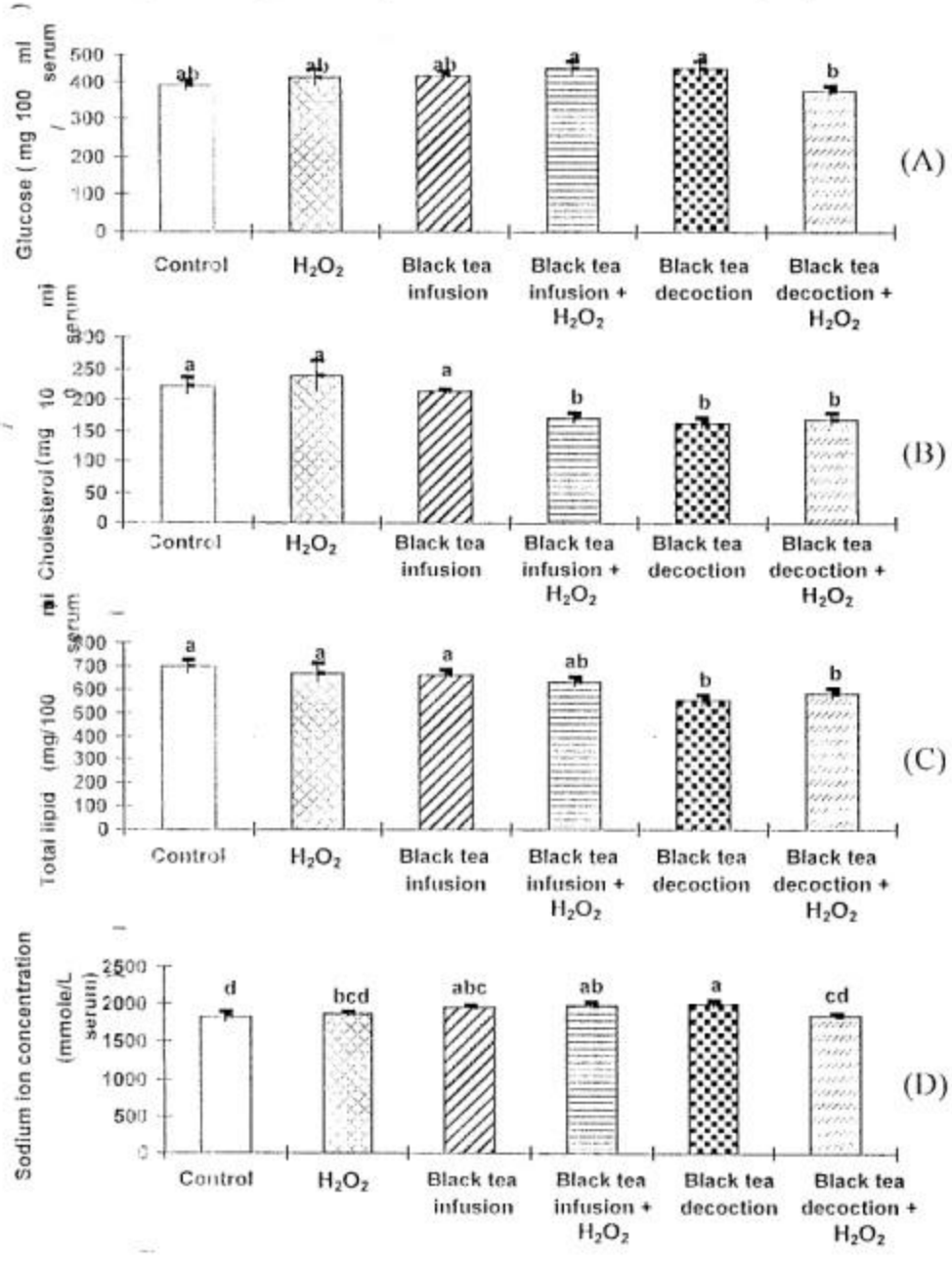
decoction compared with control and H_2O_2 treated groups and black tea decoction with H_2O_2 group (Fig. 1-d, e).

