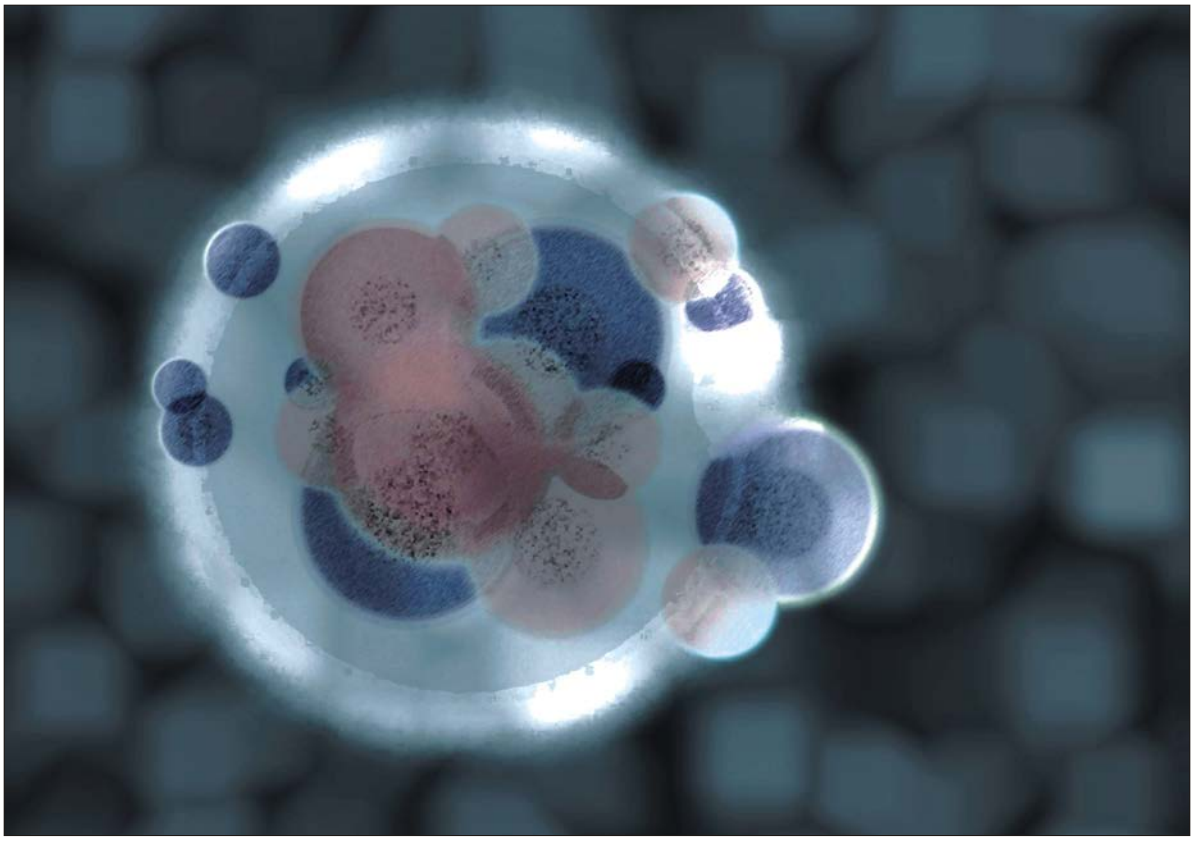

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Research Article
Effect of Aqueous Extract of Green Tea (*Camellia sinensis*) on Hematology and Oxidative Stress Biomarkers in Rats Intoxicated with Carbon Tetrachloride

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Abstract
The present study aimed to investigate the effects of aqueous extract of green tea (*Camellia sinensis*) on selected hematological and chemical biomarkers in rats intoxicated with carbon tetrachloride (CCl_4). Therefore, 20 rats were fed on standard diet and divided into four groups. Rats in the first and second groups were injected i/p with paraffin oil and received either normal water or aqueous extract of green tea, respectively. Rats in the third and fourth groups were injected i/p with CCl_4 and received either normal water or aqueous extract of green tea, respectively. Blood and liver samples were collected at the end of the experiment (5 weeks). Whole blood samples were used for estimation of selected hematological parameters, harvested sera were used for detection of selected biochemical indices and liver tissues were used for estimation of selected oxidative stress biomarkers. Hematological findings revealed that, there was a significant reduction in Total Erythrocytic Count (TEC) and hemoglobin (Hb) concentration with significant increase in Total Leucocytic Count (TLC) in blood of rats that were exposed to CCl_4 without treatment with green tea aqueous extract when compared with control. In addition, CCl_4 elevated serum transaminases, hepatic lipid peroxidation and hepatic enzymatic antioxidants activities. Oral green tea extract attenuated the detrimental effects of CCl_4 and corrected all examined biomarkers toward the control levels. Further works are recommended for the evaluation of the effect of green tea (*Camellia sinensis*) on oxidative stress biomarkers at molecular level.

