The Effect of Some Beverage Rich in Chlorogenic acid on Rats suffering from Chronic Liver Damage Induced by Carbon Tetrachloride

Usama El-Sayd, Hanan ELghadour, Nahed Mohamed Hussein, Alaa Tarek Hosney

Abstract

Chronic liver disease refers to on-going inflammation in the liver for at least 6 months with the potential to progress to cirrhosis and end-stage liver disease. In the present study, the effect of some beverage rich in chlorogenic acid on rats suffering from chronic liver induced by carbon tetrachloride (CCl4) was studied in Male albino rats. Forty-two rats were divided into six groups. Group 1: negative control group (normal, fed on basal diet), Group 2: positive group, injected with CCl4 (fed on basal diet). Groups 3, 4, 5 and 6 were injected with CCl4 co-administered with green coffee, roasted coffee, green tea and black tea. The results showed that the administration of CCl4 resulted in significant increase in liver enzyme, in addition, CCl4 disturbed liver histology. There was a statistically significant difference between the positive control group when compared to green coffee and green tea groups (192.33 ± 6.83 g vs. 193.67 ± 7.50 g, respectively) in the final weight. Beverage extract has a beneficial effect on serum glucose, lipid profile, kidney, and liver function. Histopathological examination indicated that groups administered with green tea, roasted coffee have normal histological structure.

Keywords: Chronic liver disease (CLD), (Carbon Tetrachloride) CCL, (4 Chlorogenic acid (CGA), green coffee, roasted coffee, green tea, black tea, kidney, liver function, HDL-c, LDL-c and VLDL-c.

Introduction

Nowadays human nutrition science and medicine look at food not only for its function of nourishing and satisfying human senses but also as tool to help people to maintain a healthy status preventing the risk of many common pathologies and thus reducing the costs of public healthcare. Recent evidence highlights that a dietary approach that is constituted by the increase of fruit and vegetable as well as the reduction of
alcoholic beverage and of fat-rich food consumption also for patients with liver diseases may be counselled.1

Chronic liver disease (CLD) refers to on-going inflammation in the liver for at least 6 months with the potential to progress to cirrhosis and end-stage liver disease. It involves a spectrum of conditions that includes chronic hepatitis and cirrhosis. The underlying etiology of CLD is diverse; common causes include hepatitis B and C infection, alcohol abuse, hepatotoxic drugs and non-alcoholic steatohepatitis.2 Recently, a major role of fats and oxidative stress has been shown in liver disease pathogenesis.3 Liver diseases include a number of causes, such as parasites, infections, deficiency of certain nutrients, inborn errors, toxic agents, and malignancy. Viral hepatitis is the leading cause of liver disease in Egypt.4 Egypt has launched a national treatment program to provide cures for hepatitis C virus-infected patients, aimed at eradicating viral hepatitis by 2030.5

Toxin-induced hepatic injury are caused by certain antibiotic, chemotherapeutic agents, carbon tetrachloride (CCL₄), thioacetamide (TAA), D-galactosamine, paracetamol, heavy alcohol consumption and microbes.6 CCl₄ is commonly used in animal models to cause hepatic fibrosis and cirrhosis.4 There is accumulating experimental evidence that chlorogenic acid (CGA) has a variety of pharmacological actions, including anti-inflammatory, anti-oxidative and anti-carcinogenic.7

One of the most available phenolic acid compounds in foods, such as coffee and tea, is CGA. Many polyphenols, especially CGAs, which are considered well-known antioxidant agents, are isolated from coffee. Caffeoylquinic acid (CQA) is characterized as one of the main polyphenols contained in the coffee abundance families of Asteraceae and Lamiaceae, which is commonly known as CGA and is also a quinic acid ester.8

Dietary and beverages play an important role in the prevention of chronic diseases.9 Coffee is the most popular
beverage around the world. Coffee drinking is closely related to human health. Long-term research studies show that the bioactivity of coffee is closely related to CGAs, caffeine, trigonelline, and coffee melanoidins. (11) Recently, the generally beneficial effects of coffee consumption on reducing cause-specific and total mortality have attracted much attention. The increasing evidence has reported that coffee consumption has protective effects on liver disease progression and development. (12)

Tea is one of the most widely popular beverages worldwide, categorized into three types, based on the level of fermentation, i.e., green (unfermented), oolong (partially fermented) and black (fermented) tea. Interestingly, the use of tea extracts as dietary supplements arises from the perception that some tea compounds have beneficial protective effects against chronic diseases. (13)

This study aimed at evaluating the effect of some beverage rich in chlorogenic acid such as green coffee, roasted coffee, green tea and black tea on serum glucose, liver function, kidney function and serum lipoproteins of rats suffering from chronic liver damage induced by carbon tetrachloride.

