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CO-TREATMENT WITH CRANBERRY AND VITAMIN-C AMELIORATES THE HEPATO-RENAL TOXICITIES INDUCED BY PHENOBARBITAL IN WISTAR RATS

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Although phenobarbital (PB) is used to treat epilepsy, its long-term treatment leads to liver and kidney dysfunctions. Natural products, however, can provide essential resources to alleviate hepato-renal toxicities due to their antioxidant's properties. This study evaluated the impact of cranberries (CB) and Vitamin-C (Vit-C) on hepato-renal toxicities induced by PB in rats. Forty male Wistar rats were allocated into five groups (N= 8) as follows: Group 1 (Gp1) was served as a negative control. Gp2 had administered with PB (160 mg/kg), orally for a month. Gp3 and Gp4 had administered with PB as in Gp2, then administered with CB (500 mg/kg) and Vit-C (27 mg/kg), respectively for a month. Gp5 had administered with PB, then administered with a combination of CB and Vit-C as in Gp3 and Gp4. Liver transaminases, albumin, total protein, creatinine, urea, total lipids, cholesterol, as well as catalase (CAT), super oxide dismutase (SOD), glutathione reduced (GSH) and malondialdehyde (MDA) were assessed. The liver and kidney histopathological alterations were examined. The treatment with PB decreased the percentage of the body weight changes, impaired the liver and kidney functions, increased the levels of total lipids, cholesterol, MDA levels, decreased the SOD, CAT activities, and changed the histological architectures of both kidney and liver. Treatment with CB or Vit-C ameliorates the adverse effects that were induced by PB, however, PB-injected rats that were co-treated with CB/Vit-C showed the highest therapeutic effects. Collectively, co-treatment with CB and Vit-C mitigated hepato-renal toxicities induced by PB-treatment in rats.

Keywords: *Cranberry; Vitamin-C; Phenobarbital; Hepato-renal; Toxicity.*

INTRODUCTION

Phenobarbital (PB) is belonged to a group of medications known as barbiturates that used as anti-seizure agents[1- 3]. PB is used to alleviate sleep anxiety, drug withdrawal issues and facilitate surgery[4]. Long-term use of PB has been linked to adverse outcomes such as irritability, lack of desire, soreness in the joints, bones, sadness, and liver failure[5].

PB has been observed to enhance the formation of reactive oxygen species (ROS), possibly via the uncoupling of cytochrome P450[6]. For this reason, PB altered the liver physiology through the hepatic hypertrophy, hyperproliferation of the smooth endoplasmic reticulum, and activation or repression of several genes, including those encoding the cytochrome P450 enzyme[6&7]. In addition, PB caused nephrotoxicity in rats and elevated levels of urea nitrogen, uric acid, and creatinine[6]. PB intoxication includes drowsiness, respiratory depression, coma, hypotension, cardiovascular collapse, and hypothermia[8&9]. Long term use of PB causes a rise in plasma total cholesterol concentrations in rodents and humans[10]. PB is also the model for liver tumor promoters, drastically

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increasing the tumor numbers when chronically provided following the first treatment with genotoxic carcinogens[11]. Previous study showed that the treatment with PB increase MDA level associated with significant decreases in antioxidant concentration in treated rats[12].

Natural products that were extracted from the medicinal plants have several phytochemical compounds as polyphenolic, flavonoids, and proanthocyanins with various health advantages in inflammatory processes, treatment of cancer, diabetes, cardiovascular disorders, and age-related neurodegenerative diseases[13-16]. Cranberry (CB) is a medicinal plant of family Ericaceae. CB has high levels of dietary fiber, organic acids, flavonoids, glycosides, terpenoids, tannins, and alkaloids[17]. These compounds act as potent antioxidant agents and provided crucial protective effects as anticancer, anti-inflammatory, and anti-mutagenic agents[18-21].

CB is also helpful in the management of hyperglycemia, diabetes, hypercholesterolemia, rheumatoid arthritis, urinary tract infections, obesity, insulin resistance, and intestinal inflammation[22 25]. CB can significantly increase the antioxidant defenses against free radicals-induced oxidative stress, and indicate remarkable hepatoprotective efficacy versus chemical-induced hepatotoxicity[7&26&27]. A previous study reported that CB has a protective effect against the renal damage that induced to toxic agents[28]. In addition, the phytochemicals in CB may possibly also hinder LDL oxidation and platelet aggregation, increase expression of LDL receptor, and decrease blood pressure[29].

Vitamin-C (Vit-C) is a water-soluble vitamin with outstanding reducing characteristics. It is well recognized for its high antioxidant capacity due to the neutralization of free radicals generated during cell metabolism[30]. Vit-C performs a part in several enzyme reactions, involving those important to the synthesis of amino acids and peptide hormones. In addition, Vitamin C functions as an important cofactor in oxidative stress pathways. Therefore, Vit-C is potentially useful as a therapeutic agent that could be used to treat the illnesses that were related with free radicals[31]. Vit-C has been shown to have hepatoprotective benefits and able to reduce the liver damage that induced by some chemical agents[32]. Consequently, this study aims to determine the impact of the treatment with cranberry, Vit-C or their combination on the hepato-renal damages that were induced by the phenobarbital in Wistar male rats.

MATERIALS AND METHODS

Chemicals

Phenobarbital (PB) was obtained from Sigma Company (USA). Cranberry (CB) and Vitamin-C (Vit-C) were purchased from the pharmacy (Cairo, Egypt). Kits for albumin, total protein, aspartate amino transferase (AST), alanine amino transferase (ALT), creatinine, urea, cholesterol, total lipid, catalase (CAT), superoxide dismutase (SOD), glutathione reduced (GSH) and malondialdehyde (MDA) were purchased from Bio-diagnostic Company.