Key words: Carbon tetrachloride, green tea, antioxidants, physiology, serum, liver

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Carbon tetrachloride (CCl_4) is a highly toxic chemical agent, the most famous drug used to induce liver damage experimentally (Kim *et al*., 2010). Histological sectioning of liver tissues indicated that CCl_4 induced fibrosis, cirrhosis and hepatocarcinoma (Junnila *et al*., 2000). The toxic effect of CCl_4 is attributed to trichloromethyl radical produced during oxidative stress (Stoyanovsky and Cederbaum, 1999). Once the liver became injured, its efficient treatment with famous chemical drugs is limited (Lee *et al*., 2007). Therefore, interest concerned the use of alternative medicines for the treatment of hepatic disease has been arisen. Natural products received great attention as potentially antitoxic and antioxidants agents (Lee *et al*., 2007). Polyphenols found in green tea has received special attention as antioxidant and anti-inflammatory agent (Kuroda and Hara, 1999; Varilek *et al*., 2001). Green tea is produced from the dried leaves of the plant *Camellia sinensis* and contains several polyphenolic components. Most of these polyphenols are flavonols, which is usually called catechins. Epigallocatechin gallate is the most abundant catechin, accounting for about 65% of green tea catechin content and is also the component with the highest antioxidant properties (Guo *et al*., 1996). Previous publications reported many beneficial effects of green tea such as anti-obesity, anticarcinogenic (Zaveri, 2006), hypochlosterolemic (Cooper *et al*., 2005), anti-neurodegenerative (Crespy and Williamson, 2004) and antioxidant (Erba *et al*., 2005) properties. The protective effect of green tea as antitoxic was not investigated so far. Therefore, the present study aimed to investigate the capacity of green tea (*Camellia sinensis*) water extract against CCl_4-induced liver toxicity by assessing hematology, serum biochemistry and hepatic oxidative stress biomarkers.

MATERIALS AND METHODS

Chemicals and kits: Commercial diagnostic kits for glucose (EP37L-660), total proteins (EP56-660), albumin (EP03-570), TAG (EP59-660), cholesterol (EP24-660), ALT (EP07-500), AST (EP15-500), BUN (EP20-420), uric acid (EP61-620), Creatinine (EP33K-660), calcium (EP22-660), phosphorus (EP46-660), magnesium (EP50-660), chloride (EP27-500) were purchased from United Diagnostic Industry, UDI, Dammam, Saudi Arabia. Paraffin oil, carbon tetrachloride (Spectrosol® BHD chemicals Ltd pool, England) and other chemicals and solvents used in the study were of highest grade and commercially available.

Green tea (*Camellia sinensis*) aqueous extract: Green tea extract was made according to Maity *et al.* (1998) by soaking 30 g of green tea powder in 1 L of boiling distilled water for 5 min. The solution was filtered to make 3% green tea extract. This solution was provided to rats as their sole source of drinking water.

Animals and treatment: A total of 20 albino rats (200-250 g) was obtained from Laboratory House of College of Veterinary Medicine and Animal Resources, King Faisal University, Al-Ahsa, Saudi Arabia and acclimated for 10 days before starting the experiment. All animals were housed in standard cages (5 rats/cage), fed with standard laboratory diet and tap water *ad libitum*. The experimental animals were housed in air-conditioned rooms at 21-23°C and 60-65% of relative humidity and kept on a 12 h light/12 h dark cycle. The animals received humane care in accordance with the Guide for the Care and Use of Laboratory Animals, published by ethics of scientific research committee of king Faisal University, Saudi Arabia.

Induction of hepatotoxicity by CCl_4: Liver toxicity was induced by the intraperitoneal injection of CCl_4 (1 mL kg^{-1} b.wt.), 1:1 diluted with paraffin oil, for two successive days of the experiment (Althnaian *et al*., 2013; El-Bahr, 2014; Al-Sultan and El-Bahr, 2015).

Experimental groups and protocol: The rats were divided randomly into 4 groups comprising 5 rats in each group and fed the same diet throughout the experimental period. Group 1: Control rats fed only with diet and tap water and injected i/p with paraffin oil, group 2: Rats injected i/p with paraffin oil and received green tea (Positive control, 3%) as their sole source of drinking water, group 3: Diseased control group intoxicated with CCl_4 (1 mL kg^{-1} b.wt.), 1:1 diluted with paraffin oil in first two days of the experiment and received normal tap water and group 4: Rats were intoxicated with CCl_4 (1 mL kg^{-1} b.wt.), 1:1 diluted with paraffin oil on first two days of the experiment and treated with green tea 3% as their sole source of drinking water.

Samples collection: At the end of the experiment (5 weeks), the overnight fasted rats were anesthetized with diethyl ether. Blood samples were collected by cardiac puncture before incision of the abdomen in plain and EDTA vacutainers. Whole blood was used for the determination of selected hematological parameters. Sera were harvested and stored at -30°C until the time of analysis of serum glucose, total proteins, albumin, triglyceride, total cholesterol, ALT, AST, BUN, uric acid, creatinine, calcium, phosphorus, magnesium and chloride using commercial assay kits according to the manufacture instruction. The liver tissues were removed and

liver fragments were immediately frozen and stored at -80°C for biochemical analysis of selected antioxidant enzymes, namely catalase (CAT) and glutathione peroxidase (GPX).

Hematological analysis: The TEC and TLC were determined by standard hematological techniques (Feldman *et al*., 2000). The Hb% was assessed according to Drabkin (1946).

Analysis of selected biochemical parameters: Serum glucose, total proteins, Albumin, TAG, cholesterol, ALT, AST, BUN, uric acid, creatinine, calcium, phosphorus, magnesium and chloride levels were determined by using commercial kits (United Diagnostic Industry, UDI, Dammam, Saudi Arabia) on ELIPSE full automated chemistry analyzer (Rome, Italy). Concentration of the biochemical constituents was calculated according to the manufacture instruction.

Analysis of hepatic lipid peroxidation and selected hepatic Antioxidant enzymes: The concentration of TBARS (μM, Cayman Chemical Company, USA, Catalog No. 10009055) and the activities of CAT (nmol/min/g tissue; Cayman Chemical Company, USA, Catalog No. 707002) and GPX (nmol/min/g tissue; Cayman Chemical Company, USA, Catalog No.703102) were determined by ELISA reader (Absorbance Microplate Reader ELx 800TM BioTek®, USA). Results were calculated according to the manufacture instructions.

Statistical analysis: All data are presented as the mean±standard error of the mean using one way analysis of variance (ANOVA). All tests were performed using a statistical analysis system program (SAS., 2002).