**Material and methods:**

**Material**

(1) Green coffee, roasted coffee, green tea and black tea were obtained from local market, Cairo government, Egypt.

(2) Casein, vitamins, minerals, cellulose and choline chloride were purchased from Sigma Company, Cairo, Egypt.

(3) CCl₄ purchased from Sigma Company, Egypt. The storage and use of CCl₄ was in accordance with the institutional chemical safety guidelines.

(4) Kits used to determine serum AST, ALT, ALP, total cholesterol, triglycerides, VLDL-c, uric acid, urea nitrogen and glucose were obtained from Sigma Company, Cairo, Egypt.
Methods

Experiment design

Forty-two male albino rats weighing 150±10 g used in the present investigation. The animals were provided by the animal house colony of the National Research Center, Dokki, Cairo, Egypt. Rats were housed 7 per cage with wood shave bedding and were fed standard basal diet as reported by (14) and water *ad libitum*. The experimental protocols were approved and carried out according to the local experimental ethics committee.

All animals were conditioned at room temperature at a natural photoperiod for 1 week before experiment execution, then divided into two main groups as follows:

**The first main group**: (7 rats) fed on basal diet, as a negative control group (normal).

**The second main group**: (35 rats) fed on basal diet and injected with CCl$_4$ (2mL/kg body weight), dissolved in paraffin oil (%50) V/V intraperitoneally twice/week for 2 weeks, to induce chronic liver damage, according to the method of (15).

**The second main group divided into five subgroups each subgroup contains** (7 rats) and divided as follows-:

**The first subgroup**: rats fed on basal diet, as a positive control group.

**The second subgroup**: rats fed on basal diet and ingested with 2.5 mL green coffee aqueous extract) equivalent to 150 mg of CGA (orally by gastric gavage once daily for 6 weeks, according to the method of (16).

**The third subgroup**: rats fed on basal diet and ingested with 2.5 mL roasted coffee aqueous extract) equivalent to 150 mg of CGA (by gastric gavage once daily for 6 weeks, according to the method of (17).
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**The fourth subgroup**: Rats fed on basal diet and ingested with 2.5 mL green tea aqueous extract) equivalent to 150 mg of CGA (by gastric gavage once daily for 6 weeks, according to the method of. (18)

**The fifth subgroup**: Rats fed on basal diet and ingested with 2.5 mL black tea aqueous extract) equivalent to 150 mg of CGA (by gastric gavage once daily for 6 weeks, according to the method of. (18)

**At the end of the experimental period 8) weeks**, rats were fasted overnight before sacrificing. Blood samples were collected from each rat and centrifuged at 3000 rpm to separate the serum. Serum was carefully separated and transferred into dry clean Eppendorf tubes and kept frozen at -20° C till analysis.

**Biochemical analysis**:

**Serum Glucose**. Serum glucose was determination according to the procedure as described by. (19)

**Liver function**. Blood samples collected just before sacrifice from the tail veins for assessment of liver function. Alanine aminotransferase (ALT) and Aspartate Aminotransferase (AST) activities were determined according to the method of. (20) Alkaline Phosphates (ALP) activities were determined according to the method of. (21)

**Kidney function**. Urea was determined using commercial kit) Egyptian Company for Biotechnology, Egypt) according to. (22)Serum creatinine was determined according to the method reported by. (23)

**Serum lipoproteins**. High density lipoprotein cholesterol (HDL-c) was determined using the method declared by. (24) Very low-density lipoprotein cholesterol (VLDL-c) and Low density of lipoprotein cholesterol (LDL-c) were calculated as reported by. (25)
Histopathological examination:

For histomorphology evaluation, a portion of liver tissue was fixed in Bouin’s solution. Paraplast-embedded liver was sectioned (4μm thick) and stained with hematoxylin and eosin (H and E) and Masson’s trichrome stain. Representative histopathological features such as necrosis or inflammatory cell infiltration and fibrosis were examined under a microscope in a blind manner. Intensity of necro-inflammatory lesions was graded as follows: none, mild, moderate and severe, according to the method illustrated by.(26)

Statistical analysis:

Data presented here expressed as Mean ± SD and one-way analysis of variance (ANOVA) was undertaken followed by a post-hoc LSD (Least Significant Difference) test. A p value > 0.05 considered statistically significant. A Statistical analysis performed with the Statistical Package for the Social Sciences for Windows (SPSS, version 17.0, Chicago, IL, USA) (Steel and Torri.1980).

Result and discussion:

Effect of Some Beverage Rich in Chlorogenic acid on body weight gain (g) in the studied rat groups.

On the basis of initial weight, there was no statistically significant difference between the negative control group, the positive control group, and all treated groups) P) (41. =Table 1. (There was a statistically significant difference between the positive control group when compared to green coffee group and green tea group 6.83 ± 192.33) g vs. 193.67 ± 7.50 g, respectively) in the final weight.
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Table : (1) Effect of Some Beverage Rich in Chlorogenic acid on body weight gain (g) in the studied rat groups.

<table>
<thead>
<tr>
<th>Groups</th>
<th>(g) initial weight</th>
<th>(g) final weight</th>
</tr>
</thead>
<tbody>
<tr>
<td>(-) Cont</td>
<td>179.67±2.73</td>
<td>215.67±18.42</td>
</tr>
<tr>
<td>(+) Cont</td>
<td>178.00±5.59</td>
<td>210.33±3.72</td>
</tr>
<tr>
<td>Green Coffee</td>
<td>175.00±1.79</td>
<td>192.33±6.83</td>
</tr>
<tr>
<td>Roasted Coffee</td>
<td>177.33±6.28</td>
<td>207.67±4.93</td>
</tr>
<tr>
<td>Green Tea</td>
<td>176.33±1.86</td>
<td>193.67±7.50</td>
</tr>
<tr>
<td>Black Tea</td>
<td>176.33±2.25</td>
<td>204.33±9.40</td>
</tr>
</tbody>
</table>

LSD : Least significant differences (p > 0.05). Data were presented as mean ± standard deviation.

Means with the same latter in each column are not significantly different.

Fig : (1) Effect of Some Beverage Rich in Chlorogenic acid on initial weight and final weight (grams) in all the studied rat groups.

Data were presented as mean ± standard deviation.

Con : negative control group ; con + : positive control group.

Effect of Some Beverage Rich in Chlorogenic acid on serum glucose (mg/dl) and Liver enzymes levels (u/l) in the studied rat groups.
The results shown in Table 2 indicate that rats injected with CCl₄ induced damage to the liver of positive control group, which led to increased serum glucose level (140.00 ± 5.59 mg/dl), as compared to the negative control group and other treated groups. Green coffee group has pronounced effect in lowering the level of serum glucose (137 ± 125.67) The positive control group recorded significant increase in serum AST level, as compared to healthy rats (negative control group) 1.29 ± 24.20) vs. 17.36 ± 2.23 u/l, respectively. On the other hand, all treated groups recorded significant decrease in serum AST enzyme as compared to the control positive group, except for black tea group (1.76 ± 22.06)u/l.

There was significant difference in the serum level of ALT between positive control group and all treated groups. Green coffee, and green tea (10.21 ± 0.93, 11.04 ± 0.89 respectively) have similar effect on serum level of ALT. There was no significant difference in ALP enzyme between the positive control group and the negative control group 6.26 ± 209.00) u/l. Among the treated groups, the lowest mean value of ALP enzyme was found in black tea 4.41 ± 203.33) u/l), green tea (204.67 ± 4.93 u/l) followed by roasted coffee (206.33 ± 5.82 u/l).