Rats and experimental design

Adult male Wistar Rats with an average weigh 120 ± 4 g, were taken from National Research Center (NRC, Cairo, Egypt). Rats were kept for a week for adaptation. The temperature and relative humidity were around 22 ± 1 ºC and 55 ± 5%, respectively. Light-dark (day/night) cycle was attained and the animals were managed permitting to the ethical guidelines approved by the animal care and use committee, Faculty of Science, Tanta University (Protocol number: IACUC-SCITU 0139).

Group 1 (Gp1) was served as a negative control. Gp2 had administered with PB (160 mg/kg) orally, for a month. Gp3 and Gp4 had administered with PB as in Gp2, then treated orally daily for a month with CB (500 mg/kg) and Vit-C (27mg/kg), respectively. Gp5 had administered with PB as in Gp2 and with CB/Vit-C as in Gp3 and Gp4 (Figure 1). Sera samples, liver tissues from all groups were collected for assessment of biochemical markers. kidney and liver tissues were collected for histopathological examinations.

Determination the total body weight changes

Each group of rats was weighted at the start of the experiment (I.B.wt) and at the end of the experiment (F.B.wt). The percentage of the change in the total body weight (% b.wt)

was estimated as follow: (F.B.wt – I.B.wt / I.B.wt) × 100.

Determination of the biochemical parameters

Serum aminotransferases (ALT and AST) activities were determined according to Reitman and Frankel[33]. Serum levels of albumin, and total proteins were determined according to Doumans et al and Gornal et al [34, 35], respectively. Creatinine and urea levels were assessed by colorimetric methods according to Fawcett and Scott[36] and Bartles et al.[37], respectively. Serum total lipids and cholesterol were measured according to Friedman[38] and Allain et al[39], respectively. The hepatic levels of catalase (CAT) were measured according to Aebi [40], superoxide dismutase (SOD) Nishikimi et al [41], reduced glutathione GSH, and MDA were assessed according to Beutler et al and Buege and Aust,[42&43], respectively.

Histological investigations

The liver and kidneys specimen of various groups were fixed in 10% formal. The dehydration process was done with a succession of weak alcohol solutions. Paraffin blocks were primed after finishing the tissue processing in various grade of alcohol and xylene. Sections (5 μm) were made from paraffin blocks applying microtome, stained with hematoxylin and eosin, which studied under light microscope (Optika light microscope (B- 350) to examine gross cellular damage[44].

Statistical analysis

SPSS program was selected for data analysis. The outcomes of the study were presented as mean ± standard error means (SEM). One-way analysis of variance (ANOVA) was employed to evaluate the data, followed by the Tukey test for multiple comparisons. Values with a $P < 0.05$ were statistically significant.

RESULTS AND DISCUSSION

Results

Effect of the treatment with CB or/and Vit-C on the percentages of the body weight change in PB-treated rats

Rats were distributed into five experimental groups (Figure 1). The findings revealed that the group of rats that were treated with PB revealed a significant reduction in the % b.wt change ($p \leq 0.05$) up to -10%, when compared to the negative control group that represented 22.5%. The group of PB-treated rats that were administered with CB indicated a significant increase ($p \leq 0.05$) in the % b.wt change (13%) when compared to the PB-treated rats alone (Figure 2). The group of PB-injected rats that were treated with Vit-C showed also a significant increase in the % b.wt change (9%) but not much as CB treatment when compared to the PB-treated rats alone ($p \leq 0.05$). As compared to the PB-treated rats, the group of PB-treated rats that were treated with a combination of CB and Vit-C showed significant increase ($p \leq 0.01$) in the % b.wt changes, represented by 19% (Figure 2).