RESULTS

The effects of CCl_4 administration and the modulatory activities of green tea aqueous extract on the hematological parameters of rats are shown in Table 1. There was a significant reduction ($p<0.05$) in the TEC and Hb concentration in rats that were exposed to CCl_4 without treatment with green tea aqueous extract (group 3) when compared with those of the normal control (group 1). However, there was an increase in these parameters in the rats that were treated with green tea extract (group 2). The TLC increased significantly ($p<0.05$) in rats exposed to CCl_4 when compared to those of the control. However there was a decrease in these parameters in the rats that were treated with green tea extract (group 2).

The activities of AST and ALT were estimated in serum samples as indictors of liver function. These results are given in Table 2. The CCl_4 treatment markedly affected the liver specific enzymes. A significant increase ($p<0.05$) of serum AST and ALT activities was observed in untreated CCl_4 intoxicated rats (group 3) compared to controls (group 1 and 2). This result suggests that these hepatic biomarkers are elevated in the serum due to release of the enzymes from damaged liver. However, a significant decrease ($p<0.05$) was observed in the respective serum activities in rats intoxicated with CCl_4 and treated with aqueous extract of green tea (group 4) compared to untreated CCl_4 intoxicated rats (group 3). The levels of serum kidney biomarkers of rats in all groups showed in Table 2. Serum kidney biomarkers (BUN, uric acid and creatinine) levels remained unchanged in all treated group ($p>0.05$). The profiles of serum protein, lipid and glucose are presented in Table 3. A significant decrease ($p<0.05$) in serum glucose, total proteins and albumin ($p<0.05$) was observed in untreated CCl_4 intoxicated rats (group 3) compared to controls (group 1 and 2). However, administration of aqueous extract of green tea corrected the

Table 1: Effects of CCl_4 and green tea water extract on hematological parameters in rats

	Groups			
Parameters	C	GT	CCl_4	GT+CCl_4
TEC (10^{12} L^{-1})	11.1±0.9	10.8±1.0	8.3±2.0*	10.0±1.0**
TLC (10^9 L^{-1})	10.9±1.0	11.0±0.5	14.2±1.1*	10.2±1.4**
Hb (g dL^{-1})	12.0±1.5	12.1±1.0	8.5±0.7*	11.5±1.0**

Each value represents the mean±standard error of means (SEM) of 5 rats, TEC: Total erythrocyte count, TLC: Total leucocyte count, Hb: Hemoglobin, *Significantly different from control group ($p<0.05$), **Significantly different from CCl_4 group ($p<0.05$), C: Control, GT: Green tea, CCl_4: Carbon tetrachloride

Table 2: Effects of CCl_4 and green tea water extract on serum liver transaminases in rats

	Parameters	
Groups	AST (IU L^{-1})	ALT (IU L^{-1})
C	120.4±4.0	33.5±0.1
GT	131.3±5.0	30.1±1.4
CCl_4	145.0±5.0*	45.0±4.0*
C	115.5±6.0**	35.1±2.9**

Each value represents the mean±standard error of means (SEM) of 5 rats, *Significantly different from control group ($p<0.05$), **Significantly different from CCl_4 group ($p<0.05$), C: Control, GT: Green tea, CCl_4: Carbon tetrachloride

Table 3: Effects of CCl_4 and green tea water extract on kidney biomarkers in rats

	Parameters		
Groups	BUN (mg dL^{-1})	Uric acid (mg dL^{-1})	Creatinine (mg dL^{-1})
C	9.9±0.4	1.5±0.3	0.4±0.04
GT	10.1±0.5	1.3±0.3	0.4±0.03
CCl_4	10.5±1.2	1.8±0.5	0.4±0.02
GT+CCl_4	10.0±1.0	1.7±0.3	0.4±0.05

Each value represents the mean±standard error of means (SEM) of 5 rats, C: Control, GT: Green tea, CCl_4: Carbon tetrachloride, *Significantly different from control group ($p<0.05$)

Table 4: Effects of CCl_4 and green tea water extract on protein, lipids and glucose profiles in rats

	Parameters				
Groups	Glucose (mg dL^{-1})	Total protein (g dL^{-1})	Albumin (g dL^{-1})	TAG (mg dL^{-1})	Cholesterol (mg dL^{-1})
C	320.0±1.9	6.1±1.1	3.7±0.1	60.4±3.0	51.0±2.0
GT	325.5±6.1	6.0±1.0	3.9±0.1	55.3±2.0	52.2±3.0
CCl_4	241.0±0.9*	4.5±0.9*	2.2±0.1*	74.3±5.0*	59.5±1.0*
GT+CCl_4	315.8±7.1**	6.0±1.0**	3.1±0.2**	51.4±5.0**	48.9±6.0**

Each value represents the mean±standard error of means (SEM) of 5 rats, *Significantly different from control group ($p<0.05$), **Significantly different from CCl_4 group ($p<0.05$), C: Control, GT: Green tea, CCl_4: Carbon tetrachloride

Table 5: Effects of CCl_4 and green tea water extract on minerals profile in rats

	Parameters			
Groups	Calcium (mg dL^{-1})	Phosphorus (mg dL^{-1})	Magnesium (mg dL^{-1})	Chloride (mEq L^{-1})
C	8.9±1.1	2.5±0.7	3.7±0.3	40.1±4.0
GT	10.0±2.0	2.4±0.9	3.5±0.3	44.2±3.0
CCl4	9.0±1.1	2.1±0.2	3.0±0.3	41.0±2.5
GT+CCl_4	9.9±1.0	2.5±0.3	3.7±0.3	43.7±2.0