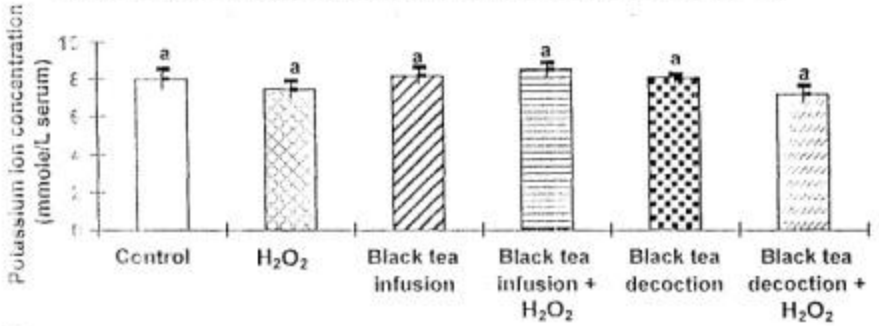
Combined treatment with H_2O_2 and black tea produced no significant changes in ALT and AST concentration compared with control and H_2O_2 treated groups. However, a significant decrease in AST was observed in groups treated with black tea decoction alone or joined with H_2O_2 (Fig. 1-g and h).

Liver glycogen concentration significantly increased in the group treated with 0.5% H_2O_2 compared with control group accompanied with a significant decrease in liver glycogen concentration in other different treatments. (Fig. 1-g)

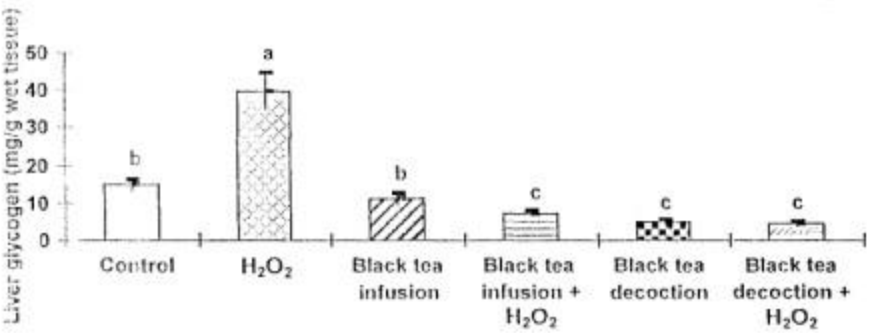
DISCUSSION

The present study demonstrated that H_2O_2-induced oxidative stress resulted in a significant decrease in the body weight. This decrease in the body weight seems to agree with that reported by Aziz in mice (19), Al-Kennany in chickens (20) and Wohaieb *et al.* in rabbits (21). The change in body weight may be due to a decrease in food intake in animals treated with H_2O_2 (20). Abdul-Rahman (22) observed that oral administration of 1% H_2O_2 to male rats caused no change in body weight. The decrease in body weight in groups treated with black tea was in agreement with that reported by Landau *et al.* (15) who observed a decrease in body weight of mice treated with black tea infusion as the sole source of drinking fluid compared with the control group despite that black tea treated animals consumed more food and fluid than control animals. The decrease in body weight of animals treated with black tea in the present study may be due to suppression of the digestive enzymes and this possibility needs to be further investigated. No changes were observed in serum glucose, cholesterol and total lipids in animals treated with H_2O_2 (0.5%) as compared with control group. These observations are in agreement with those obtained by Aziz in rats (19) and Wohaieb *et al.* in rabbits (21). On the other hand, Al-Kennany (20) demonstrated a significant increase in serum cholesterol and total lipids in chickens treated with 0.5% H_2O_2 with drinking water. Moreover, Abdul-Rahman (22) reported a significant decrease in serum glucose level associated with a significant increase in serum cholesterol level in rats administered 1% H_2O_2 in drinking water. Black tea infusion or decoction alone or in combination with H_2O_2 for 3 weeks caused no significant changes in serum glucose level. These results are consistent with those reported by Gomes *et al.* (8) in normal rats. Deng *et al.* (23) noticed that consumed diets with adding black tea and its water