The current results in concordance with (27) who reported that coffee consumption has been associated with decreased levels of AST, ALT and ALP. It has been reported that high coffee consumption 2 (<) cups per day) was associated with a low risk of elevated ALT levels by 44% and a lower risk of chronic liver disease compared to non-coffee drinkers. (28) Moreover, coffee consumption also had beneficial associations with renal stones and for liver conditions including hepatic fibrosis, cirrhosis, cirrhosis mortality, and chronic liver disease combined. (29)

(30) suggested that higher coffee consumption was associated with lower concentrations of log-transformed ALP, ALT and AST. Higher coffee consumption was associated with lower odds of having elevated levels of all studied liver enzymes. An inverse association between higher coffee consumption and
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serum concentrations of baseline liver enzymes and NAFLD risk. These findings suggest that habitual consumption of coffee may be protective of liver health.

There was a significant lowering effect of green tea intake on liver enzymes; ALT and AST compared to no green tea intake. The authors suggested that green tea could be added to the daily dietary program to improve cardiovascular status with no possible risk of liver cancer. It even may have a protecting effect against liver cancer in the usual daily number of cups (up to 5 a day).(31) Moreover, epidemiological studies have indicated that green tea has beneficial effects in liver diseases. In a clinical experiment conducted on nine cases of intractable chronic hepatitis C with a high viral load, found that combination therapy of 6 g of green tea powder/day and interferon/ribavirin showed 3.5-fold efficacy compared with interferon/ribavirin therapy alone.(32) A significant reduction in serum levels of ALT (%38) and AST (23%–34%) was observed (p < 0.05) with black tea. the reduction in the level of ALT and AST observed in that study is therefore an indication of the lack of damage to the liver.(33) It has been indicated that regular consumption of theaflavin-enriched black tea extract can achieve the effect of preventing liver or kidney fibrosis.(34)

The mechanism by which tea polyphenols produce antioxidant effects includes the following processes: the increase in activity of antioxidant enzymes, the inhibition of lipid peroxidation, the scavenging of free radicals in synergy with other nutrients, and the reduction of oxidation via chelation of metal ions.(35)
Table 2: Effect of Some Beverage Rich in Chlorogenic acid on serum glucose and Liver enzymes levels in the studied rat groups.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Glucose (mg/dl)</th>
<th>AST (u/l)</th>
<th>ALT (u/l)</th>
<th>ALP (u/l)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cont(-)</td>
<td>129.00cd3.90</td>
<td>17.36c2.23</td>
<td>10.12c0.94</td>
<td>209.00ab6.26</td>
</tr>
<tr>
<td>Cont(+)</td>
<td>140.00a5.59</td>
<td>24.20a1.29</td>
<td>16.19a1.78</td>
<td>212.67a1.37</td>
</tr>
<tr>
<td>Green Coffee</td>
<td>125.67d1.37</td>
<td>17.45c3.56</td>
<td>10.21c0.93</td>
<td>209.00ab1.79</td>
</tr>
<tr>
<td>Roasted Coffee</td>
<td>130.67bc1.86</td>
<td>20.29bc1.11</td>
<td>12.76b1.32</td>
<td>206.33b5.82</td>
</tr>
<tr>
<td>Green Tea</td>
<td>133.33b2.25</td>
<td>18.76c2.79</td>
<td>11.04c0.89</td>
<td>204.67b4.93</td>
</tr>
<tr>
<td>Black Tea</td>
<td>130.00bc1.79</td>
<td>22.06ab1.76</td>
<td>14.13b0.93</td>
<td>203.33b4.41</td>
</tr>
</tbody>
</table>

LSD : Least significant differences (p > 0.05). Data were presented as mean ± standard deviation.

Means with the same latter in each column are not significantly different.

ALP: Alkaline phosphatase; AST: Aspartate aminotransferase; ALT: Alanine aminotransferase.

Fig 2: Effect of Some Beverage Rich in Chlorogenic acid on glucose levels (mg/dl) in all the studied rat groups.

Data were presented as mean ± standard deviation.

Con :- negative control group ; con +: positive control group.
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**Fig : (3) . Effect of Some Beverage Rich in Chlorogenic acid on AST enzyme (u/l) in all the studied rat groups.**

[Graph showing AST enzyme levels in different groups]

Data were presented as mean ± standard deviation.

Con :-negative control group ; con +: positive control group; AST: Aspartate aminotransferase

**Fig : (4) . Effect of Some Beverage Rich in Chlorogenic acid on ALT enzyme (u/l) in all the studied rat groups.**

[Graph showing ALT enzyme levels in different groups]

Data were presented as mean ± standard deviation.