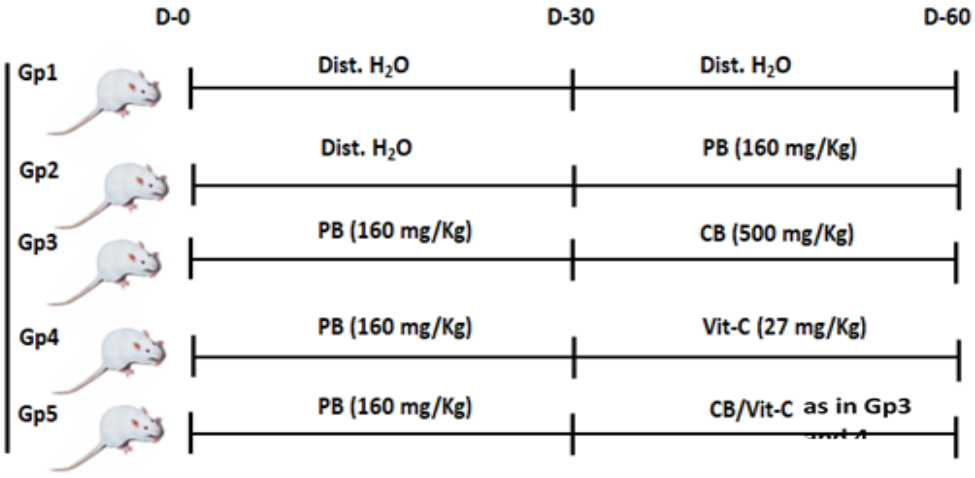

Fig. 1: The time-line of the experimental groups under the study

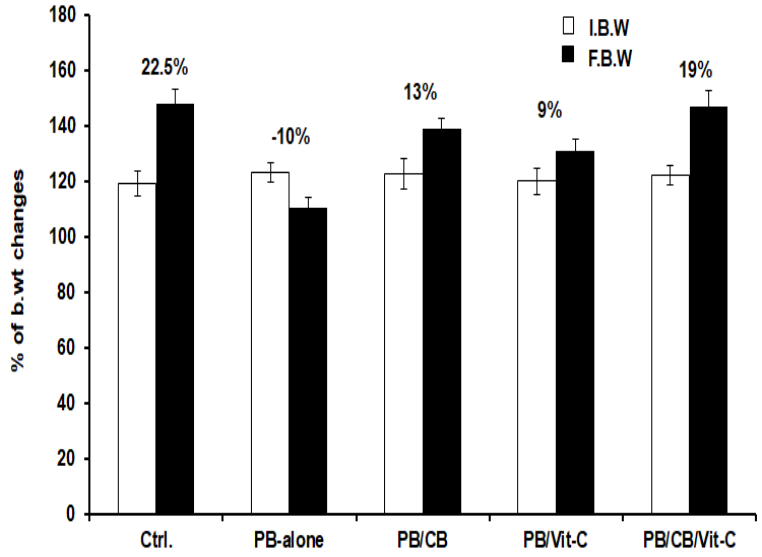

Fig. 2 : The initial, final body weight, and their percentages of body weight change in the different groups under the study. The values represented mean ± SE; **I.B.wt:** Initial body weight; **F.B.wt:** Final body weigh**; Ctrl:** Control; **PB:** Phenobarbital**; CB:** Cranberry; **Vit-C:** Vitamin C. P value < 0.05 was considered to be statistically significant.

Effect of the treatment with CB or/and Vit-C on the total protein and albumin levels in PB-injected rats

The concentrations of the albumin and total protein were significantly reduced in the group of rats that were treated with PB when compared to control group ($p \leq 0.01$). Treatment of PB-injected rat with CB or Vit-C led to significant increase the levels of the total protein and albumin as compared to PB-treated rats alone ($p \leq 0.01$). The treatment with a combination of CB/Vit-C, however, led to a much more improvement in the albumin and total protein concentrations when compared to the group of rats that were injected with PB-alone ($p \leq 0.01$) (Figure 3).

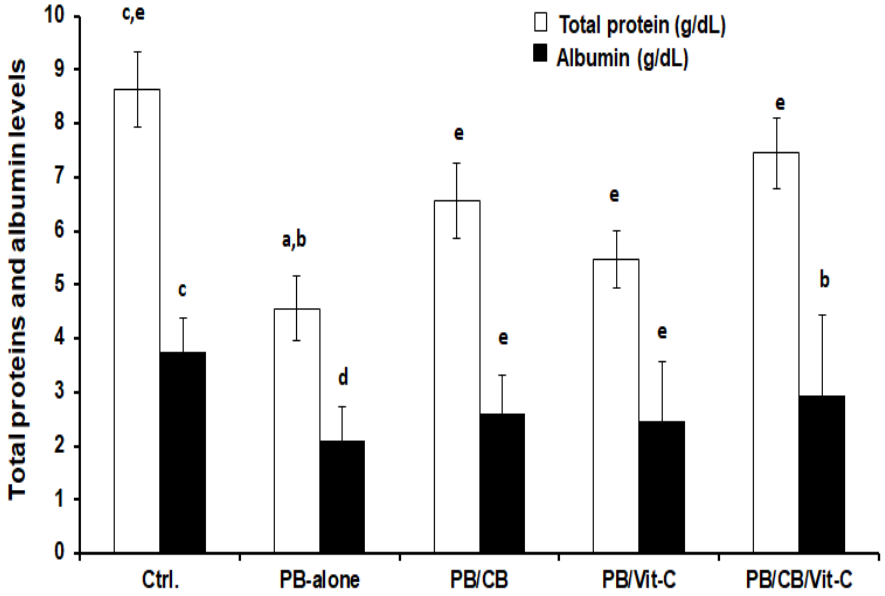

Fig. 3: Total proteins and albumin levels in the different groups under the study. The values represented mean ± SE; **Ctrl:** Control; **PB:** Phenobarbital**; CB:** Cranberry; **Vit-C:** Vitamin C. P value < 0.05 was considered to be statistically significant.

Co-treatment with CB/Vit-C mitigated the hepatic and renal dysfunctions in the PB-injected rats

Compared to the control group, the PB-injected group did show a significant increase ($p \leq 0.05$) in the in ALT and AST activities. PB-injected rats that were treated with CB or Vit-C did show significant decline ($p \leq 0.05$) in ALT and AST levels as compared to the PB-injected rats alone. As compared to PB-treated rats alone, the PB-treated group that was co-treated with CB and Vit-C did show the most significant reduction ($p \leq 0.05$) in the liver transaminases levels (Figure 4).

Compared to the control group, the PB-injected rats did show a significant increase ($p \leq 0.05$) in the kidney biomarkers levels (urea and creatinine). Rats that were treated with CB or Vit-C did show significant decrease ($p \leq 0.05$) in creatinine and urea levels as compared to PB-injected rats. Rats that were treated with a combination of CB/Vit-C showed much more improvement in kidney functions that evidenced by significant reduction ($p \geq 0.05$) in the levels of creatinine and urea when compared to PB-treated rats (Table 1).