Each value represents the mean±standard error of means (SEM) of 5 rats, C: Control, GT: Green tea, CCl_4: Carbon tetrachloride

values of glucose and total protein towards the control values in CCl_4 intoxicated rats. A significant increase ($p<0.05$) in serum cholesterol and triglyceride was observed in untreated CCl_4 intoxicated rats (group 3) compared to controls (group 1 and 2) (Table 4). However, administration of aqueous extract of green tea corrected the values of glucose and total protein towards the control values in CCl_4 intoxicated rats. A significant change was not observed in calcium, phosphorus, magnesium and chloride levels of control and all treated groups ($p>0.05$) throughout the experimental period. Results in Table 5 shows that the TBARS level was significantly ($p<0.05$) increased in the liver of CCl_4 intoxicated compare to the controls. Treatment of CCl_4 intoxicated rats with aqueous extract of green tea caused significant ($p<0.05$) decrease in TBARS level of liver tissue compared to that in liver from untreated CCl_4 intoxicated rats. Administration of aqueous extract of green tea only did not affect significantly ($p>0.05$) the level of TABARS in rat liver compare to the control. The results that presented in the same table (Table 5) indicated that, All the antioxidant enzyme activities were reduced significantly ($p<0.05$) in CCl_4 intoxicated rats compare to the control. However, all activities of these enzymes showed a significant ($p<0.05$) recovery in response to administration of aqueous extract of green tea to CCl_4 intoxicated rats compare to untreated CCl_4 intoxicated rats. The activities of hepatic antioxidant enzymes of rats treated only with aqueous extract of green tea were comparable to that of the control (Table 5).

DISCUSSION

The current study demonstrated clearly that, CCl_4 administration produced microcytic hypochromic anemia as evidenced by the reduction in the RBC and Hb concentration. Similar result was reported in rats (Saba *et al*., 2010). The current findings indicated that, CCl_4 administration induced leucocytosis. The reduction in the formed elements in the blood and leucocytosis are stress induced phenomenon (Saba *et al*., 2010). Acute stress in animals has been widely reported to be associated with increased white blood cell count occasioned by significant increase in the neutrophil count and the neutrophil/lymphocyte ratio (Larson *et al*., 1985; Huff *et al*., 2005). Previous study on the effects of CCl_4 on haematological parameters showed that acute CCl_4 toxicity led to transient decrease in the Hb concentration and RBC counts (Mortiz and Pankow, 1989). Parallel to the current findings, a significant increase of Hb and TEC was observed in rats administered green tea (El-Kott and Bin-Meferij, 2008; Gad and Zaghloul, 2013). In the same way of the current findings, administration of different concentrations of green tea produced significant decrease of TLC and significant increase of Hb and TEC in rats (Al-Shawi, 2014).

In the present study serum hepatic biomarkers, AST and ALT activities were greatly increased in untreated CCl_4 intoxicated rats compared to control animals. The increased serum levels of hepatic transaminases biomarkers have been attributed to the liver injury, because these enzymes are place in cytoplasmic area of the cell and are released into circulation in case of cellular damage (Brent and Rumack, 1993; Recknagel *et al*., 1989). The CCl_4 induced the increase of serum ALT and AST levels which source from cell membrane and mitochondrial damages in liver cells (Zimmerman *et al*., 1965; El-Bahr, 2014; Al-Sultan and El-Bahr, 2015). Many publications rep orted that, hepatic transaminases enzymes activities (ALT and AST) were significantly elevated after CCl_4 treatment

Table 6: Effects of CCl_4 and green tea water extract on lipid peroxidation and oxidative stress biomarkers in rats

	Parameters		
Groups	TBARS (μmol g^{-1} tissue)	CAT (nmol/min/g tissue)	GPX (nmol/min/g tissue)
C	25.5±0.5	30.1±1.0	312.5±5.6
GT	26.6±1.0	31.2±2.0	301.4±6.0
CCl4	37.8±1.0*	22.3±2.3*	250.7±9.9*
GT+CCl_4	28.1±4.1**	29.0±2.1**	300.8±9.0**

Each value represents the mean±standard error of means (SEM) of 5 rats, *Significantly different from control group ($p<0.05$), ** Significantly different from CCl_4 group ($p<0.05$), C: Control, GT: Green tea, CCl_4: Carbon tetrachloride, TBARS: Lipid peroxidation biomarke, CAT: Catalase, GPX: Glutatnione peroxidase