Con :-negative control group ; con +: positive control group; ALT: Alanine aminotransferase.
Fig. (5) Effect of Some Beverage Rich in Chlorogenic acid on ALP enzyme (u/l) in all the studied rat groups.

Data were presented as mean ± standard deviation.

Con :-negative control group ;con +: positive control group; ALP: Alkaline phosphatase.

**Effect of Some Beverage Rich in Chlorogenic acid on kidney functions (mg/dl) in the studied rat groups.**

Serum urea nitrogen was significantly lower in the control negative group as compared to the control positive control group (1.44 ± 30.50) vs. 43.20 ± 1.99 mg/dl, respectively). The highest serum urea nitrogen level was recorded in the control positive group (43.20 ± 1.99 mg/dl) compared with all other treated group. The highest mean value of serum creatinine level was found in the positive control group (1.00 ± 0.06 mg/dl), as compared to the negative control group (0.69 ± 0.13 mg/dl) as well as other treated groups, except for the group treated with roasted coffee (0.90 ± 0.24 mg/dl). There was a significant difference in serum creatinine level between the positive control group, green coffee, green and black tea (0.65 ± 0.10 mg/dl, 0.69 ± 0.11 vs ± 0.76 ± 0.13 mg/dl, respectively.)
A BENEFICIAL EFFECT OF COFFEE ON KIDNEY FUNCTION HAS BEEN REPORTED BY .(36) A SYSTEMATIC REVIEW AND META-ANALYSIS OF CLINICAL STUDIES INVESTIGATED THE EFFECT OF COFFEE CONSUMPTION ON RENAL OUTCOME, 12 STUDIES WERE INCLUDED IN THE ANALYSIS (7 PROSPECTIVE, 5 CROSS-SECTIONAL) INVOLVING 505,841 SUBJECTS, 7 STUDIES SHOWED THAT COFFEE INTAKE WAS DOSE-DEPENDENTLY ASSOCIATED WITH LOWER INCIDENT OF CHRONIC KIDNEY DISEASE (CKD) .(37) THE ASSOCIATION BETWEEN COFFEE AND CKD HAS BEEN INVESTIGATED IN 14,209 PARTICIPANTS AGED 45 TO 64 YEARS FROM THE ATHEROSCLEROSIS RISK IN COMMUNITIES (ARIC) STUDY, THE STUDY CONCLUDED THAT PARTICIPANTS WHO DRANK HIGHER AMOUNTS OF COFFEE HAD LOWER RISK FOR INCIDENT CKD (PARTICIPANTS NEVER CONSUMED COFFEE 2->1 ; CUPS PER DAY; 2 3->-CUPS PER DAY; ≥3 CUPS PER DAY) .(38)

These results are in accordance with previous findings by (39) who showed that tea exerts many protective effects against stone formation, through the accompanying water intake, the action of caffeine and the effects of components with antioxidant properties. Moreover, green tea had a significantly favorable effect on serum urea and serum creatinine, green tea had ameliorative effects on the ibuprofen-induced changes in the biochemical markers of the adult rat kidneys .(40) It was also concluded by (41) that the consumption of green tea extract produced a significant reduction in glucose level in diabetic rats. In addition, it can improve the impaired kidney functions in diabetic anima.
Table : (3) Effect of Some Beverage Rich in Chlorogenic acid on kidney functions in the studied rat groups.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Urea) Mg/dl(</th>
<th>Creatinine) Mg/dl(</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cont(-)</td>
<td>30.50cd1.44±</td>
<td>0.69c0.13±</td>
</tr>
<tr>
<td>Cont(+)</td>
<td>43.20a1.99±</td>
<td>1.00a0.06±</td>
</tr>
<tr>
<td>Green Coffee</td>
<td>28.60d1.82±</td>
<td>0.65c0.10±</td>
</tr>
<tr>
<td>Roasted Coffee</td>
<td>36.20b6.00±</td>
<td>0.90ab0.24±</td>
</tr>
<tr>
<td>Green Tea</td>
<td>30.40cd0.95±</td>
<td>0.69c0.11±</td>
</tr>
<tr>
<td>Black Tea</td>
<td>33.53bc1.12±</td>
<td>0.76bc0.13±</td>
</tr>
</tbody>
</table>

LSD :Least significant differences (p .0.05< Data were presented as mean ± standard deviation.