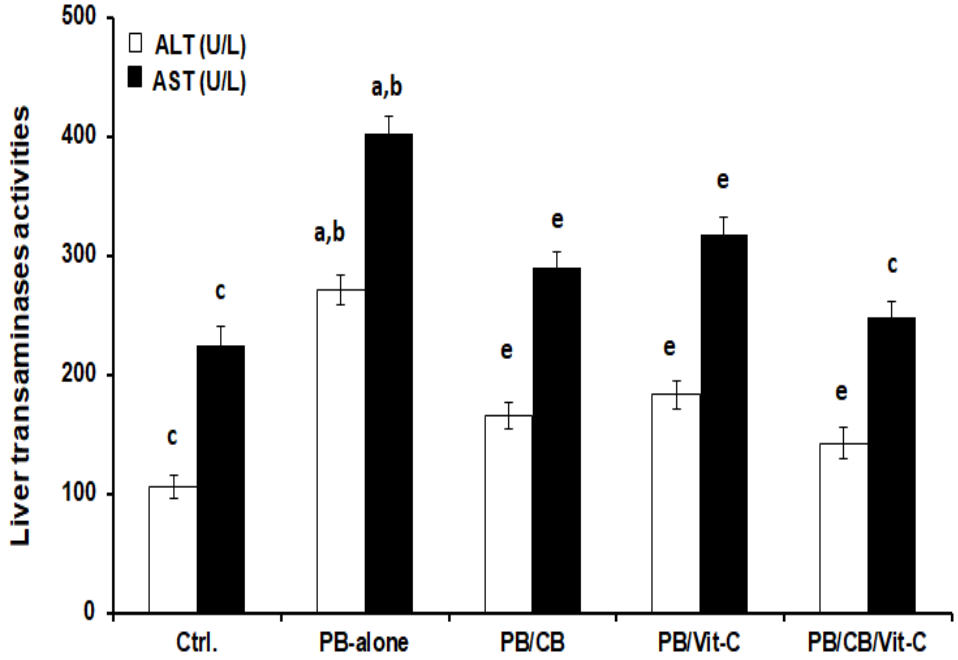

Fig. 4: Liver transaminases (ALT and AST) activities in the different groups under the study. The values represented mean ± SE; **Ctrl:** Control; **PB:** Phenobarbital**; CB:** Cranberry; **Vit-C:** Vitamin C. P value < 0.05 was considered to be statistically significant.

Table 1: The urea and creatinine levels in different groups under the study

Groups	Urea (mg/dl)	Creatinine (mg/dl)
Ctrl.	27.64 ± 2.08^{c}	0.60 ± 0.07^{c}
PB-alone	48.93 ± 3.83^{d}	1.18 ± 0.09^{b}
PB/CB	33.62 ± 2.45^{a}	0.84 ± 0.09^{e}
PB/Vit-C	$36.3 \pm 3.04^{a, b}$	0.96 ± 0.08^{e}
PB/CB/Vit-C	30.854 ± 2.95^{a}	$0.75 \pm 0.09^{e, d}$

The values represented mean ± SE; Ctrl: Control; PB: Phenobarbital; CB: Cranberry; Vit-C: Vitamin C. P value < 0.05 was considered to be statistically significant. Means that do not share a letter are significantly different.

Impact of the treatment with CB or/and Vit-C on the oxidant/antioxidant status in PB-treated rats

Compared to the control group, rats that were treated with PB showed significant decrease ($p \leq 0.05$) in the CAT and SOD activities. Also, the PB-treated rats showed a significant decrease in GSH level and significant increase ($p \leq 0.05$) in the MDA level when compared to the control group. PB-injected rats that were treated with CB or/and Vit-C did show significant increase in the CAT and SOD activities, in addition to a significant increase in GSH level and significant decrease ($p \leq 0.05$) in the MDA level when compared to PB-treated rats (Table 2).

Effect of the treatment with CB or/and Vit-C on the total lipid cholesterol levels in PB-treated rats

The levels of the total lipid and total cholesterol were significantly increased ($p \leq 0.01$) in the group of rats that were treated with PB when compared to control group. Treatment of PB-injected rats with CB or Vit-C led to significant decrease ($p \leq 0.01$) in the levels of the total lipid and cholesterol as compared to PB-treated rats alone. The treatment of PB-injected rats with a combination of CB and Vit-C, however, led to a significant decrease ($p \leq 0.01$) in the total lipid and cholesterol levels more than treatment with CB or Vit-C alone when compared to the group of rats that were treated with PB-alone (Figure 5).

Table 2: Oxidative stress biomarkers of hepatic tissue in the different groups under the study.

Groups	SOD (U/g tissue)	CAT (U/g tissue)	GSH (mmol/g tissue)	MDA (nmol/g tissue)
Ctrl.	45.18 ± 3.61^{c}	71.90 ± 3.17^{b}	$3.95 \pm 0.49^{a, b, c}$	40.07 ± 3.28^{d}
PB-alone	21.05 ± 2.33^{e}	32.26 ± 3.50^{e}	1.09 ± 0.92^{d}	102.37 ± 3.45^{a}
PB/CB	36.91 ± 2.13^{d}	53.89 ± 4.00^{c}	$2.02 \pm 0.90^{c, d}$	65.99 ± 5.95^{c}
PB/Vit-C	32.57 ± 3.65^{d}	45.45 ± 4.59^{d}	1.75 ± 0.82^{d}	82.76 ± 4.39^{b}
PB/CB/Vit-C	39.59 ± 2.05^{d}	63.14 ± 3.51^{c}	$2.50 \pm 0.98^{b, c, d}$	57.11 ± 5.59^{c}

The values represented mean ± SE; SOD: Superoxide dismutase; CAT: Catalase; GSH: Reduced glutathione; MDA: Malondialdehyde; Ctrl: Control; PB: Phenobarbital; CB: Cranberry; Vit-C: Vitamin C. P value < 0.05 was considered to be statistically significant. Means that do not share a letter are significantly different.