(Arici and Cetin, 2011; Althnaian *et al*., 2013; Das, 2014; El-Bahr, 2014; Al-Sultan and El-Bahr, 2015). The hepatotoxic effects of CCl_4 were attributed to lipid peroxidation caused by its active metabolite CCl_3 radicals (Park *et al*., 2005). These activated radicals bind covalently to the macromolecules and induce peroxidative degradation of membrane lipids of endoplasmic reticulum which are rich in polyunsaturated fatty acids. This leads to formation of lipid peroxides, which in turn gives products like melanodialdehyde (MDA) that cause damage to the membrane. This lipid peroxidative degradation of biomembrane is one of the principle causes of hepatotoxicity of CCl_4 (Park *et al*., 2005). On the other hand, current findings indicated that, the increase in serum AST and ALT activities induced in untreated CCl_4 intoxicated rats was significantly corrected towards normal control values when these rats treated with aqueous extract of green tea. This finding implies that, green tea aqueous extract challenge to protect liver tissue from CCl_4 injury. The reversal of increased serum enzymes in CCl_4-induced liver damage by green tea aqueous extract may be due to the prevention of the leakage of intracellular enzymes by its membrane stabilizing activity (Issabeagloo *et al*., 2012). This is in agreement with the commonly accepted view that serum levels of transaminases return to normal with the healing of hepatic parenchyma and the regeneration of hepatocytes (Thabrew *et al*., 1987). Similar findings have provided a considerable support for evidencing the protective effects of green tea aqueous extract on liver damage at the level of tansaminases enzymes (Almurshed, 2006; Hashimoto *et al*., 2007; Sengottuvelu *et al*., 2008; Issabeagloo *et al*., 2012). Green tea aqueous extract decreased CCl_4 induced elevated enzyme levels in tested groups, indicating the protection of structural integrity of hepatocytic cell membrane or regeneration of damaged liver cells (Palanivel *et al*., 2008). It has been demonstrated that, the protective effect of green tea aqueous extract against CCl_4-induced oxidative stress in rats was due to its antioxidant properties (Sengottuvelu *et al*., 2008). The antioxidant activity of green tea could be attributed to the presence of epicatechins which act as scavengers of reactive oxygen species (El-Beshbishy, 2005; Issabeagloo *et al*., 2012). The antioxidant activity of green tea aqueous extract has been approved in the current study as reflected on significant reduction of lipid peroxidation biomarker (TBARS) and significant elevation of antioxidant enzymes activities (CAT and GPX) in liver of CCL_4 intoxicated rats (Table 6). As in the current experiment, previous experimental studies have shown that $CC1_4$ decreased total protein (Fahim *et al*., 1999; Khan and Alzohairy, 2011; Althnaian *et al*., 2013) and albumin (Fahim *et al*., 1999; Khan and Alzohairy, 2011; Althnaian *et al*., 2013) levels. However, there is a controversy about the effect of $CC1_4$ on serum creatinine level. While, some investigator (Cruz *et al*., 1993) found a decrease in serum creatinine in CCl_4 toxicity, parallel to the present study others (Palaparthy *et al*., 2001; Wirth *et al*., 1997; Khan and Alzohairy, 2011; Althnaian *et al*., 2013) found no significant changes (Table 2). The restoration of total protein and albumin in serum of rats intoxicated with CCl_4 and treated with green tea aqueous extract compared to untreated CCl_4 intoxicated rats might attributed to the antioxidant properties of green tea which improves organ functions (El-Beshbishy, 2005). The enhancement of protein biosynthesis may accelerates the regeneration process and protect the liver cells against CCl_4 toxicity.

The current study reported that, calcium, phosphorus, chloride and magnesium values in $CC1_4$ administrated rats were not statistically different from control values (Ogeturk *et al*., 2004).

In the present study, serum glucose was reduced in CCl_4-treated rats which were prevented by green tea aqueous extract. Studies have demonstrated decreased hepatic glycogen content after treatment with CCl_4, reflecting decreased gluconeogenesis by the liver Muriel *et al*. (2001). It has been known that, hypoglycemia is main feature of carbon tetrachlorid toxicity (Mion *et al*., 1996). The same author reported that liver cirrhotic rats exhibited hypoglycemia. The normoglycemic (antihyperglycemic) effect of green tea extract may be due to the fact that, green tea extract contains polyphenols namely catechin, epicatechin, epigallocatechin and their gallates (Kasahara and Kato, 2003; Haidari *et al*., 2012). Several mechanisms have been suggested for

antihyperglycemic effect of green tea that include enhancing insulin-stimulated glucose uptake, suppressing glucose absorption by sodium dependent glucose transporter (Babu *et al*., 2006a), suppressing gluconeogenesis by decreasing the expression some genes such as phosphoenolpyrovate carboxykinase and glucose-6-phosphotase (Waltner-Law *et al*., 2002) and ameliorating insulin resistance by increasing expression of glucose transporter IV (Wu *et al*., 2004).

The results of the present study have also established that the CCl_4 treatment could have affected the lipid metabolism of liver (triglyceride and cholesterol levels). This is evidenced from the present observations that, CCl_4 was caused a significant increase in the levels of lipid parameters. Muller *et al.* (1974) stated that CCl_4 intoxication is similar the hepatitis in case of the triglycerides catabolism. This situation could be also attributed to the reduction of lipase activity, which could lead to decrease in triglyceride hydrolysis (Jahn *et al*., 1985). On the other hand, it can be assumed that hypercholesterolemia in CCl_4 intoxicated rats has resulted in damage of hepatic parenchymal cells that lead to disturbance of lipid metabolism in liver (Havel*,* 1986). When CCl_4 intoxicated rats treated with green tea aqueous extract, a significant decline in above mentioned parameters (triacylglycerol and cholesterol) has been observed compare with untreated CCl_4-intoxicated rats. Similar work demonstrated the hypocholesterolemic effect of green tea in rats (Babu *et al*., 2006b; Haidari *et al*., 2012). The hypolipidemic effect of green tea observed in the current study agrees with previous work (Velayutham *et al*., 2008; Amanolahi and Rakhshande, 2013). It has been reported that, catechins contents of green tea are responsible for the improvement of lipid profile of treated animals (Koo and Noh, 2007). The possible mechanisms by which green tea extract can exert cholesterol lowering effect are reducing the absorption of dietary and biliary cholesterol and promoting its fecal excretion (Koo and Noh, 2007). Furthermore, recent study (Singh *et al*., 2009) indicated that, green tea extract decreased the synthesis of cholesterol in cultured rat hepatoma cells.

The significant increase of TBARS in liver of untreated CCl_4 intoxicated rats suggests enhanced peroxidation leading to tissue damage and the failure of the antioxidant mechanisms in preventing of excessive free radicals (Romero-Alvira and Roche, 1996). This confirmed by the results of the current study which reported a significant decrease of activities of antioxidant enzymes (CAT and GPX) in CCl_4 intoxicated rats compare to controls which impose in elevation of TBARS levels. Presumably a decrease in CAT activity could be attributed to cross linking and inactivation of the enzyme protein in the lipid peroxides. Decreased CAT activity is linked to exhaustion of the enzyme as a result of oxidative stress caused by CCl_4. These findings were found to be in agreement with other previous studies in rats liver intoxicated with CCl_4 (Yousef, 2004; Aranda *et al*., 2010; Al-Fartosi *et al*., 2012; Bona *et al*., 2012; Pirinccioglu *et al*., 2012; El-Bahr, 2014). The significant decrease of TBARS in liver of rats intoxicated with CCl_4 and treated with green tea aqueous extract compare to untreated CCl_4 intoxicated rats suggested the antioxidant effect of green tea aqueous extract against CCl_4-induced oxidative stress and agrees with previous results in rats (Sengottuvelu *et al*., 2008).