Means with the same latter in each column are not significantly different.

Fig : (6) .Effect of Some Beverage Rich in Chlorogenic acid on urea nitrogen levels (mg/dl) in all the studied rat groups.

Data were presented as mean ± standard deviation.

Con :-negative control group ;con +: positive control group.
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**Fig :** Effect of Some Beverage Rich in Chlorogenic acid on serum creatinine levels (mg/dl) in all the studied rat groups.

![Bar Chart](image)

Data were presented as mean ± standard deviation.

Con :-negative control group ;con +: positive control group.

**Effect of Some Beverage Rich in Chlorogenic acid on serum lipid profile levels in the studied rat groups.**

As indicated in) **Table 4**, (serum cholesterol (mg/dl) in all treated groups decreased significantly as compared to the positive control group. There was a significant difference in serum triglycerides) mg/dl (level in the positive control group as compared to all treated groups ,which indicating a lowering effect of the treatment used (42) .found a positive dose-dependent association between self-reported coffee intake and plasma concentration of total-C, with the highest lipid levels seen among participants reported drinking 6 < cups/day).

Results in) **Table 4** (revealed a significant difference between the positive control group (33.67 ± 1.37) and the negative control group (43.67 ± 4.03). In addition, all treated groups showed significant increase in serum HDL-c as compared to the positive control group except for black tea group. The highest serum LDL-c was found in the positive control group (57.73 ± 1.62 mg/dl), which was significantly different when
compared to the negative control group (36.20 ± 13.49) and when compared to all treated groups. The highest serum VLDL-c was found in the positive control group (23.60 ± 1.12 mg/dl), and that was significantly different compared with all type of treatment. Coffee consumption of ≥5 cups per week was positively associated with HDL-C (p=0.0442) compared with the lowest level (1) cup/week) of consumption. (43)

(44) STUDIED THE EFFECTS AND MOLECULAR MECHANISMS OF NINGHONG BLACK TEA EXTRACT IN NONALCOHOLIC FATTY LIVER DISEASE OF RATS AND STATED THAT NINGHONG BLACK TEA EXTRACT RELIEVES HIGH-FAT DIET-INDUCED NAFLD, BY PROMOTING THE EXPRESSION OF PEROXISOME PROLIFERATOR-ACTIVATED RECEPTOR A (PPARΑ), WHICH IS IMPORTANT IN FATTY ACID B-OXIDATION AND MICROSOMAL TRIGLYCERIDE TRANSFER PROTEIN (MTP) IN LIVER TISSUE AND THEREBY PROMOTING FATTY ACID B-OXIDATION AND VLDL SYNTHESIS.

(45) investigated the effect of coffee on body weight, serum glucose, uric acid and lipid profile levels in male albino Wistar rats fed on high fructose diet, the results indicated that fasting serum TG and LDL-C levels were significantly lower in rats treated with 284 mg/kg BW/day of coffee compared to fructose control group) p (0.046 ; 0.031 = respectively. Fasting serum levels of TC and HDL-C were non-significantly lower and higher respectively, in the treated group.
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Table : (4) Effect of Some Beverage Rich in Chlorogenic acid on serum lipid profile levels in the studied rat groups.