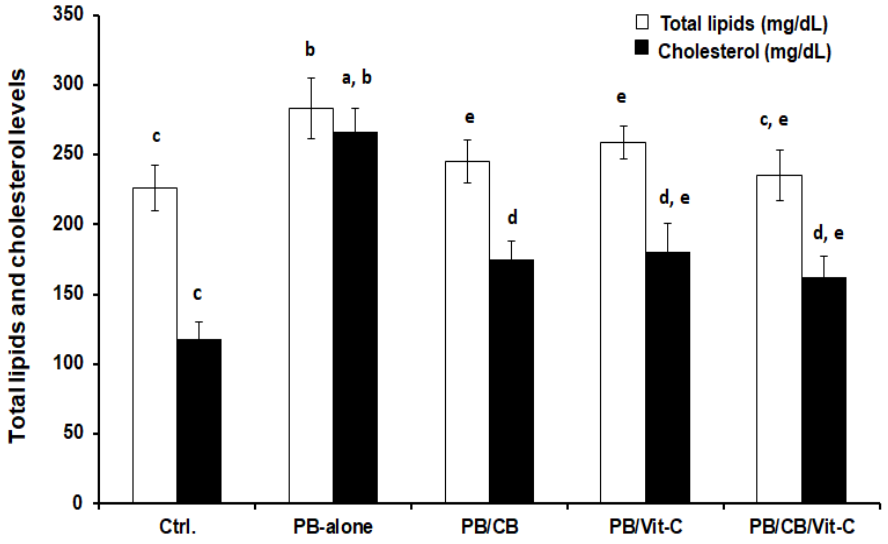

Fig. 5: Total lipids and cholesterol levels in the different groups under the study. The values represented mean ± SE; **Ctrl:** Control; **PB:** Phenobarbital; **CB:** Cranberry; **Vit-C:** Vitamin-C. P value < 0.05 was considered to be statistically significant.

Co-treatment with CB/Vit-C mitigated the histopathological changes in the hepatic tissues that induced with PB injections

Examination of control liver sections showed normal structure with no histopathological changes in the liver architectures**.** The liver sections of these rats revealed normal-like hepatic anatomy, a normal central vein, and a normal radiating hepatic strand, as well as normal blood sinusoids and normal phagocytic Kupffer cells (Figure 6 A). The examination of the liver sections of the PB-treated rats revealed loss of cellular organization, and hydropic deteriorated cells changed lobular shape, nuclear degradation in some spots, disarrangement of normal hepatic cells, necrosis, and severe fatty degeneration were all visible. Enlargement and congestion of the hepatic portal vein were discovered. Intrusion of lymphocytes was discovered in the portal area. Some hepatic cells showed degenerated alteration (pyknosis and karyolysis nuclei) and indicated a thick wall portal vein (Figure 6 B).

The liver sections of the PB/CB revealed mild fatty degeneration and moderate congestion following a phenobarbital injection. No signs of necrosis were found. With normal-appearing hepatic anatomy, a normal central vein, and a normal radiating hepatic strand, along with normal blood sinusoids and phagocytic Kupffer cells, certain hepatic cells displayed a minor degree of degeneration (nuclear degradation and pyknosis nucleus) (Figure 6 C). The liver sections of the treated rats with Vit-C after PB- injection showed moderate vein congestion, and no fatty deterioration was seen. Blood sinusoids appeared normal. Some hepatic cells showed a mild degree of the degenerated hepatocyte. No pyknotic nuclei were seen (Figure 6 D). Furthermore, the assessment of the liver sections of the rats that were treated with CB/Vit-C after PB injection showed adequate improvement, and the hepatic cords were properly positioned around the sinusoids. Hepatocytes and portal components appeared to be in acceptable condition. No Congestion and pyknotic nuclei were seen. There are few fatty degenerations. No indication of lymphocyte infiltration was seen (Figure 6 E).

Co-treatment with CB/Vit-C ameliorates the histopathological changes in the renal tissues that induced with PB injections

The renal parenchyma of control kidney sections had normal Bowman's capsules, glomerular capillaries, renal (Malpighian) corpuscles, and urine space were all concentrated in the renal cortex. At the base of the tubule, proximal convoluted tubules have a narrow lumen encircled by cubical cells with spherical nuclei. Large, simple cubical cells with sphere nuclei in the middle or at the apex of the tubule walls bordered the distal convoluted tubules' lumen (Figure 7 A). The histological changes in the kidney of PB-injected rats appeared in the form of some vacuolated glomeruli, congestion, fractured and shrunken size inside Bowman's capsules and irregular capsular space volume. Cell nuclei exhibited complete or partial damage and degenerated renal tubules in the proximal and distal convoluted tubules.

Additionally, the nucleus has been observed to compact (pyknotic nuclei). There lymphatic inflammatory cells infiltration, high degree of congestion, and cellular remains were found (Figure 7 B).

The renal sections of the PB/CB revealed some tubular epithelial cells in renal convoluted tubules exhibited mild vacuolation and degenerative changes. The glomerulus is of normal size within Bowman's capsules, with a normal volume of capsular space. Other glomeruli were fragmented, vacuolated, and swollen by mild rate. Few pyknotic nuclei are seen in only a few tubules. There was no evidence of lymphatic invasion. There was no area of hemorrhage. Congestion was noted to be moderate. No luminal cast was seen (Figure 7 C). The kidney sections of the treated rats with Vit-C after PB-injection showed tubular epithelial cells in renal convoluted tubules exhibited moderate vacuolation and degenerative changes. The glomerulus is of normal size within Bowman's capsules, with a normal volume of capsular space. Numerous glomeruli were atrophied. Few pyknotic nuclei are seen in only a few tubules. Degeneration of the tubular hydropic membrane. There was no evidence of lymphatic invasion. There was no area of hemorrhage. Congestion was noted to be mild (Figure 7 D). The examination of the kidney sections of the group of rats that was

treated with CB/Vit-C after PB-injection showed normal structure of glomeruli with normal size inside Bowman's capsule. Congestion was noted to be moderate, and pyknotic nuclei were visible. No luminal cast was seen. There was no evidence of lymphatic invasion (Figure 7 E).