The protective effect of green tea aqueous extract was augmented by increasing the activities of antioxidant enzymes (CAT and GPX) when administered to CCl_4 intoxicated rats. Consistent to the current findings, stimulation of antioxidant enzymes activities by green tea has been documented previously (Sengottuvelu *et al*., 2008). Epicatechins contents of green tea may be responsible for the antioxidant enzyme activation and subsequent scavenging of free radical species (El-Beshbishy, 2005).

CONCLUSION

Administration of CCl_4 to rats induced elevation of hepatic transaminases, lipid peroxidation and antioxidant enzymes activities. Oral green tea aqueous extract attenuated the detrimental effects of CCl_4 and corrected all examined biomarkers toward the control values. The ameliorative effects of green tea aqueous extract were in the form of improving of serum transaminases, attenuation of hepatic lipid peroxidation and activation of hepatic antioxidant enzymes. Further work is suggested for the evaluation of the effect of green tea (*Camellia sinensis*) on oxidative stress biomarkers at molecular level.

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REFERENCES

Al-Fartosi, K.G., A. Majid, M.A. Auda and M.H. Hussein, 2012. The role of camel's milk against some oxidant-antioxidant markers of male rats treated with CCl_4. Int. J. Res. Pharmaceut. Biomed. Sci., 3: 385-389.

Al-Shawi, N.N., 2014. Impacts of different concentrations of aqueous green tea extract administered during methotrexate treatment on some selected blood indices in rats. Int. J. Pharm. Pharmaceut. Sci., 6: 175-178.

Al-Sultan, S.I. and S.M. El-Bahr, 2015. Effect of aqueous extract of fenugreek (*Trigonella foenum-graecum* L.) on selected biochemical and oxidative stress biomarkers in rats intoxicated with carbon tetrachloride. Int. J. Pharmacol., 11: 43-49.

Almurshed, K.S., 2006. Protective effect of black and green tea against carbon tetrachloride-induced oxidative stress in rats. Saudi Med. J., 27: 1804-1809.

Althnaian, T., I., Albokhadaim and S.M. El-Bahr, 2013. Biochemical and histopathological study in rats intoxicated with carbontetrachloride and treated with camel milk. Springer Plus, Vol. 2. 10.1186/2193-1801-2-57

Amanolahi, F. and H. Rakhshande, 2013. Effects of ethanolic extract of green tea on decreasing the level of lipid profile in rat. Avicenna J. Phytomed., 3: 98-105.

Aranda, M., C.D. Albendea, F. Lostale, L. Lopez-Pingarron and L. Fuentes-Broto *et al*., 2010. *In vivo* hepatic oxidative stress because of carbon tetrachloride toxicity: Protection by melatonin and pinoline. J. Pineal Res., 49: 78-85.

Arici, O.F. and N. Cetin, 2011. Protective role of ghrelin against carbon tetrachloride (CCl_4)-induced coagulation disturbances in rats. Regul. Pept., 166: 139-142.

Babu, P.V.A., K.E. Sabitha and C.S. Shyamaladevi, 2006a. Green tea impedes dyslipidemia, lipid peroxidation, protein glycation and ameliorates Ca^{2+}-ATPase and Na^{+}/K^{+}-ATPase activity in the heart of streptozotocin-diabetic rats. Chemico-Biol. Interact., 162: 157-164.

Babu, P.V.A., K.E. Sabitha and C.S. Shyamaladevi, 2006b. Therapeutic effect of green tea extract on oxidative stress in aorta and heart of streptozotocin diabetic rats. Chemico-Biol. Interact., 162: 114-120.

Bona, S., L.I. Filippin, F.C. Di Naso, C. de David and B. Valiatti *et al*., 2012. Effect of antioxidant treatment on fibrogenesis in rats with carbon tetrachloride-induced cirrhosis. ISRN Gastroenterol. 10.5402/2012/762920

Brent, J.A. and B.H. Rumack, 1993. Role of free radicals in toxic hepatic injury II. Are free radicals the cause of toxin-induced liver injury? J. Toxicol.: Clin. Toxicol., 31: 173-196.

Cooper, R., D.J. Morre and D.M. Morre, 2005. Medicinal benefits of green tea: Part I. Review of noncancer health benefits. J. Altern. Complement. Med., 11: 521-528.

Crespy, V. and G. Williamson, 2004. A review of the health effects of green tea catechins in *in vivo* animal models. J. Nutr., 134: 3431S-3440S.

Cruz, C., M.E. Ibarra-Rubio and J. Pedraza-Chaverri, 1993. Circulating levels of active, total and inactive renin (prorenin), angiotensin I and angiotensinogen in carbon tetrachloride-treated rats. Clin. Exp. Pharmacol. Physiol., 20: 83-88.

Das, S., 2014. Hepatoprotective activity of methanol extract of *Fenugreek* seeds on rats. Int. J. Pharmaceut. Sci. Res., 5: 1506-1513.

Drabkin, D., 1946. Spectrophotometric studies XIV. The crystallographic and optical properties of the hemoglobin of man in comparison with those of other species. J. Biol. Chem., 164: 703-723.

El-Bahr, S.M., 2014. Camel milk regulates gene expression and activities of hepatic antioxidant enzymes in rats intoxicated with carbon tetrachloride. Asian J. Biochem., 9: 30-40.