<table>
<thead>
<tr>
<th>Groups</th>
<th>HDL-c (Mg/dl)</th>
<th>LDL-c (Mg/dl)</th>
<th>VLDL-c (Mg/dl)</th>
<th>Cholesterol (Mg/dl)</th>
<th>Triglycerides (Mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cont(-)</td>
<td>4.03 ±a 43.67</td>
<td>13.49 ±b 36.20</td>
<td>1.19 ±cd 17.47</td>
<td>11.04 ±b 97.33</td>
<td>5.96 ±cd 87.33</td>
</tr>
<tr>
<td>Cont(+)</td>
<td>1.37 ±bc 33.67</td>
<td>1.62 ±a 57.73</td>
<td>1.12 ±a 23.60</td>
<td>1.79 ±a 115.00</td>
<td>5.59 ±a 118.00</td>
</tr>
<tr>
<td>Green Coffee</td>
<td>2.73 ±ab 42.67</td>
<td>4.25 ±b 30.53</td>
<td>0.55 ±bc 19.13</td>
<td>2.25 ±b 92.33</td>
<td>2.73 ±bc 95.67</td>
</tr>
<tr>
<td>Roasted Coffee</td>
<td>1.86 ±ab 42.33</td>
<td>4.12 ±b 34.13</td>
<td>0.47 ±b 20.20</td>
<td>2.73 ±b 96.67</td>
<td>2.37 ±b 101.00</td>
</tr>
<tr>
<td>Green Tea</td>
<td>2.37 ±b 40.00</td>
<td>11.54 ±b 38.33</td>
<td>1.71 ±cd 18.00</td>
<td>1.104 ±b 96.33</td>
<td>8.53 ±cd 90.00</td>
</tr>
<tr>
<td>Black Tea</td>
<td>2.37 ±c 34.00</td>
<td>3.80 ±b 37.73</td>
<td>0.47 ±b 19.60</td>
<td>2.73 ±b 91.33</td>
<td>2.37 ±b 98.00</td>
</tr>
</tbody>
</table>

LSD : Least significant differences (p > 0.05) Data were presented as mean ± standard deviation.

Means with the same latter in each column are not significantly different.

LDL-c : Low-Density Lipoprotein cholesterol; HDL-c : High-Density Lipoprotein cholesterol; VLDL : Very Low-Density Lipoprotein cholesterol.

Fig : (8) Effect of Some Beverage Rich in Chlorogenic acid on HDL-c, LDL-c and VLDL-c levels mg/dl (in all the studied rat groups).

Data were presented as mean ± standard deviation.

Con :- negative control group ;con +: positive control group ;HDL-c : High-Density Lipoprotein cholesterol ;LDL-c: Low-Density Lipoprotein cholesterol ;VLDL-c : Very Low-Density Lipoprotein cholesterol.
**Histopathological Results**

Microscopical examination of liver of negative control group revealed no histopathological alteration, normal histological structure of the central vein as well as portal area. The liver of the positive control group showed thickening and fibrosis in the Glisson’s capsule in focal manner and focal inflammatory cells infiltration in between the fatty cells at the parenchyma. Green tea, roasted coffee groups showed normal histological structure. Kidney of roasted coffee group, showed normal histological structure) **Fig. (10)**.

**Conclusion:**

Beverage rich in chlorogenic acid such as tea and coffee proved its ameliorative effect on serum glucose, lipid profile, liver and kidney functions. Green tea, roasted coffee showed normal histological structure. Besides, green coffee and green tea have better effects on body weight gain.
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- Con

Liver

Kidney

+ Con

Green coffee

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Fig) : (10) . a (liver of negative control group, showing normal histological structure of the central vein and surrounding hepatocytes in the penenchy (H&E X 40) . b (liver of positive control group, showing focal inflammatory cells information in
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between group fat cells in hepatocytes) H&E x) .(40 c (liver of green coffee group, showing few numbers y newly bile ductules formarien in poiraluaea (H&E x) .(40 d (liver of roasted coffee group, showing normal histological structure of the central vein and surrounding hepatocytes in the parenchyma (H&E x) .(40 e (liver of green tea group, showing normal histological structure of the central vein and surrounding hepatocytes in the parenchyma (H&E x) .(40 f (liver of black tea group, showing of inflammatory cells infiltration in portal area (H&E x) .(40 g (kidney of negative control group, showing normal histological structure of the tubules at the corticomedullary portion (H&E X 40) .(h (kidney of positive control group, showing focal hemorrhage in between the degenerated tubules at the corticomedullary portion (H&E x) .(40i (kidney of green coffee group, showing decyenencerion in lining tubular epithelium at the coitex (H&E x) .(40 j (kidney of roasted coffee group, showing normal histological structure (H&E x) .(40 k (kidney of green tea group, showing focal inflammatory cells information in between the tubules at the corticomedullary portion (H&E x) .(40 l (kidney of black tea group, showing decyenencerion in lining tubular epithelium of the corticomedullary portion (H&E x). (40

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