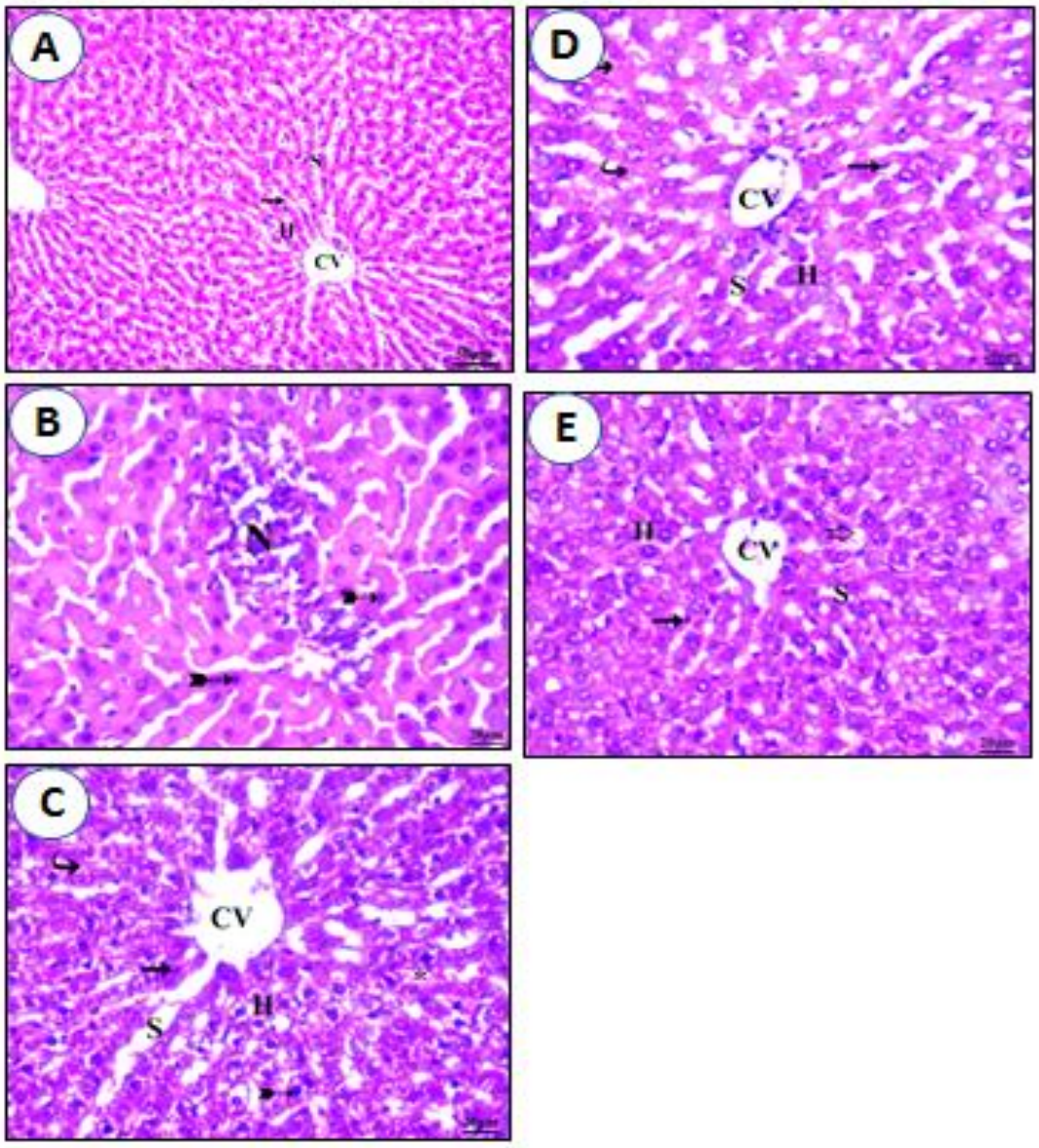

Fig. 6: Photomicrographs of a section in the liver of control rat **(A),** showing the normal histological structure of hepatic lope, central vein (CV) and radiating polygonal hepatocyte (H), blood sinusoids (S), Kupffer cells (arrow). PB **(B),** necrosis (N), pyknotic cell (Bifid arrow). CB after PB administration **(C),** showing maker improvement hepatic structure, normal radiating hepatocytes from the central vein (CV), hepatocytes (H), blood sinusoid (S), some hepatic cells still showing degenerative changes (curves arrow). Vit-C after PB administration **(D),** the hepatic cords was properly positioned around the sinusoids (S) and normal central vein (CV), Hepatocytes (H), degenerated hepatocyte (curved arrow), Kupffer cell (arrow). CB /Vit-C after PB-administration **(E),** showing maker improvement hepatic structure, hepatocytes (H) appeared to be in good condition, central vein (CV), Kupffer cell (arrow). Few fatty degeneration (hollow arrow), blood sinusoid (S) were noticed (H&E stain).