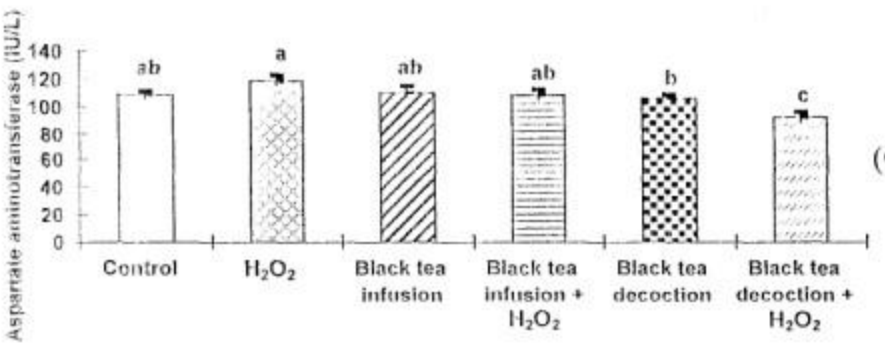

Glucose (mg 100 ml serum)
500
400
300
200
100
0
ab
ab
ab
a
a
b
Control
H2O2
Black tea infusion
Black tea infusion + H2O2
Black tea decoction
Black tea decoction + H2O2
(A)
ml serum
300
250
200
150
100
50
0
a
a
a
b
b
b
Control
H2O2
Black tea infusion
Black tea infusion + H2O2
Black tea decoction
Black tea decoction + H2O2
(B)
Total lipid (mg/100 ml serum)
800
700
600
500
400
300
200
100
0
a
a
a
ab
b
b
Control
H2O2
Black tea infusion
Black tea infusion + H2O2
Black tea decoction
Black tea decoction + H2O2
(C)
Sodium ion concentration (mmole/L serum)
2500
2000
1500
1000
500
0
d
bcd
abc
ab
a
cd
Control
H2O2
Black tea infusion
Black tea infusion + H2O2
Black tea decoction
Black tea decoction + H2O2
(D)


Potassium ion concentration (mmole/L serum)
a
a
a
a
a
a
Control
H2O2
Black tea infusion
Black tea infusion + H2O2
Black tea decoction
Black tea decoction + H2O2

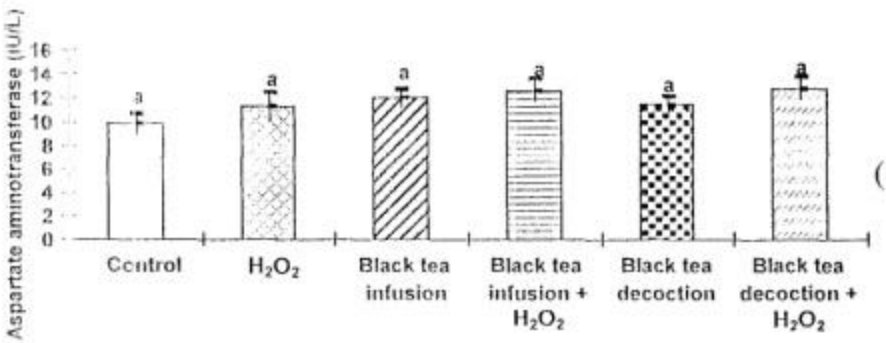
(E)


Liver glycogen (mg/g wet tissue)
50
40
30
20
10
0
b
a
b
c
c
c
Control
H2O2
Black tea infusion
Black tea infusion + H2O2
Black tea decoction
Black tea decoction + H2O2

(F)