El-Beshbishy, H.A., 2005. Hepatoprotective effect of green tea (*Camellia sinensis*) extract against tamoxifen-induced liver injury in rats. J. Biochem. Mol. Biol., 38: 563-570.

El-Kott, A.F. and M.M. Bin-Meferij, 2008. Influence of green tea on haematological and lung histological disorders induced by malathion in rats. Res. J. Environ. Toxicol., 2: 85-91.

Erba, D., P. Riso, A. Bordon, P. Foti, P.L. Biagi and G. Testolin, 2005. Effectiveness of moderate green tea consumption on antioxidative status and plasma lipid profile in humans. J. Nutr. Biochem., 16: 144-149.

Fahim, F.A., A.Y. Esmat, H.M. Fadel and K.F. Hassan, 1999. Allied studies on the effect of *Rosmarinus officinalis* L. on experimental hepatotoxicity and mutagenesis. Int. Food Sci. Nutr., 50: 413-427.

Feldman, B.F., J.G. Zinkl and N.C. Jain, 2000. Schalm's Veterinary Hematology. 5th Edn., John Wiley and Sons, New York, USA., ISBN-13: 9780683306927, pp: 1085-1088.

Gad, S.B. and D.M. Zaghloul, 2013. Beneficial effects of green tea extract on liver and kidney functions, ultrastructure, lipid profile and hematological parameters in aged male rats. Global Veterinaria, 11: 191-205.

Guo, Q., B. Zhao, M. Li, S. Shen and W. Xin, 1996. Studies on protective mechanisms of four components of green tea polyphenols against lipid peroxidation in synaptosomes. Biochimica Biophysica Acta (BBA)-Lipids Lipid Metab., 1304: 210-222.

Haidari, F., M.M. Shahi, M. Zarei, H. Rafiei and K. Omidian, 2012. Effect of green tea extract on body weight, serum glucose and lipid profile in streptozotocin-induced diabetic rats. A dose response study. Saudi Med. J., 33: 128-133.

Hashimoto, T., M. Goto, H. Sakakibara, N. Oi, M. Okamoto and K. Kanazawa, 2007. Yellow tea is more potent than other types of tea in suppressing liver toxicity induced by carbon tetrachloride in rats. Phytother. Res., 21: 668-670.

Havel, R.J., 1986. Functional activities of hepatic lipoprotein receptors. Annu. Rev. Physiol., 48: 119-134.

Huff, G.R., W.E. Huff, J.M. Balog, N.C. Rath, N.B. Anthony and K.E. Nestor, 2005. Stress response differences and disease susceptibility reflected by heterophil to lymphocyte ratio in turkeys selected for increased body weight. Poult. Sci., 84: 709-717.

Issabeagloo, E., F. Ahmadpoor, P. Kermanizadeh and M. Taghizadiyeh, 2012. Hepatoprotective effect of green tea on hepatic injury due to leflunomide in rat. Asian J. Exp. Biol. Sci., 3: 136-141.

Jahn, C.E., E.J. Schaefer, L.A. Taam, J.H. Hoofnagle and F.T. Lindgren *et al*., 1985. Lipoprotein abnormalities in primary biliary cirrhosis. Association with hepatic lipase inhibition as well as altered cholesterol esterification. Gastroenterology, 89: 1266-1278.

Junnila, M., T. Rahko, A. Sukura and L.A. Lindberg, 2000. Reduction of carbon tetrachloride-induced hepatotoxic effects by oral administration of betaine in male Han-Wistar rats: A morphometric histological study. Vet. Pathol., 37: 231-238.

Kasahara, T. and T. Kato, 2003. Nutritional biochemistry: A new redox-cofactor vitamin for mammals. Nature, 422: 832-832.

Khan, A.A. and M.A. Alzohairy, 2011. Hepatoprotective effects of camel milk against CCl_4-induced hepatotoxicity in rats. Asian J. Biochem., 6: 171-180.

Kim, H.Y., J.K. Kim, J.H. Choi, J.Y. Jung and W.Y. Oh *et al*., 2010. Hepatoprotective effect of pinoresinol on carbon tetrachloride-induced hepatic damage in mice. J. Pharmacol. Sci., 112: 105-112.

Koo, S.I. and S.K. Noh, 2007. Green tea as inhibitor of the intestinal absorption of lipids: Potential mechanism for its lipid-lowering effect. J. Nutr. Biochem., 18: 179-183.

Kuroda, Y. and Y. Hara, 1999. Antimutagenic and anticarcinogenic activity of tea polyphenols. Mutat. Res./Rev. Mutat. Res., 436: 69-97.

Larson, C.T., W.B. Gross and J.W. Davis, 1985. Social stress and resistance of chicken and swine to *Staphylococcus aureus* challenge infections. Can. J. Comp. Med., 49: 208-210.

Lee, C.H., S.W. Park, Y.S. Kim, S.S. Kang, J.A. Kim, S.H. Lee and S.M. Lee, 2007. Protective mechanism of glycyrrhizin on acute liver injury induced by carbon tetrachloride in mice. Biol. Pharmaceut. Bull., 30: 1898-1904.

Maity, S., J.R. Vedasiromoni and D.K. Ganguly, 1998. Role of glutathione in the antiulcer effect of hot water extract of black tea (*Camellia sinensis*). Jpn. J. Pharmacol., 78: 285-292.

Mion, F., A. Geloen, E. Agosto and Y. Minaire, 1996. Carbon tetrachloride-induced cirrhosis in rats: Influence of the acute effects of the toxin on glucose metabolism. Hepatology, 23: 582-588.

Mortiz, R.P. and D. Pankow, 1989. [Effect of carbon tetrachloride and chloroform on hematologic parameters in rats]. Folia Haematologica, 116: 283-287, (In German).