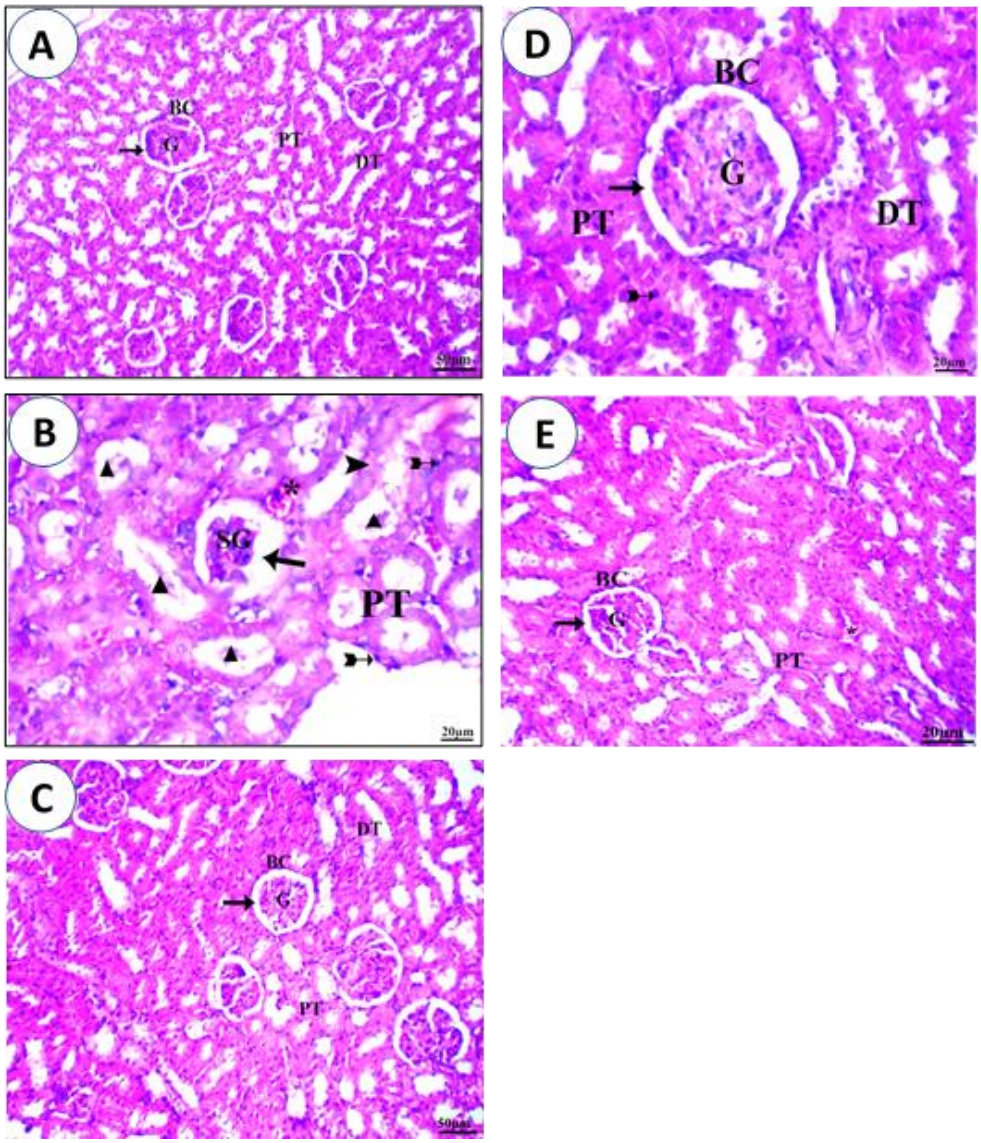

Fig. 7: Photomicrographs of a section in the kidney of control rat **(A),** were showing the normal histological structure of the cortex region, normal glomeruli (G), normal Bowman's capsule (BC), normal proximal tubule (PT), capsular space (arrow), normal structure of distal tubule (DT). PB- induced rat **(B)** showing renal congestion (*), shrunk glomeruli (SG) were visible, luminal cast (Triangle) and pyknotic nuclei (bifid arrow) detectable, (H&E stain). Photomicrographs of section of kidney of rat protected with CB after PB- injection **(C),** Vit-C after PB- injection **(D),** CB/Vit-C after PB- injection **(E),** showing maker improvement in renal structure, normal glomeruli (G), normal Bowman's capsule (BC), capsular space (arrow), normal proximal tubule (PT), distal tubules (DC) and few congestion (*) was noticed (H&E stain).

Discussion

Phenobarbital (PB) is the drug of choice for long-term treatment of epilepsy but it exerts some undesirable side effects[5, 7]. This study aimed to investigate whether CB or/and Vit-C have ameliorative effects against the hepato-renal toxicity that induced by PB-treatment. According to this study, PB (160 mg/kg) administration for one month caused a significant decrease the body weight. A previous study revealed that PB administration induced body weights loss significantly[45]. The treatment with CB or Vit-C or with their combination after PB-injection increased the body weight gain. This could explain the therapeutic effects of the CB and Vit-C due to their potential antioxidant properties[31, 28].

In this study, it was found that the PB treatment led to hepatotoxicity evidenced by significant decrease in the albumin, and TP

levels along with significant increase the ALT, AST activities. Consistent with this finding, a previous study reported that PB is triggering proliferation of the hepatocyte smooth endoplasmic reticulum, boosted liver weight and serum liver enzyme activities[46, 5, 7]. Treatment with CB or/and Vit-C after PB-injection led to significant reduction in the liver transaminases activity, total protein and albumin levels. This result was reliable with prior studies that have stated that both of CB and Vit-C have a protective effect against hepatoxicity[32, 47]. The treatment with PB led to increasing serum urea, and creatinine levels and this could explain the renal damage. This result was consistent with a preceding research which reported that PB caused nephrotoxicity in rats and elevated levels of urea nitrogen, uric acid, and creatinine[6]. Treatment with CB or/and Vit-C after PB-injection, however, led to significant diminution in the levels of creatinine and urea. This result was reliable with earlier studies that have informed that both of CB and Vit-C have a protective effect against the renal damage that induced to toxic agents[48, 28].

Glutathione (GSH) and superoxide dismutase (SOD) are known as reactive oxygen species (ROS) scavenging agents. Although, ROS have a significant part in several cellular signaling processes at low levels, however, they employ a cytotoxic impact by destruction macromolecules such as proteins, lipids and nucleic acids. Therefore, the cellular antioxidants enzymes, CAT and SOD detoxify the ROS and the subsequent oxidative stress. This study reported that the PB-caused hepatotoxicity was correlated with oxidative stress triggered by the decline in the antioxidant enzymes (SOD and CAT) indicating pronounced oxidative stress. This finding was similar to previous study which showed that the treatment with PB increase MDA level associated with significant decreases in CAT and SOD levels in treated rats[12].