Aspartate aminotransferase (IU/L)
140
120
100
80
60
40
20
0
ab
a
ab
ab
b
c
Control
H2O2
Black tea infusion
Black tea infusion + H2O2
Black tea decoction
Black tea decoction + H2O2

(G)


Aspartate aminotransferase (IU/L)
16
14
12
10
8
6
4
2
0
a
a
a
a
a
a
Control
H2O2
Black tea infusion
Black tea infusion + H2O2
Black tea decoction
Black tea decoction + H2O2

(H)

Fig. (1) Effect of three weeks oral administration of 0.5% H_2O_2, 20% black tea infusion, 20% black tea infusion with 0.5% H_2O_2, 20% black tea decoction and 20% black tea decoction with 0.5% H_2O_2 on blood glucose concentration (A), cholesterol concentration (B), Total lipid concentration (C), sodium ion concentration (D), potassium ion concentration (E), Liver glycogen concentration (F), blood AST (G), blood ALT (H), in broiler chickens. The different letters on histograms indicate significant difference at $p \leq 0.05$.

extract decreased blood glucose significantly in rats. Gomes *et al.* (8) showed that black tea administration lowered blood glucose level in streptozotocin diabetic rats. However, the unchanged serum glucose level in the present study may be either due to the short duration (3 weeks) of treatment or species variations, since authors suggest that tea play a role in controlling dietary glucose uptake in the intestinal tract and it possibly contribute to the blood glucose homeostasis, but these possibilities need further studies (24, 25).

In the present study, black tea infusion with H_2O_2 reduced serum cholesterol level, while black tea decoction alone or conjugated with H_2O_2 resulted in a decrease in both cholesterol and total lipid level. The effects of black tea on lipids and lipoprotein were, controversial, some reported a reduction in blood cholesterol while others failed to show any effect. Gangnly (26) noticed that black tea administration for 3 weeks reduced levels of serum cholesterol, triglycerides, low density lipoprotein (LDL) and very low density lipoprotein (VLDL) to some extent in normal rats. Also, black tea had a hypocholesterolemic effect in cholesterol fed rats owing to decreased plasma lipid levels, increased fecal excretion of total lipids and cholesterol (26). Black tea significantly reduced rat's blood triglycerides (23). On the other hand, Chan (28) showed that the hypolipidemic activity of tea epicatechins in hamsters is due to higher fecal excretions of total fatty acids, neutral sterols and acidic sterols or its effect on the rate limiting enzyme of cholesterol biogenesis by binding to the enzyme and by scavenging reactive oxygen species required for monooxygenase reaction (29). However, the effect of tea catechins mixture that reduced cholesterol absorption from rat intestine may be due to the reduction of cholesterol solubility in mixed bile salts micelles (30).

AST is an indicator of liver injury. In the present study, different black tea treatments showed no significant changes in both AST and ALT except black tea decoction alone or combined with H_2O_2 exerted AST lowering effect compared with 0.5% H_2O_2 treated group, this view may be supported by Wada *et al (31), and*

Sugiyama *et al.* (32) who reported that black tea significantly suppressed D-galactosamine-induced enhancement of enzyme activities ALT and AST. The hepatoprotective effect of black tea owing to tea catechins that resulted in the reduction in membrane fluidity as proposed by Tuschiya (33).

Oral administration of 0.5% H_2O_2 alone or combine with different black tea treatments produced no significant changes in serum sodium and potassium concentrations as compared with control group. The mechanism leading to the significant increase in serum sodium concentration following treatment with black tea decoction is unknown. Liver glycogen concentration increased significantly in animals treated for 3 weeks with 0.5% H_2O_2 as compared with control group. Contrary to these results combined treatments of black tea and H_2O_2 was associated with significant decrease in liver glycogen concentration even it returns to the normal level as in the control group. These results indicate that black tea administration can effectively overcome the alteration in liver glycogen concentration caused by H_2O_2 administration.

In conclusion, black tea may have a protective role on some biochemical parameters in broiler exposed to experimental hydrogen peroxide-induced oxidative stress.

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