Muller, P., R. Fellin, J. Lambrecht, B. Agostini, H. Wieland, W. Rost and D. Seidel, 1974. Hypertriglyceridaemia secondary to liver disease. Eur. J. Clin. Invest., 4: 419-428.

Muriel, P., N. Alba, V.M. Perez-Alvarez, M. Shibayama and V.K. Tsutsumi, 2001. Kupffer cells inhibition prevents hepatic lipid peroxidation and damage induced by carbon tetrachloride. Comp. Biochem. Physiol. Part C: Toxicol. Pharmacol., 130: 219-226.

Ogeturk, M., I. Kus, A. Kavakli, I. Zararsiz, N. Ilhan and M. Sarsilmaz, 2004. Effects of melatonin on carbon tetrachloride-induced changes in rat serum. J. Physiol. Biochem., 60: 205-210.

Palanivel, M.G., B. Rajkapoor, R. Senthil Kumar, J.W. Einstein and E.P. Kumar *et al*., 2008. Hepatoprotective and antioxidant effect of *Pisonia aculeata* L. against CCl_4-induced hepatic damage in rats. Scientia Pharmaceutica, 76: 203-215.

Palaparthy, R., H. Kastrissios and A. Gulati, 2001. Pharmacokinetics of diaspirin cross-linked haemoglobin in a rat model of hepatic cirrhosis. J. Pharm. Pharmacol., 53: 179-185.

Park, W.H., S.K. Lee and C.H. Kim, 2005. A Korean herbal medicine, *Panax notoginseng*, prevents liver fibrosis and hepatic microvascular dysfunction in rats. Life Sci., 76: 1675-1690.

Pirinccioglu, M., G. Kizil, M. Kizil, G. Ozdemir, Z. Kanay and M.A. Ketani, 2012. Protective effect of Okuzgozu (*Vitis vinifera* L. cv.) grape juice against carbon tetrachloride induced oxidative stress in rats. Food Funct., 3: 668-673.

Recknagel, R.O., E.A. Glende Jr., J.A. Dolak and R.L. Waller, 1989. Mechanisms of carbon tetrachloride toxicity. Pharmacol. Ther., 43: 139-154.

Romero-Alvira, D. and E. Roche, 1996. High blood pressure, oxygen radicals and antioxidants: Etiological relationships. Med. Hyphothesis, 46: 414-420.

SAS., 2002. Statistical Analysis System, Users Guide, SAS/STAT Version. 8th Edn., SAS Institute, Inc., Cary, North Carolina, USA.

Saba, A.B., A.A. Oyagbemi and O.I. Azeez, 2010. Amelioration of carbon tetrachloride-induced hepatotoxicity and haemotoxicity by aqueous leaf extract of *Cnidoscolus aconitifolius* in rats. Niger. J. Physiol. Sci., 25: 139-147.

Sengottuvelu, S., S. Duraisami, J. Nandhakumar, R. Duraisami and M. Vasudevan, 2008. Hepatoprotective activity of *Camellia sinensis* and its possible mechanism of action. Iran. J. Pharmacol. Therapeut., 7: 9-14.

Singh, D.K., S. Banerjee and T.D. Porter, 2009. Green and black tea extracts inhibit HMG-CoA reductase and activate AMP kinase to decrease cholesterol synthesis in hepatoma cells. J. Nutr. Biochem., 20: 816-822.

Stoyanovsky, D.A. and A.I. Cederbaum, 1999. Metabolism of carbon tetrachloride to trichloromethyl radical: An ESR and HPLC-EC study. Chem. Res. Toxicol., 12: 730-736.

Thabrew, M.I., P.D.T.M. Joice and W. Rajatissa, 1987. A comparative study of the efficacy of *Pavetta indica* and *Osbeckia octandra* in the treatment of liver dysfunction. Planta Medica, 53: 239-241.

Varilek, G.W., F. Yang, E.Y. Lee, W.J.S. deVilliers and J. Zhong *et al*., 2001. Green tea polyphenol extract attenuates inflammation in interleukin-2-deficient mice, a model of autoimmunity. J. Nutr., 131: 2034-2039.

Velayutham, P., A. Babu and D. Liu, 2008. Green tea catechins and cardiovascular health: An update. Curr. Med. Chem., 15: 1840-1850.

Waltner-Law, M.E., L.X. Wang, B.K. Law, R. Hal, M. Nawano and D.K. Granner, 2002. Epigallocatechin gallate, a constituent of green tea, represses hepatic glucose production. J. Biol. Chem., 277: 34933-34940.

Wirth, K.J., M. Bickel, M. Hropot, V. Gunzler, H. Heitsch, D. Ruppert and B.A. Scholkens, 1997. The bradykinin B_2 receptor antagonist icatibant (HOE 140) corrects avid Na^+ retention in rats with CCl_4-induced liver cirrhosis: Possible role of enhanced microvascular leakage. Eur. J. Pharmacol., 337: 45-53.

Wu, L.Y., C.C. Juan, L.S. Hwang, Y.P. Hsu, P.H. Ho and L.T. Ho, 2004. Green tea supplementation ameliorates insulin resistance and increases glucose transporter IV content in a fructose-fed rat model. Eur. J. Nutr., 43: 116-124.

Yousef, M.I., 2004. Aluminum-induced changes in hemato-biochemical parameters, lipid peroxidation and enzyme activities of male rabbits: Protective role of ascorbic acid. Toxicology, 199: 47-57.

Zaveri, N.T., 2006. Green tea and its polyphenolic catechins: Medicinal uses in cancer and noncancer applications. Life Sci., 78: 2073-2080.

Zimmerman, H.J., Y. Kodera and M. West, 1965. Effects of carbon tetrachloride poisoning on the plasma levels of cytoplasmic and mitochondrial enzymes in animals with nutritional fatty metamorphosis. J. Lab. Clin. Med., 66: 324-333.