The increase in hepatic lipid peroxidation was associated with initiation of cytochrome P_{450} and cytochrome c reductase after the treatment with PB[49]. Previous report suggested that natural antioxidants protect against PB-hepatotoxicity[50]. Treatment with CB post PB-injection rise the levels of CAT and SOD that indicated the improvement of antioxidant status of the livers, these observations confirm the finding of studies, which stated the improvement of antioxidant enzyme activities upon administration of CB or Vit-C to experimental animals[29]. Treatment with CB or/and Vit-C after PB injection provided protection against lipid peroxidation, which could be attributed to the free radical removing ability. These findings were reliable with the prior research paper shown the role of CB and Vit-C in ameliorating the lipids peroxidation induced by toxic agents in experimental animals[7, 27, 30].

The levels of the total lipid and total cholesterol were increased significantly in the group of rats that were treated with PB. Previous study showed that long term use of PB causes an increase in plasma total cholesterol levels in rodents and humans[10].

Treatment of PB-injected rats with CB or Vit-C led to decrease the levels of the total lipid and cholesterol, this could be due to the potential effect of CB and Vit-C to reduce the lipid content in PB-treated rats[29]. Earlier studies have reported that PB-treatment could cause liver and kidney toxicity[7]. PB is possibly triggers sinusoidal obstruction syndrome, causing a directly toxic impact on hepatic sinusoidal cells, thus stimulating necrosis, obstruction, and obliteration of hepatic veins. The treatment with CB or/and Vit-C ameliorated the hepatic and renal dysfunctions and decreases the hepatic and renal histological changes which induced by PB-injection. The results showed that in PB-treated rats, the liver tissue loss the cellular organization, with a marked histological alteration. This finding agreed with earlier research by[5], who informed that the PB-treatment led to histopathological changes in the liver tissue. Treatment with CB or/and Vit-C ameliorated these changes that induced with PB-treatment. This finding could be due to the potential anti-oxidant properties of CB and Vit-C [32, 7, 27].

The histological changes in the kidney of PB-injected rats appeared in the form of some vacuolated glomeruli, congestion, fractured and shrunken size inside Bowman's capsules and irregular capsular space volume. These results could explain the direct toxic effect of PB on the renal tissues. This result was in agreement with previous study showed that PB caused

nephrotoxicity in rats[6]. The treatment with CB, Vit-C or their combination led to an improvement in the kidney structure which postulated that both of them have potent therapeutic effect against the renal toxicities that induced with PB-treatment. In conclusion, this study showed the potential ameliorative effects of CB, Vit-C and their combination against liver and kidney toxicities that were induced by phenobarbital.

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العلاج بكل من التوت البري و فيتامين سي يقلل من السمية الكبدية والكلوية الناتجة عن حقن الفينوباربتال في الجرذان المعملية

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على الرغم من أن عقار الفينوباربيتال يستخدم لعلاج مرضى الصرع، إلا أن أستخدامه علي المدى الطويل يؤدى إلى أختلال فى كل من وظائف الكبد والكلى. وتعتبر المنتجات الطبيعية من العناصر الأساسية التى تستخدم لحماية الكبد والكلي من الاثار الضارة نتيجة لخصائصها المضادة للأكسدة. هدفت هذه الدراسة إلى معرفة الدور العلاجى لكل من التوت البري و فيتامين سي او كلاهما ضد السمية الكبدية والكلوية الناتجة عن استخدام عقار الفينوباربيتال في الجرذان البيضاء. وقد تم تقسيم عدد أربعون من ذكور الجرذان إلى خمس مجموعات متساوية كل مجموعة تحتوى علي ثمانية جرذان كالتالي: المجموعة الأولى : المجموعة الضابطة تم اعطائها مياه مقطره عن طريق الفم لمدة ٦٠ يوم يوميا. المجموعة الثانية: تم اعطائها عن طريق الفم عقار الفينوباربيتال (١٦٠ ملجم / كجم) لمدة ٣٠يوم يوميا. المجموعة الثالثة : تم اعطائها التوت البرى (٥٠٠ ملجم / كجم) لمدة ٣٠ يوم يوميا بعد حقنها بعقار الفينوباربتال لمدة ٣٠ يوما. المجموعة الرابعة: تم اعطائها عن طريق الفم من فيتامين سي (٢٧ ملجم / كجم) لمدة ٣٠ يوم يوميا بعد حقنها بعقارالفينوباربتال لمدة ٣٠ يوما. المجموعة الخامسة : تم اعطائها عن طريق الفم كل من التوت البرى مع فيتامين سي لمده٣٠ يوم يوميا بعد حقنها بعقار الفينوباربتال لمده ٣٠ يوما. وقد تم تجميع العينات من كل المجموعات لقياس وظائف الكبد ALT ، AST ، البروتين الكلي ، الألبومين ، وظائف الكلي اليوريا ، الكرياتينين ، وكذلك المؤشرات الحيوية المضادة للأكسدة/المؤكسدة ؛ SOD، CAT، MDA، .GSHوكذلك تم معرفة التغيرات النسيجية للكبد والكلى. اظهرت النتائج أنه العلاج بعقار الفينوباربيتال يقلل من النسبة المئوية في وزن الجسم للجرذان ، وخلل فى وظائف كل من الكبد والكلى، وزيادة ملحوظة فى مستويات الدهون الكلية، والكوليسترول، ومستويات MDA، وقد ادي انخفاض مستويات SOD، CAT، GSHفي الأنسجة وتغيير البنى النسيجية لكل من الكبد والكلى. كما أوضحت النتائج أن العلاج باستخدام التوت البرى و فيتامين سي قد ادى الى تقليل الآثار الضارة التي أحدثها العلاج بعقار الفينوباربتال، كما أظهرت النتائج ان الجرذان المحقونة بعقار الفينوباربتال والتي تمت معالجتها بكل من التوت البرى و فيتامين سي قد اظهرت أعلى الآثار الوقائية.